Risk factors for urinary bladder cancer

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**Risk Factors
for
Urinary Bladder Cancer**

Maurice P.A. Zeegers

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voor mijn moeder en Jos

1 **Introduction**

INTRODUCTION

Bladder cancer is the seventh most frequent tumour in men worldwide, accounting for approximately 200,000 men with newly diagnosed bladder cancer each year. Bladder cancer is considerably more common in men than in women, but still yearly approximately 60,000 women are diagnosed with the disease worldwide.¹ In The Netherlands, bladder cancer is the fifth most frequent invasive tumour in men. In this country, 1,653 men and 532 women were diagnosed with bladder cancer in 1996.² The age-standardised (European Standard Population) incidence rates for bladder tumours for men and women were 22.2 and 5.0 per 100,000 person-years in this population in 1996, respectively. Like many solid tumours, bladder cancer incidence increases with age. Tumours of the bladder rarely occur before the age of 40 to 50, but most commonly arise in the seventh decade of life.² The incidence of bladder cancer varies considerably between countries.^{1,3} In general, the highest incidence rates for bladder cancer are found in developed countries like Western Europe and North America; relatively low rates are found in Eastern Europe and several areas of Asia.^{1,3} In The Netherlands in 1996 750 men with bladder cancer (61%) and 325 women with bladder cancer (45%) died from the disease.²

About 90% of bladder cancers are transitional cell carcinomas, cancers that begin in the urothelial cell lining. The renal pelvis, ureters, and urethra are covered with the same urothelium. Therefore, unless otherwise specified, the term bladder cancer is used as a synonym for these neoplasms. About 80% of bladder tumours are confined to the bladder mucosa, the so-called superficial tumours, and 20% invade the muscle layer.⁴ Haematuria, pain during urination and frequent urination are the most common symptoms of both types of cancer. However, the management and prognosis of these cancers are completely different: superficial tumours are fairly benign, and invasive tumours are highly malignant. Depending on its stage and grade, bladder cancer may be treated with surgery, radiation therapy, chemotherapy, or immunotherapy.

Along with lung cancer, bladder cancer was one of the earliest cancers to undergo serious epidemiologic investigation. As a consequence, some important causes of bladder cancer such as cigarette smoking, occupational exposure, and chronic infections with *schistosoma haematobium* have been known for decades.⁵ However, at present there is few detailed information for specific smoking characteristics or specific occupational carcinogenic agents other than aromatic amines available. The recent controversy concerning the influence of fluid consumption and diet on the risk of bladder cancer points to some of the still unanswered questions about the aetiology of the disease. Therefore, research is warranted to study bladder cancer aetiology, in which possible risk factors for bladder cancer are explicitly tested. Knowledge about these determinants may play an important role in bladder cancer prevention.

In this dissertation, analyses regarding the associations between several environmental risk factors (i.e., tobacco smoking, fluid consumption, diet and occupation) and bladder cancer incidence are reported. These analyses were carried out through meta-analyses of epidemiologic studies and through specific analyses within the context of the Netherlands Cohort Study on diet and cancer.

META-ANALYSES OF EPIDEMIOLOGIC STUDIES

Although meta-analyses are traditionally used to combine evidence from experimental research we applied this technique to observational epidemiologic studies.^{6,7} In meta-analyses all previous epidemiologic literature concerning cigarette smoking, fluid consumption and bladder cancer incidence was examined and evaluated systematically in order to identify potential risk factors and to summarize their potential effects on bladder cancer incidence. Furthermore, the potentially profitable areas of further investigation were identified.

The epidemiologic literature was identified through computerized database searches for follow-up and case-control studies published from 1966 to 2000 and through reference

checking.⁷ From each study both qualitative and quantitative data were abstracted. A criteria list was developed for the assessment of quality items in observational cancer research.⁸ A statistical method was developed to abstract the quantitative data from the component studies.⁹ In the meta-analyses, summary effects were estimated, both adjusted and unadjusted for potential confounders with random effects meta regression analysis.⁶ Meta-analyses concerning diet and occupation were not addressed in this thesis either because a meta-analysis of the recent dietary literature already exists^{10,11} or because it was not possible to conduct since too much heterogeneity existed between the studies. The reader is referred to other publications instead.^{12,13}

THE NETHERLANDS COHORT STUDY

In the meta-analyses, no data were available to explore dose-response associations or to conduct detailed analyses on smoking or drinking characteristics. These results prompted us to investigate the associations between smoking, fluid consumption, diet, occupation and bladder cancer incidence in more detail in the ongoing Netherlands Cohort Study.

In 1986, a large-scale prospective cohort study, the Netherlands Cohort Study on diet and cancer,¹⁴ was initiated among 120,852 men and women aged 55-69 years. The subjects originated from 204 municipal registries throughout the country. Data regarding potential risk factors for cancer were collected by means of a self-administered questionnaire. Follow-up for incident bladder cancer was established by record linkage to all regional cancer registries in The Netherlands and with the national pathology register (PALGA). The case cohort approach was used for data processing and analysis. Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after the baseline exposure measurement. 619 Incident bladder cancer cases were detected during a follow-up period of 6.3 years (September 1986 - December 1992).¹⁵

OUTLINE OF THE THESIS

Chapter 2 describes a statistical method, which was used to abstract quantitative data from the component studies in the meta-analyses. In the next three chapters the epidemiologic literature on smoking (Chapter 3), alcohol consumption (Chapter 4), coffee and tea consumption (Chapter 5) and bladder cancer incidence are systematically reviewed in terms of meta-analyses. Thereafter, our findings from the Netherlands Cohort Study regarding possible associations between the incidence of bladder cancer and active and environmental tobacco smoking (Chapter 6), alcohol consumption (Chapter 7), coffee, tea and total fluid consumption (Chapter 8), consumption of vegetables and fruits (Chapter 9), retinol, vitamin C, vitamin E, folate and carotenoids intake (Chapter 10), prediagnostic toenail selenium (Chapter 11), and occupational exposure to carcinogens (Chapter 12) are presented. Finally, the findings described in this thesis are discussed in the epilogue (Chapter 13).

REFERENCES

1. Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer*. 1999;80:827-41.
2. Visser O, Coebergh JWW, Schouten LJ, van Dijck JAAM. Incidence of cancer in the Netherlands:1996. Utrecht: Association of comprehensive cancer centres, 2000.
3. Schottenfeld D, Fraumeni JF. *cancer epidemiology and prevention*. New York Oxford: Oxford university press, 1996.
4. van der Meijden APM. Bladder cancer. *Br Med J*. 1998;317:1366-9.
5. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol*. 1996;23:536-45.

6. Zeegers MPA, Heisterkamp SH, Kostense PJ, van der Windt DAWM, Scholten RJPM. De praktijk van systematische reviews: VIII Het combineren van de resultaten van observationele onderzoeken. *Ned Tijdschr Geneeskd*. 2000;144:1393-7.
7. van der Windt DAWM, Zeegers MPA, Kemper HGC, Scholten RJPM, Assendelft WJJ. De praktijk van systematische reviews. VII Zoeken, selecteren en methodologische beoordeling van observationeel onderzoek. *Ned Tijdschr Geneeskd*. 2000;144:1210-4.
8. Zeegers MP, Tan FE, Dorant E, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer*. 2000;89:630-9.
9. Tan FES, Zeegers MPA. An alternative method of combining exposure specific odds ratios in epidemiologic studies. Research report. Maastricht: Maastricht University, Department Methodology and Statistics, 1999.
10. Steinmaus CM, Nunez S, Smith AH. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol*. 2000;151:693-702.
11. Boffetta P, Silverman DT. A meta-analysis of bladder cancer and diesel exhaust exposure. *Epidemiology*. 2001;12:125-30.
12. Vineis P, Simonato L. Proportion of lung and bladder cancers in males resulting from occupation: a systematic approach. *Arch Environ Health*. 1991;46:6-15.
13. t Mannetje A, Kogevinas M, Chang Claude J, Cordier S, Gonzalez CA, Hours M, Jockel KH, Bolm Audorff U, Lynge E, Porru S, Donato F, Ranft U, Serra C, Tzonou A, Vineis P, Wahrendorf J, Boffetta P. Occupation and bladder cancer in European women. *Cancer Causes Control*. 1999;10:209-17.
14. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol*. 1990;43:285-95.
15. Barlow WE, Ichikawa L, Rosner D, Izumi S. Analysis of case-cohort designs. *J Clin Epidemiol*. 1999;52:1165-72.

2

An Unbiased Estimator of Exposed versus Non-Exposed Odds Ratio from Summarised Dose-Response Data

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ABSTRACT

Summary effect measures in meta-analysis of published epidemiological studies are often based on (adjusted) odds ratios reported for several exposure levels with varying arrangements and number of levels across studies. Usually only two way contingency tables together with exposure specific adjusted odds ratios and corresponding standard errors are presented in articles. An unbiased estimate of exposed versus non-exposed adjusted odds ratio from summarised dose-response data is proposed. This estimate is based on the weighted sum of the exposure specific odds ratios, with the prevalences of the control group as weights. Large sample variance is derived accounting for the dependency between exposure specific adjusted odds ratios. A summary adjusted odds ratio between studies is then obtained by using standard methods common in stratified analysis.

INTRODUCTION

In the last decade there is a rapidly growing interest to systematically review and summarize results from several homogeneous studies¹. Important arguments in favor of meta-analysis are to investigate potential heterogeneity between different studies, to generalise results of single studies and the possibility to quantify a summary risk estimate from several published data. Furthermore, many relevant but statistically non significant results pointing in the same direction can be pooled into one summary measure with an increase in statistical power. An example of such conclusion can be found in a recent paper of Zeegers et. al.² They have performed a random effects meta-analysis to estimate a summary association between current alcohol consumption and cancer of the urinary tract. In order to perform that analysis, odds ratios, unadjusted and adjusted for confounders, were extracted directly from the primary studies. However, those primary studies present separate adjusted odds ratios with study specific arrangements and number of levels of the determinant.

A major problem that we often encounter is the lack of agreement regarding how criterion variables should be operationalised. For example, the increase risk due to alcohol consumption in the articles Zeegers et al.² have reviewed is based on different exposure comparisons. Some authors compared non-users with current users. Others reported comparisons between non-users, those who drank only one glass and those who drank two or more glasses within a given time unit. In most cases, two way contingency tables, exposure specific adjusted odds ratios and corresponding standard errors or confidence bounds are the only summary table and statistics presented in the results section of the primary studies. The lack of consistency across studies makes it difficult to pool study specific estimates, because they are not directly comparable to each other due to at least different exposure comparisons. The problem that we are dealing with in this article, is how to combine odds ratios or adjusted (for confounding) odds ratios measuring different exposure comparisons into a single odds ratio. Several pooling methods exist in literature. Greenland and Longnecker³ proposed to use regression methods and to use the weighted average of all within study regression slopes as a summary estimate. This method has been applied by others.^{4,5} In some situations, however, the dose-response method is not recommended. Regarding the study of Zeegers et al.,² for example, in four primary studies (25 %) the determinant has only two levels. Twelve primary studies (75 %) consists of less than four drinking levels (the last level is an open category). As Berlin, Longnecker and Greenland⁶ have pointed out, the estimates of the slopes are highly sensitive to the choice of method for assigning values to the exposure levels; especially if small numbers of exposure levels and open categories are involved.

Another way to make effect estimates comparable across studies is to only compare exposed with non-exposed. Thus by combining the different exposure levels within each study into one large user level which is to be compared with the reference level. The corresponding (adjusted) odds ratio for each study is derived from the available exposure specific odds ratios, standard errors and information from the collapsed two way contingency table. Boyd, Martin, Noffel, Lockwood and Tritchler,⁷ proposed to average the effect estimates across the exposure levels. Others have proposed to calculate an estimate 'at any degree' based on weighting the exposure specific effect estimates. Some⁸ used the reciprocal of the variance as a weight for each estimate.

In this article we propose an alternative method that estimates an exposed versus non-exposed odds ratio from summarised dose-response data. In section two the estimator of exposed versus non-exposed is defined and the properties of it are discussed. In section three a large sample variance of the estimator is derived accounting for dependency between exposure specific adjusted odds ratios. The method is illustrated by means of both hypothetical and real data in section four. Finally, we end up with some discussion in section five.

AN UNBIASED ESTIMATOR OF EXPOSED VERSUS NON-EXPOSED ODDS RATIO BASED ON REPORTED RESULTS

Many studies, dealing with the relationship between some determinant and an outcome and controlling for several confounders, only report exposure specific (adjusted) odds ratios, their standard errors (or confidence intervals), and a two way contingency table, cross-classifying the determinant and outcome variables. To be more specific, we first consider the complete source, based on which the results are presented in the results section of the primary study. Then, we will point out which information is often not presented. Table 1 shows in general form the cross-classified table that contains all necessary information.

Table 1. Full information cross-classified table with confounders

Confounder (i)	Determinant (j)	Dependent		Row total
		Case (code 1)	Non-case (code 0)	
1	0	X ₁₀₁	X ₁₀₀	n ₁₀
	1	X ₁₁₁	X ₁₁₀	n ₁₁
	.	.	.	.
	K	X _{1K1}	X _{1K0}	n _{1K}
2	0	X ₂₀₁	X ₂₀₀	n ₂₀
	1	X ₂₁₁	X ₂₁₀	n ₂₁
	.	.	.	.
	K	X _{2K1}	X _{2K0}	n _{2K}
.	.	.	.	.
.	.	.	.	.
.	.	.	.	.
L	0	X _{L01}	X _{L00}	n _{L0}
	1	X _{L11}	X _{L10}	n _{L1}
	.	.	.	.
	K	X _{LK1}	X _{LK0}	n _{LK}

The first column denotes all stratum combinations of one or more confounders. Let $\{X_{ij1}\}_{i=1,\dots,L}, j=0,\dots,K$ be independent binomials with $X_{ij1} \sim \text{Bin}(n_{ij}, p_{ij})$, where $n_{ij} = X_{ij0} + X_{ij1}$ is the number of subjects from stratum i and exposure category j , X_{ij0} is the number of non-cases, and X_{ij1} is the number of cases from stratum i and exposure category j . The risk p_{ij} is estimated by $\hat{p}_{ij} = \frac{X_{ij1}}{n_{ij}}$. Furthermore, let $n_0 = \sum_{i=1}^L n_{i0}$ be the number of subjects in the non-exposed group with corresponding risk $p_0 = \frac{1}{n_0} \sum_{i=1}^L p_{i0} n_{i0}$. Denote the odds ratio from stratum i and exposure level j as $\hat{\alpha}_{ij} = \frac{\hat{p}_{ij}}{1 - \hat{p}_{ij}} \cdot \frac{1 - \hat{p}_{i0}}{\hat{p}_{i0}}, j = 1, \dots, K$. Most papers do not present full information like table 1. For ease of presentation, this table is often collapsed into a two way contingency table. Thus, the crude number of cases, say $C_j = \sum_{i=1}^L X_{ij1}$ and non cases, say $NC_j = \sum_{i=1}^L X_{ij0}, j = 0, \dots, K$ are only presented in the primary study. Denote the estimated crude odds ratio of exposure category j as $\hat{\alpha}_j = \frac{NC_0 * C_j}{NC_j * C_0}, j = 1, \dots, K$. Often the estimated adjusted odds ratios $\hat{\alpha}_j^{adj}$ of all exposure categories $j = 1, \dots, K$ and their corresponding standard errors are also presented. Based on the reported results, the problem that we tackle in this paper is how to estimate the adjusted odds ratio of only the exposed versus the non-exposed group. Before we go into that in detail, consider first the unadjusted situation. Let $\hat{\alpha}_{exposed}$ be the crude estimated odds ratio of exposed versus non-exposed,

i.e., $\hat{a}_{exposed} = \frac{NC_0 * \sum_{j=1}^K C_j}{C_0 * \sum_{j=1}^K NC_j}$. It appears, that $\hat{a}_{exposed}$ can be represented in terms of the exposure specific estimated crude odds ratios $\hat{a}_j$ as follows (see appendix A1),

$$\hat{a}_{exposed} = \frac{\sum_{j=1}^K NC_j * \hat{a}_j}{\sum_{j=1}^K NC_j}. \quad (1)$$

Equation (1) shows that the odds ratio of the reduced (into only exposed versus non-exposed) table can be expressed as a weighted average of the available exposed specific estimated odds ratios. The weight appears to be just the non-cases of the exposure levels. If the number of non-cases is constant across exposure levels and not necessarily equal to NC_0 , then $\hat{a}_{exposed}$ of the reduced table is equal to the average of the exposure specific $\hat{a}_j$. In many research situations, however, the distribution of the non-cases across exposure levels is skewed and the $\hat{a}_j$'s differ from each other.

Two problems arise, if we try to extend the above results to adjusted versions of the odds ratios. The first problem deals with confounding aspects. In view of the (meta-analysis) application that we have in mind, the following assumption seems reasonable. Let $\emptyset$ be any rearrangement of exposure level categories. We assume that the expected stratum (with respect to the confounders) specific odds ratios are all equal, i.e. $E(\hat{a}_{ij}) = E(\hat{a}_j^{adj})$. For all strata $i = 1, \dots, L$ and exposure levels $j \in \emptyset$. This assumption can be motivated as follows. Suppose that a meta-analysis is performed based on a (large) number of papers dealing with the same research design. Suppose further, that each paper presents adjusted odds ratios controlling for, in essence, the same set of covariates. In general, any rearrangement of the exposure levels, may introduce (but not necessarily) an interaction between the determinant and the covariates. The fact that, for these studies, exposure specific odds ratios are presented for different arrangement and number of exposure levels, suggests that the covariates mentioned in those papers remain confounders irrespective of how the determinant is classified. Let $\hat{a}_{exposed}^{adj}$ be any estimator of the adjusted odds ratio, comparing only exposed with non-exposed group (reduced table), satisfying $E(\hat{a}_{i exposed}) = E(\hat{a}_{exposed}^{adj})$ for all $i = 1, \dots, L$, where $\hat{a}_{i exposed}$ is an estimator of the stratum specific odds ratio in the reduced table. It can be shown (appendix A2), that the estimator of the adjusted odds ratio in the reduced table must satisfy

$$E(\hat{a}_{exposed}^{adj}) = \frac{\sum_{j \in \emptyset} NC_j * E(\hat{a}_j^{adj})}{\sum_{j \in \emptyset} NC_j}. \quad (2)$$

Note that, if $\hat{a}_j^{adj}$ and $\hat{a}_{i exposed}$ are unbiased estimates of the exposure specific and stratum specific (adjusted) odds ratios, respectively, then the estimator defined as

$$\hat{a}_{exposed}^{adj} = \frac{\sum_{j \in \emptyset} NC_j * \hat{a}_j^{adj}}{\sum_{j \in \emptyset} NC_j} \quad (3)$$

is also an unbiased estimate of the adjusted odds ratio in the reduced table. This follows immediately from the definition of confounding, i.e. $E(\hat{a}_{ij}) = E(\hat{a}_j^{adj})$, and equality (1) for each stratum i . It follows in particular that $E(\hat{a}_{exposed}^{adj}) = E(\hat{a}_{i exposed})$ for all $i = 1, \dots, \emptyset$. In many articles, marginal two way contingency tables are reported as well as exposure specific odds ratios adjusted for several confounders. Thus in particular, NC_j and $\hat{a}_j^{adj}$ for all j are known. Hence, adjusted odds ratios for the reduced situation can be estimated without bias

from the reported results of the primary study. In many cases, standard errors of the (logarithm of the) exposure specific adjusted odds ratios are also reported. In that case it is also possible to estimate the variance of $\hat{\alpha}_{exposed}^{adj}$ as will be shown in the next section.

VARIANCE OF THE ESTIMATED ADJUSTED ODDS RATIO OF EXPOSED VERSUS NON-EXPOSED

A problem that is also mentioned by Greenland and Longnecker³, is the fact that in practice the odds ratios $\hat{\alpha}_j^{adj}$'s are heavily correlated. Many authors use in their primary study an adjustment method proposed by Woolf.⁹ Basically the adjustments are made by calculating the weighted average of the stratum specific odds ratios, which is of the form

$$\ln(\hat{\alpha}_j^{adj}) = \sum_{i=1}^L \omega_{ij} \ln \hat{\alpha}_{ij}, \quad (4)$$

where $\sum_{i=1}^L \omega_{ij} = 1$. Here, we suppose for ease of presentation that there are K exposure levels numbered as $j = 1, \dots, K$. It can be shown that (appendix A3), asymptotically the

$$\text{cov}(\hat{\alpha}_m^{adj}, \hat{\alpha}_n^{adj}) = \alpha_m^{adj} \alpha_n^{adj} \sum_{i=1}^L \frac{\omega_{im} \omega_{in}}{n_{i0} p_{i0} (1 - p_{i0})}, \quad (5)$$

following equation holds.

for all $m, n = 1, \dots, K$. It should not be difficult for authors of primary studies to calculate the covariances according to equation (5). Unfortunately, most papers do not report these values. The quantities ω_{im} , ω_{in} and p_{i0} are not reported in most cases, so that the covariances can not be calculated from the results section of the primary studies according to equation (5). Equation (5) reduces to

$$\text{cov}(\hat{\alpha}_m^{adj}, \hat{\alpha}_n^{adj}) = \frac{\alpha_m^{adj} \alpha_n^{adj}}{n_0 p_0 (1 - p_0)}, \quad (6)$$

if the crude odds ratio is approximately equal to the adjusted odds ratio. Note that, this condition is the same as the first assumption mentioned by Greenland and Longnecker.³ Numerical calculations, however, show that equation (6) is a good approximation of equation (5). It is now straightforward that the asymptotic variance of $\hat{\alpha}_{exposed}^{adj}$ is equal to

$$\begin{aligned} \text{Var}(\hat{\alpha}_{exposed}^{adj}) = & \\ \frac{1}{(\sum_{j=1}^K NC_j)^2} & (\sum_{j=1}^K NC_j^2 \text{var}(\hat{\alpha}_j^{adj}) + \frac{2}{n_0 p_0 (1 - p_0)} \sum_{m < n} NC_m NC_n \alpha_m^{adj} \alpha_n^{adj}). \end{aligned} \quad (7)$$

In the literature the variance of log transform of the odds ratio is often calculated. The variance of the odds ratio can be approximated by

$$\text{var}(\hat{\alpha}_j^{adj}) = \text{var}(\ln(\hat{\alpha}_j^{adj})) * \alpha_j^2. \quad (8)$$

It should be noted, that the covariance between two arbitrarily chosen exposure specific estimated adjusted odds ratios depends on the number of non-exposed subjects and the probability of a case in the non-exposed group. This is not a surprise, because all covariation can be explained by the common reference group.

ILLUSTRATIONS

We have selected two published data that also reported three way contingency tables showing the relationship between determinant and bladder cancer for each stratum of a third factor.^{10,11} For each study, we have calculated the exposure specific (adjusted) odds ratios as well as the (adjusted) odds ratio of the reduced table. The first study deals with the risk of coffee consumption for males and females.¹⁰ Coffee consumption consists of three exposure levels. The second study deals with the risk of smoking for alcohol drinkers and abstainers.¹¹. Smoking consists of four exposure levels. Table 2 shows some input data that should be known for the authors of the primary study.

Table 2. Input data known to the author of the primary study

Confounder	determinant	case	Control	exp(osure) spec(ific) $\hat{\alpha}_{ij}$	exp. spec. var(ln($\hat{\alpha}_{ij}$))	adjusted exp.spec. $\hat{\alpha}_i^{adj}$	exp. spec. var(ln($\hat{\alpha}_i^{adj}$))
Gender				Pujolar et al.¹⁰			
Male	≤1	34	103	1.00 (reference)		1.00 (reference)	
	2-7	138	326	1.28	0.05	1.23	0.04
	8-14	130	294	1.34	0.05	1.31	0.04
	≥15	135	263	1.56	0.05	1.42	0.04
	≤1	5	10	1.00 (reference)			
Female	2-7	17	37	0.92	0.39		
	8-14	24	42	1.14	0.37		
	≥15	13	38	0.68	0.40		
Alcohol				Bravo et al.¹¹			
Drinker	0	28	122	1.00 (reference)		1.00 (reference)	
	<20	24	30	3.49	0.12	3.63	0.09
	21-30	50	40	5.45	0.09	5.35	0.07
	31-40	125	35	15.56	0.08	15.04	0.07
	>40	55	40	5.99	0.09	5.80	0.06
Non-drinker	0	13	47	1.00 (reference)			
	<20	8	7	4.13	0.37		
	21-30	14	10	5.06	0.27		
	31-40	6	2	10.85	0.76		
	>40	30	20	5.42	0.18		
Confounder		Determinant		Hypothetical data			
1	0	7	12	1.00 (reference)		1.00 (reference)	
	1	20	10	3.43	0.38	3.40	0.18
	2	140	30	8.00	0.27	8.00	0.14
	3	14	3	8.00	0.63	8.30	0.43
	≥4	14	21	1.14	0.35	1.20	0.20
2	0	10	18	1.00 (reference)			
	1	15	8	3.38	0.35		
	2	40	9	8.00	0.29		
	3	5	1	9.00	1.36		
	≥4	5	7	1.29	0.50		

The last example deals with hypothetical data. These hypothetical data are constructed to fulfil the assumption of invariance of confounding against any rearrangement of the exposure categories. Table 3 shows the results of the estimated (adjusted) odds ratios $\hat{\alpha}_{exposed}$ ($\hat{\alpha}_{exposed}^{adj}$) when using information only from the collapsed two way contingency table of disease versus determinant and the exposure specific (adjusted) odds ratios $\hat{\alpha}_j$ ($\hat{\alpha}_j^{adj}$) and the variance of the log transform (Table 2).

Table 3. Exposed versus non-exposed oddsratio from full data and summarised data

Study	Full data		Summarised data	
	Estimated oddsratio $\hat{\alpha}_{\text{exposed}}$	Estimated variance $\text{var}(\ln(\hat{\alpha}_{\text{exposed}}))$	Estimated oddsratio $\hat{\alpha}_{\text{exposed}}$	Estimated variance $\text{var}(\ln(\hat{\alpha}_{\text{exposed}}))$
Pujolar et al.¹⁰				
Adjusted	1.32	0.04	1.32	0.04
Unadjusted	1.32	0.04	1.32	0.04
Max. correlation $\hat{\alpha}_i, \hat{\alpha}_j$	0.78			
Bravo et al.¹¹				
Adjusted	6.92	0.04	7.10	0.04
Unadjusted	6.99	0.04	6.99	0.04
Max. correlation $\hat{\alpha}_i, \hat{\alpha}_j$	0.48			
Hypothetical data				
Adjusted	4.84	0.11	4.94	0.12
Unadjusted	5.02	0.11	5.02	0.12
Max. correlation $\hat{\alpha}_i, \hat{\alpha}_j$	0.58			

The quality of these estimates based on summarised data is related to the estimates based on full data information. From table 3 it can be concluded, that the estimates based on summarised data are close to the estimates if full data information were used. Note that, as it should be according to equation (1), the unadjusted estimates based on summarised data are exactly equal to the estimates based on full data information. The variances of the summary measures are calculated by using the approximation given in equation (7). As can be seen from table 3, these values are close to the 'correct' variance estimates based on full data information.

DISCUSSION

If in 778dividual data from all studies are available, then many shortcomings of meta-analysis based on published data are avoidable. More important is the possibility to adjust the summary odds ratio for confounders using individual data. Unfortunately, meta-analysis based on individual data has major obstacles. Blettner et al.¹ noticed that it is more expensive, time consuming, requires close cooperation between study coordinators and takes several years to complete.

In this article, a new method is proposed to make adjusted odds ratios across studies more comparable. The proposed method to combine exposure specific adjusted odds ratio at any degree within each study is equal to the correct value if the assumption of no interaction holds irrespective of how the determinant is classified. If interaction exists, the method can lead to incorrect standardised adjusted odds ratios. If there exists a determinant versus external variable interaction, however, then the odds ratios should not be adjusted for these external variables.

Interaction may also appear due to the different stratum specific determinant distribution. Combining exposure levels may then introduce an interaction between determinant and the covariate. The fact that, different studies used different arrangements of exposure levels, suggests that - at least for those studies- such interaction did not exist.

The proposed exposed versus non-exposed measure of the adjusted odds ratio is an unbiased estimator, given that the estimators of the exposure specific odds ratios in the primary study are also unbiased. The illustrations on both hypothetical and real data show that, the point estimate as well as the estimated large sample variance based on summarised data are almost equal to the correct point estimate and correct estimated variance (if full information data is used), respectively.

APPENDIX

Let $\{X_{ijl}\}_{i=1,\dots,L, j=1,\dots,K}$ be independent binomials with $X_{ijl} \sim \text{Bin}(n_{ij}, p_{ij})$, where $n_{ij} = X_{ij0} + X_{ij1}$ is the number of subjects, X_{ij0} is the number of non-cases, and X_{ij1} is the number of cases from stratum i and exposure category j . (Table 1) The risk p_{ij} is estimated by $\hat{p}_{ij} = \frac{X_{ij1}}{n_{ij}}$.

Furthermore, let $n_0 = \sum_{i=1}^L n_{i0}$ be the number of subjects in the non-exposed group with corresponding risk $p_0 = \frac{1}{n_0} \sum_{i=1}^L p_{i0}$. Denote the odds ratio from stratum i and exposure

level j as $\hat{\alpha}_{ij} = \frac{\hat{p}_{ij}}{1 - \hat{p}_{ij}} \cdot \frac{1 - \hat{p}_{i0}}{\hat{p}_{i0}} =: \frac{\hat{O}_{ij}}{\hat{O}_{i0}}, j = 1, \dots, K$. For the reduced table, let $C_j = \sum_{i=1}^L X_{ij1}$ and

$NC_j = \sum_{i=1}^L X_{ij0}$. Denote the estimated crude odds ratio of exposure category j as $\hat{\alpha}_j = \frac{NC_0 * C_j}{NC_j * C_0}$, and $\hat{\alpha}_{exposed}$ as the crude estimated odds ratio of exposed versus non-

exposed, i.e., $\hat{\alpha}_{exposed} = \frac{NC_0 * \sum_{j=1}^K C_j}{C_0 * \sum_{j=1}^K NC_j}$.

PROPERTY 1

Let $\hat{\alpha}_j, j=1,\dots,K$ be the crude estimated odds ratio of exposure level i relative to the zero reference level, and $\hat{\alpha}_{exposed}$ be the crude estimated odds ratio of exposed versus non-exposed. The following equation holds.

$$\hat{\alpha}_{exposed} = \frac{\sum_{j=1}^K NC_j * \hat{\alpha}_j}{\sum_{j=1}^K NC_j}. \quad (A1)$$

PROOF

$\hat{\alpha}_j = \frac{C_j * NC_0}{C_0 * NC_j}$ for each $j = 1, \dots, K$. Hence, $\sum_{j=1}^K (NC_j * \hat{\alpha}_j) = \frac{NC_0}{C_0} * \sum_{j=1}^K C_j$. Substitution into the formula for $\hat{\alpha}_{exposed}$ leads to

$$\hat{\alpha}_{exposed} = \frac{NC_0 * \sum_{j=1}^K C_j}{C_0 * \sum_{j=1}^K NC_j} = \frac{\frac{NC_0}{C_0} * \sum_{j=1}^K C_j}{\sum_{j=1}^K NC_j} = \frac{\sum_{j=1}^K NC_j * \hat{\alpha}_j}{\sum_{j=1}^K NC_j}$$

□

PROPERTY 2

Let $\wp$ be any rearrangement of the exposure level categories in table 1. Assume that the expected stratum (with respect to the confounders) specific odds ratios are all equal, i.e. $E(\hat{\alpha}_{ij}) = E(\hat{\alpha}_j^{adj})$. For all strata $i = 1, \dots, L$ and exposure levels $j \in \wp$. Let $\hat{\alpha}_{exposed}^{adj}$ be any estimator of the adjusted odds ratio, comparing only exposed with non-exposed group (reduced table), satisfying $E(\hat{\alpha}_{i\text{ exposed}}) = E(\hat{\alpha}_{exposed}^{adj})$ for all $i = 1, \dots, L$, where $\hat{\alpha}_{i\text{ exposed}}$ is an estimator of the stratum specific odds ratio in the reduced table. Then, the estimator of the adjusted odds ratio in the reduced table satisfies

$$E(\hat{\alpha}_{exposed}^{adj}) = \frac{\sum_{j \in \wp} NC_j * E(\hat{\alpha}_j^{adj})}{\sum_{j \in \wp} NC_j}. \quad (A2)$$

PROOF

Using property A1, the following equalities hold.

$$E(\hat{\alpha}_{i\text{ exposed}}) * \sum_{j \in \wp} X_{ij0} = \sum_{j \in \wp} X_{ij0} * E(\hat{\alpha}_{ij}), \text{ for all strata } i = 1, \dots, L.$$

Note that $\sum_{j \in \wp} (\sum_{i=1}^L X_{ij0}) = \sum_{j \in \wp} NC_j$. Consequently,

$$\begin{aligned} E(\hat{\alpha}_{exposed}^{adj}) &= E(\hat{\alpha}_{exposed}^{adj}) * \frac{\sum_{j \in \wp} (\sum_{i=1}^L X_{ij0})}{\sum_{j \in \wp} NC_j} = \frac{\sum_{i=1}^L (E(\hat{\alpha}_{i\text{ exposed}}^{adj}) * \sum_{j \in \wp} X_{ij0})}{\sum_{j \in \wp} NC_j} \\ &= \frac{\sum_{i=1}^L \sum_{j \in \wp} (X_{ij0} * E(\hat{\alpha}_{ij}))}{\sum_{j \in \wp} NC_j} = \frac{\sum_{j \in \wp} \sum_{i=1}^L (X_{ij0} * E(\hat{\alpha}_j^{adj}))}{\sum_{j \in \wp} NC_j} = \frac{\sum_{j \in \wp} NC_j * E(\hat{\alpha}_j^{adj})}{\sum_{j \in \wp} NC_j}. \end{aligned}$$

□

PROPERTY 3

Let $\ln \hat{\alpha}_j^{adj} = \sum_{i=1}^L w_{ij} \ln \hat{\alpha}_{ij}$, with $\sum_{i=1}^L w_{ij} = 1$ and $j = 1, \dots, K$

Then asymptotically ($n_{i0} \rightarrow \infty$, for all $i = 1, \dots, L$):

$$\text{cov}(\hat{\alpha}_m^{adj}, \hat{\alpha}_n^{adj}) = \alpha_m^{adj} \alpha_n^{adj} \sum_{i=1}^L \frac{\omega_{im} \omega_{in}}{n_{i0} p_{i0} (1 - p_{i0})}, \quad (A3)$$

for all $m, n = 1, \dots, K$.

PROOF

$$\text{cov}(\hat{\alpha}_m^{\text{adj}}, \hat{\alpha}_n^{\text{adj}}) = \text{cov}(\prod_{i=1}^L \hat{\alpha}_{im}^{w_{im}}, \prod_{i=1}^L \hat{\alpha}_{in}^{w_{in}}) = E(\prod_{i=1}^L \hat{\alpha}_{im}^{w_{im}} * \prod_{i=1}^L \hat{\alpha}_{in}^{w_{in}}) - \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} =$$

$$E(\prod_{i=1}^L \hat{O}_{im}^{w_{im}} * \prod_{i=1}^L \hat{O}_{in}^{w_{in}} \cdot \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} * \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}}) - \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} =$$

$$E(\prod_{i=1}^L \hat{O}_{im}^{w_{im}}) * E(\prod_{i=1}^L \hat{O}_{in}^{w_{in}}) \cdot E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} * \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}}) - \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} =$$

$$\alpha_m^{\text{adj}} \alpha_n^{\text{adj}} * \frac{E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} * \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})}{E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}}) * E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})} - \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} = \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} * \frac{\text{cov}(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} * \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})}{E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}}) * E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})}$$

The property follows by applying the extended version of the δ -method (Serfling, 1980, Theorem A on page 122). According to the central limit theorem we have

$\hat{p}_{ij}$ is $\text{AN}(\hat{p}_{ij}, \frac{1}{n_{ij}} * p_{ij}(1 - p_{ij}))$. A straightforward application of Theorem A leads to

$$\begin{pmatrix} \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} \\ \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}} \end{pmatrix} \text{ is AN } \left(\begin{pmatrix} \prod_{i=1}^L \frac{1}{O_{i0}^{w_{im}}} \\ \prod_{i=1}^L \frac{1}{O_{i0}^{w_{in}}} \end{pmatrix}, \begin{pmatrix} \sum_{i=1}^L \frac{w_{im}^2}{n_{i0} p_{i0} (1 - p_{i0})} \prod_{i=1}^L \frac{1}{O_{i0}^{2w_{im}}} & \sum_{i=1}^L \frac{w_{im} w_{in}}{n_{i0} p_{i0} (1 - p_{i0})} \prod_{i=1}^L \frac{1}{O_{i0}^{w_{im} + w_{in}}} \\ \sum_{i=1}^L \frac{w_{im} w_{in}}{n_{i0} p_{i0} (1 - p_{i0})} \prod_{i=1}^L \frac{1}{O_{i0}^{w_{im} + w_{in}}} & \sum_{i=1}^L \frac{w_{in}^2}{n_{i0} p_{i0} (1 - p_{i0})} \prod_{i=1}^L \frac{1}{O_{i0}^{2w_{in}}} \end{pmatrix} \right).$$

Hence asymptotically,

$$\text{cov}(\hat{\alpha}_m^{\text{adj}}, \hat{\alpha}_n^{\text{adj}}) = \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} * \frac{\text{cov}(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}} * \prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})}{E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{im}}}) * E(\prod_{i=1}^L \frac{1}{\hat{O}_{i0}^{w_{in}}})} =$$

$$\frac{\alpha_m^{\text{adj}} \alpha_n^{\text{adj}} \sum_{i=1}^L \frac{w_{im} w_{in}}{n_{i0} p_{i0} (1 - p_{i0})} \prod_{i=1}^L \frac{1}{O_{i0}^{w_{im} + w_{in}}}}{E(\prod_{i=1}^L \frac{1}{O_{i0}^{w_{im}}}) * E(\prod_{i=1}^L \frac{1}{O_{i0}^{w_{in}}})} = \alpha_m^{\text{adj}} \alpha_n^{\text{adj}} \sum_{i=1}^L \frac{w_{im} w_{in}}{n_{i0} p_{i0} (1 - p_{i0})}.$$

□

REFERENCES

1. Bleutner M, Sauerbrei W, Schlehofer B, Scheuchenspflug T, Friedenreich C. Traditional reviews, meta-analyses and pooled analyses in epidemiology. *Int J Epidemiol.* 1999;28:1-9.
2. Zeegers MPA, Tan FES, Verhagen AP, Weijnenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: A meta-analysis. *Cancer Causes and Control.* 1999;10:445-451.
3. Greenland S, Longnecker MP. Methods for trend estimation from summarised dose-response data, with application to meta-analysis. *Am J Epidemiol.* 1992;135:1301-1309.
4. Longnecker MP. Alcoholic beverage consumption in relation to risk of breast cancer: meta-analysis and review. *Cancer Causes and Control.* 1994;5:73-82.
5. Ursin G, Longnecker MP, Haile RW, Greenland S. A meta-analysis of body mass index and risk of premenopausal breast cancer. *Epidemiology.* 1995;6:137-141.
6. Berlin JA, Longnecker MP, Greenland S. Meta-analysis of epidemiologic dose-response data. *Epidemiology.* 1993;4:218-228.
7. Boyd NF, Martin LJ, Noffel M, Lockwood GA, Trichler DL. A meta-analysis of studies of dietary fat and breast cancer risk. *B J Cancer.* 1993;68:627-636.
8. Viscoli CM, Lachs MS, Horwitz RI. Bladder cancer and coffee drinking: a summary of case-control research. *Lancet.* 1993;341:1432-37.
9. Woolf B. On estimation the relation between blood group and disease. *Annals of Human Genetics.* 1954;19:251-253.
10. Pujolar AE, Gonzalez CA, Lopez-Abente G, Errezola M, Izarzugaza I, Nebot M, Riboli E. Bladder cancer and coffee consumption in smokers and nonsmokers in Spain. *Int J Epidemiol.* 1993;22:38-44.
11. Bravo MP, Del Rey Calero J, Conde M. Bladder cancer and the consumption of alcoholic beverages in Spain. *Eur J Epidemiol.* 1987;3:365-369.

3

The Impact of Characteristics of Cigarette Smoking on Urinary Tract Cancer Risk: a Meta-analysis of Epidemiologic Studies

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ABSTRACT

Background: Although narrative reviews have concluded that there is strong support for an association between cigarette smoking and urinary tract cancer, the association has never been quantified systematically in reviews. The purpose of this systematic review was to summarize and quantify the impact of different smoking characteristics (status, amount, duration, cessation, and age at first exposure) both unadjusted and adjusted for age and gender.

Methods: The authors included 43 epidemiologic studies (8 cohort and 35 case-control) and calculated summary odds ratios (SORs) by meta-regression analyses for different smoking characteristics. They also evaluated changes in summary estimates according to differences in study methodology.

Results: Smoking status and increased amount and duration of smoking were associated with a strong increased risk of urinary tract cancer. Smoking cessation and age at first exposure were negatively associated with the risk of urinary tract cancer. The age- and gender-adjusted SORs for current and former cigarette smokers compared with those for nonsmokers were 3.33 (95% confidence interval [CI], 2.63-4.21) and 1.98 (CI, 1.72-2.29), respectively. Even though the component studies differed in methodology, the results were rather consistent.

Conclusions: The results suggest a substantial increase in risk of cancer of the urinary tract for cigarette smokers. Based on the results of this study and previous literature, the authors conclude that current cigarette smokers have an approximately threefold higher risk of urinary tract cancer than nonsmokers. In Europe, approximately half of urinary tract cancer cases among males and one-third of cases among females might be attributable to cigarette smoking.

Over the last 4 decades, many epidemiologic studies have been conducted to investigate an association between cigarette smoking and the development of urinary tract cancer. Currently, a substantial amount of evidence has accumulated in support of a positive association between cigarette smoking and urinary tract cancer risk. A positive association has been consistently shown in both men and women in many different geographic areas. Similar results have been obtained in numerous case-control and follow-up studies.

Although cigarette smoking explains the occurrence of a greater amount of urinary tract cancer than does any other known environmental factor (e.g., occupation)¹ and no other environmental factor has been shown to confound this association, to our knowledge no systematic review on the association between several smoking characteristics and urinary tract cancer has been conducted.

Earlier narrative reviews on cigarette smoking and urinary tract cancer have summarized the association for current cigarette smoking compared with nonsmokers by estimating a general relative risk without calculation or systematic collection of data.¹⁻⁹ According to these narrative reviews, typical cigarette smokers have two to four times the risk of nonsmokers. Most narrative reviews suggested that the risk of urinary tract cancer increases with the number of cigarettes regularly smoked.^{1,3-11}

The magnitude of the effects of other cigarette smoking characteristics (e.g., smoking duration and cessation or age at first exposure) also has not been systematically reviewed. Although duration of smoking has been evaluated less often than intensity, some narrative reviews reported an unquantified positive dose-response relation.^{3,5,6} According to some reviews, former cigarette smokers seem to have a reduced incidence of urinary tract cancer as compared with current smokers.^{4-8,11} Age at first exposure to smoking has been reported only occasionally in narrative reviews of cigarette smoking and urinary tract cancer.

The purpose of the current study was to review all epidemiologic studies from 1966 to December 2000 more systematically; to provide quantitative summary estimates of the risk of urinary tract cancer with emphasis on smoking status, duration, amount, cessation, and age at first exposure based on these studies; and to evaluate changes in summary estimates according to differences in study methodology.

METHODS

Search strategy. The study design has been published previously.¹² Epidemiologic studies were identified through a computerized MEDLINE, CANCERLIT, and Current Contents searches for follow-up and case-control studies published until December 2000. The keywords used were urolo*, bladder, cyst*, vesic*, kidney, glomerul*, nephro*, pyel*, renal, ureteral, urethral, transitional cell, cancer, carcino*, tumor*, neoplasm*, onco*, risk, etiology, epidemiology, and caus*. References cited in published original and review articles were examined further. For inclusion in this analysis, the articles had to provide sufficient information to estimate a summary odds ratio and the associated standard error of incident primary urinary tract cancer for at least one of the following cigarette smoking characteristics: cigarette smoking status, average daily cigarette consumption, total duration of cigarette consumption, number of years since cessation, and age at first exposure of smoking. Urinary tract cancer was defined as cancer of the renal pelvis, ureter, urinary bladder, or urethra.

Data collection. We developed a criteria list for the assessment of quality items (study characteristics) in observational cancer research. This list is used to provide covariables for inclusion in meta-regression models to explore reasons for observed heterogeneity in results between observational studies. The criteria list has been validated on published articles on alcohol intake associated with bladder cancer through consensus meetings with experts on the fields of cancer and meta-analysis. The list calls for the following: general information - year of publication, research design (case-control study, follow-up study, other, unknown), and geographic area (Europe, United States, Asia, Africa,

unknown); exposure information - exposure measurement (personal interview, telephone interview, questionnaire, medical records, other, unknown), trained interviewer (yes, no, not applicable [n/a], unknown), validation exposure measurement (yes, no, unknown), and reference period (number of years, lifetime, unknown); case information - source cases (hospital, population, other, unknown), site carcinoma (renal pelvis, ureter, urinary bladder, urethra, urinary tract, unknown), histological confirmation cases (yes, no, unknown), and percentage transitional cell tumors case-control study information - source controls (hospital, population, neighborhood, other, n/a, unknown), response rate (percentage, n/a, unknown), and blinding of case status (yes, no, n/a, unknown); follow-up study information - source study population (volunteer, population, other, n/a, unknown), years of follow-up (number of years, n/a, unknown), blinding of exposure status (yes, no, n/a, unknown), and completeness of follow-up (percentage, n/a, unknown).

We extracted data allowing us to calculate both unadjusted and adjusted odds ratios to estimate the association between cigarette smoking and the risk of urinary tract cancer. We constructed two-way contingency tables for each study, based on exposure frequency distributions, to calculate the unadjusted odds ratios. Adjusted odds ratios were extracted directly from the original reports. Because we considered age and gender to be the most important confounding variables, the authors of the original articles had to have adjusted for at least these two variables for inclusion in the calculation of adjusted summary estimates. If studies reported gender-stratified age-adjusted odds ratios, we combined these estimates by calculating age- and gender-adjusted odds ratios,¹³ because from both theoretically and statistically points of view, gender is probably a confounder in the association between cigarette smoking and bladder cancer. For studies that reported separate adjusted odds ratios for several exposure strata, we combined the exposure specific odds ratios by using the prevalence of the noncases as weight.¹⁴ Summary odds ratios were calculated for smoking status (non, former, and current smoker), smoking amount (0, 1-20, and >20 cigarettes/day), smoking duration (≤ 20 and >20 years), smoking cessation (>10, and ≤ 10 years) and age at first exposure of smoking (>20 and ≤ 20 years). Unfortunately, most component studies did not include simultaneously different smoking characteristics in a regression model to estimate the impact of cigarette smoking status, amount, duration, and age at first exposure solely.

Statistical analysis. To detect publication or related biases, we explored heterogeneity in funnel plots, i.e., plots of effect estimates against their estimated precision (reciprocal of the variance).¹⁵ We examined funnel plot asymmetry visually and measured the degree of asymmetry by using Egger's unweighted regression asymmetry test.¹⁶ If a study has appeared in more than one publication, data from the last publication were used for statistical analysis. We estimated the summary odds ratios and corresponding 95% confidence intervals (CIs) with random effects meta regression analysis by using the Stata statistical software.¹⁷ The between-study variance was estimated iteratively, by using the empiric Bayes method.¹⁸ We analyzed the results for men and women both separately and combined, depending on available data in the original studies. To explore reasons for the observed heterogeneity, we performed sensitivity analyses on study characteristics and tested their influence on the association between current cigarette smoking and urinary tract cancer. We estimated the population attributable risk of urinary tract cancer for men and women based on the proportion of cigarette smokers in the European Union and the results of the current meta-analysis.

Table 1. Study characteristics of published epidemiologic studies concerning cigarette smoking and the status of the urinary tract, ordered by year of publication

Ref.	First author	Year	Country	Anatomic site	Study design			Cigarette smoking assessment
					Cohort study	Case-control study		
						Case source	Control source	
(19)	Dunham	1968	US	Bladder	-	Both	Hospital	Interview
(20)	Anthony	1970	UK	Bladder	-	Hospital	Hospital	Interview
(21)	Tyrrell	1971	Ireland	Bladder	-	Hospital	Hospital	Interview
(22)	Armstrong	1976	UK	Renal pelvis	-	Hospital	Hospital	Interview
(23)	Miller	1977	Canada	Bladder	-	Population	Population*	Interview
(24)a	Wynder	1977	US	Bladder	-	Hospital	Hospital	Interview
(25)	Tola	1980	Finland	Bladder	-	Population	Hospital	Questionnaire†
(26)b	Vineis	1981	Italy	Bladder	-	Hospital	Hospital	Interview
(27)b	Vineis	1983	Italy	Urinary tract†	-	Hospital	Hospital	Interview
(28)b	Vineis	1984	Italy	Bladder	-	Hospital	Hospital	Interview
(29)	Morrison	1984	US/UK/Japan	Urinary tract†	-	Hospital	Population	Interview
(30)c	Hartge	1985	US	Bladder	-	Population	Population	Interview
(31)c	Marret	1985	US	Bladder	-	Population	Population	Interview
(32)	Rebekalos	1985	Greece	Bladder	-	Hospital	Hospital	Interview
(33)b	Vineis	1985	Greece	Bladder	-	Hospital	Hospital	Interview
(34)a	Wynder	1985	US	Bladder	-	Hospital	Hospital	Interview
(35)	Bravo	1986	Spain	Urinary tract†	-	Hospital	Hospital	Questionnaire†
(36)	Brownson	1987	US	Bladder	-	Population	Hospital	Questionnaire†
(37)c	Hartge	1987	US	Bladder	-	Population	Population	Interview
(38)b	Vineis	1988	Italy	Bladder	-	Hospital	Hospital	Interview
(39)c	Slatterly	1988	US	Bladder	-	Population	Population	Interview
(40)	Steineck	1988	Sweden	Urinary tract†	Yes	-	-	Questionnaire†
(41)a	Augustine	1988	US	Bladder	-	Hospital	Hospital	Interview
(42)d	La Vecchia	1989	Italy	Bladder	-	Hospital	Hospital	Interview
(43)	Burch	1989	Canada	Bladder	-	Both	Population	Interview
(44)	Helzlsouer	1989	US	Bladder	Yes	-	-	Questionnaire†
(45)e	Clavel	1989	France	Bladder	-	Hospital	Hospital	Interview
(46)	Ross	1989	US	Renal pelvis§	-	Population	Neighborhood	Interview
(47)d	D'Avanzo	1990	Italy	Bladder	-	Hospital	Hospital	Interview
(48)c	Hartge	1990	US	Bladder	-	Population	Population	Interview
(49)	Iyer	1990	US	Bladder	-	Hospital	Hospital	Interview
(50)a	Harris	1990	US	Bladder	-	Hospital	Hospital	Interview
(51)d	La Vecchia	1991	Italy	Bladder	-	Hospital	Hospital	Interview
(52)	Mills	1991	US	Bladder	Yes	-	-	Questionnaire†
(53)	Lopez-Abente	1991	Spain	Bladder	-	Hospital	Both	Interview
(54)	De Stefani	1991	Uruguay	Bladder	-	Hospital	Hospital	Interview
(55)	Burns	1991	US	Bladder	-	Population	Hospital	Interview
(56)d	D'Avanzo	1992	Italy	Bladder	-	Hospital	Hospital	Interview
(57)	Kunze	1992	Germany	Urinary tract†	-	Hospital	Hospital	Interview
(58)	McLaughlin	1992	US	Renal pelvis§	-	Population	Population	Interview
(59)e	Cordier	1993	France	Bladder	-	Hospital	Hospital	Interview
(60)	Chyou	1993	US	Urinary tract†	Yes	-	-	Interview
(61)	Hayes	1993	US	Urinary tract†	-	Hospital	Population	Interview
(62)	Sorahan	1994	UK	Urinary tract†	-	Population	Both**	Questionnaire†
(63)d	Barbone	1994	Italy	Bladder	-	Hospital	Hospital	Interview
(64)	Vizcaino	1994	Zimbabwe	Bladder	-	Population	Hospital	Interview
(65)	Momas	1994	France	Bladder	-	Population	Population	Interview
(66)c	Sturgeon	1994	US	Bladder	-	Population	Population	Interview
(67)	Tremblay	1995	Canada	Bladder	Yes	-	-	Medical files
(68)d	D'Avanzo	1995	Italy	Bladder	-	Hospital	Hospital	Interview
(69)	McCarthy	1995	US	Bladder	Yes	-	-	Questionnaire†
(70)	Murata	1996	Japan	Bladder	Yes	-	-	Questionnaire†
(71)	Bruemmer	1996	US	Bladder	-	Population	Population	Both
(72)	Engeland	1996	Norwegen	Urinary tract†	Yes	-	-	Questionnaire†
(73)	Bedwani	1997	Egypt	Bladder	-	Hospital	Hospital	Interview
(74)	Donato	1997	Italy	Bladder	-	Hospital	Hospital	Interview
(75)	Teschke	1997	Canada	Bladder	-	Population	Population	Interview
(76)	Sorahan	1998	US	Urinary tract†	-	Hospital	Hospital	Questionnaire†
(77)	Koivusalo	1998	Finland	Bladder	-	Population	Population	Questionnaire†

*and neighborhood

[†]self-administered questionnaire

[‡]includes bladder carcinoma and at least one other urinary tract cancer

[§]and ureter

** only data from population controls were used

a**same study has appeared in more than one publication

RESULTS

Study Characteristics. We identified 59 articles reporting follow-up or case-control studies on cigarette smoking and incident urinary tract cancer published between 1968 and 1998¹⁹⁻⁷⁷ (Table 1). Generally, the association between cigarette smoking and urinary tract cancer was not the main research hypothesis. Eighteen articles were excluded from the analyses because the same study appeared in publications that were more recent. The remaining 41 articles described 8 follow-up studies^{40,44,52,60,67,69,70,72} and 35 case-control studies.^{9-23,25,29,32,35,36,41,43,46,49,50,53-55,57-59,61,62,64-66,68,71,73-77} One case-control study that provided separate associations for parts of the study performed in the United States, United Kingdom, and Japan was considered as three separate studies.⁹ The case-control studies were population-based (n=12),^{23,29,43,58,61,65,66,71,75,77} hospital-based (n=20),^{19-22,25,32,35,36,38,49,50,54,55,57,59,64,68,73,74,76} or neighborhood-based (n=1).⁴⁶ Two case-control studies used both population- and hospital-based controls.^{53,62} The controls in most hospital-based case-control studies did not have any smoking-related disease. The case-control studies also varied with regard to their criteria of case selection. Thirteen case-control studies identified cancer cases in defined populations,^{23,25,36,46,55,58,62,64-66,71,75,77} Twenty case-control studies selected cases from hospitals,^{20-22,29,32,35,38,49,50,53,54,57,59,61,68,73,74,76} and two case-control studies used both populations and hospitals.^{19,43} Information on cigarette smoking was obtained by interview (n=29),^{19-23,29,32,38,43,46,49,50,53-55,57-61,64-66,68,73-75} self-administered questionnaire (n=12)^{25,35,36,40,44,52,62,69,70,72,76,77} or both techniques (n=1).⁷¹ One follow-up study used medical files to obtain data on cigarette smoking.⁶⁷ Some studies included all neoplasms of the urinary tract as cases, of which greater than 90% were found to involve bladder cancer (n=11).^{29,35,40,57,60-62,72,76} Other studies defined case status by incident bladder cancer (n=29),^{19-21,23,25,32,36,38,43,44,49,50,52-55,59,64-71,73-75,77} carcinomas of the renal pelvis (n=1)²² or carcinomas of renal pelvis and ureter combined (n=2).^{46,58} Most studies used histologically confirmed cases with transitional cell carcinomas (Table 1).

Figure 1. Funnel plot for current cigarette smokers compared with nonsmokers is shown. Dashed and solid reference lines indicate no effect and total summary odds ratio, respectively

Table 2. Unadjusted summary odds ratios for different cigarette smoking characteristics

Determinants	Male			Female			Male and Female		
	n*	OR	CI	n*	OR	CI	n*	OR [†]	CI
Smoking status									
Nonsmoker		1.00	Reference		1.00	Reference		1.00	Reference
Former smoker	23	2.00	1.57-2.55	12	1.66	1.13-2.44	22	1.71	1.51-1.94
Current smoker	24	2.81	2.31-3.43	15	2.33	1.82-2.99	23	2.57	2.20-3.00
Smoking amount									
Nonsmoker		1.00	Reference		1.00	Reference		1.00	Reference
1-20 cigarettes/day	11	2.34	1.77-3.09	7	1.79	1.39-2.30	14	2.17	1.75-2.70
>20 cigarettes/day	11	2.91	2.09-4.06	5	2.57	2.24-2.94	14	2.79	2.00-3.90
Smoking duration (yrs)									
≤20		1.00	Reference		1.00	Reference		1.00	Reference
>20	10	2.59	1.83-3.67	5	2.73	1.63-4.57	5	2.13	1.70-2.67
Smoking cessation (yrs)									
>10		1.00	Reference		1.00	Reference		1.00	Reference
≤10	6	1.23	0.80-1.87	2	0.38	0.17-0.85	4	1.36	0.76-2.43
Age at first exposure (yrs)									
>20		1.00	Reference		1.00	Reference		1.00	Reference
≤20	13	1.25	1.07-1.47	3	1.70	1.09-2.65	5	1.26	1.12-1.42

OR: odds ratio; CI 95% confidence interval

*No. of analyzed studies

[†]Calculated from collapsed contingency tables

RISK ESTIMATION

We could not identify heterogeneity in funnel plots, neither visually nor in terms of statistical significance (p values ≥ 0.40 for current smoking) (Figure 1). Tables 2 and 3 summarize the unadjusted and adjusted results of observational studies reporting the associations for different cigarette smoking characteristics, respectively.

Current cigarette smokers have approximately three times the risk of urinary tract cancer of nonsmokers. The adjusted summary odds ratios for current cigarette smokers compared with nonsmokers were 3.18 (CI, 2.35-4.29) for studies with men, 2.90 (CI, 2.01-4.19) for studies with women, and 3.33 (CI, 2.63-4.21) for studies with men and women combined (Table 3). Smoking cessation might be beneficial, although former smokers still have an increased risk of urinary tract cancer compared with nonsmokers. The adjusted summary odds ratios for former smokers were 2.90 (CI, 1.41-5.98), 1.34 (CI, 1.03-1.74), and 1.98 (CI, 1.72-2.29) for studies with men only, women only, or men and women combined, respectively (Table 3).

The risk of urinary tract cancer is associated with the number of cigarettes smoked per day (Table 3). The adjusted summary odds ratios for smoking up to 20 cigarettes per day ranged from 1.66 (CI, 0.93-2.97) for studies with women to 2.66 (CI, 2.06-3.42) for studies with men. Men or women who smoked more than 20 cigarettes per day appeared to have higher risks. The adjusted summary odds ratios were: 3.51 (CI, 2.73-4.52), 2.48 (CI, 1.34-4.61), and 3.15 (CI, 2.62-3.79) for studies with men only, women only, or men and women combined, respectively (Table 3).

For both smoking status and smoking amount, the unadjusted estimation for the summary odds ratios were usually lower than the age- and gender-adjusted estimates, although the unadjusted estimates were based on a larger set of studies (Table 2). For smoking duration, smoking cessation, and age at first exposure of smoking, only unadjusted summary odds ratios could be calculated.

The risk of urinary tract cancer increased with increasing duration of cigarette smoking (Table 2). Subjects who smoked for greater than 20 years appeared to develop urinary tract cancer at 2-3 times the rate in subjects who smoked cigarettes for less than 20 years. The corresponding summary odds ratios were 2.59 (CI, 1.83-3.67) for studies with men, 2.73 (CI, 1.63-4.57) for studies with women and 2.13 (CI, 1.70-2.67) for studies in which the data for men and women were collapsed (Table 2).

Table 3. Adjusted summary odds ratios for different cigarette smoking characteristics

Determinants	Male			Female			Male and Female		
	n*	OR [†]	CI	n*	OR [†]	CI	n*	OR [†]	CI
Smoking status									
Nonsmoker		1.00	Reference		1.00	Reference		1.00	Reference
Former smoker	14	2.90	1.41-5.98	6	1.34	1.03-1.74	13	1.98	1.72-2.29
Current smoker	13	3.18	2.35-4.29	7	2.90	2.01-4.19	13	3.33	2.63-4.21
Smoking amount									
Nonsmoker		1.00	Reference		1.00	Reference		1.00	Reference
<20 cigarettes/day	9	2.66	2.06-3.42	4	1.66	0.93-2.97	6	2.04	1.82-2.30
≥20 cigarettes/day	9	3.51	2.73-4.52	4	2.48	1.34-4.61	6	3.15	2.62-3.79

OR: odds ratio; CI 95% confidence interval

*number of analyzed studies

[†]adjusted for age and gender

[‡]adjusted for age

The time since smoking cessation among former smokers also appeared to be an important smoking characteristic (Table 2). Men who stopped smoking for less than 10 years had higher risks of urinary tract cancer compared with men who stopped smoking for longer than 10 years (summary odds ratio, 1.23; CI, 0.80-1.87). However, the reduction in risk for women appeared to be greatest in the first decade after quitting, although this is only based on two case-control studies (summary odds ratio, 0.38; CI, 0.17-0.85).^{43,57} The summary odds ratio for studies with collapsed data on men and women was 1.36 (CI, 0.76-2.43) (Table 2).

Furthermore, persons who started smoking at younger ages (younger than 20 years) tended to have higher risks of urinary tract cancer compared with persons who start smoking at older ages (Table 2). The corresponding summary odds ratios were 1.25 (CI, 1.07-1.47) for studies with men, 1.70 (CI, 1.09-2.65) for studies with women and 1.26 (CI, 1.12-1.42) for studies with men and women combined (Table 2).

Figure 2. Forest plot is shown of crude summary odds ratios and 95% confidence intervals of urinary tract cancer for current cigarette smokers compared with nonsmokers, by study design, measuring instrument, source of cases and controls, and tumor site for case-control studies with men and women combined. Dashed and solid vertical reference lines indicate no effect and total summary odds ratio, respectively

Sensitivity analysis. We further examined the crude association of current smoking by geographic area, year of publication, study design, measuring instrument, sources of cases and controls, and anatomic site of the tumor to explore their influence on the outcome estimates in studies that provided information for men and women combined (Figure 2). No tests for interaction were statistically significant. Most subset specific summary odds ratios did not differ substantially, although it appeared that the odds ratios from studies published before 1980 were lower than from more recent studies. Furthermore, the summary associations for case-control studies were higher than for follow-up studies. Selection on anatomic site of the tumor did not alter the summary odds ratios (Figure 2).

Population attributable risk. In the European Union, 28% of women and 43% of men smoke cigarettes.⁷⁸ Based on these figures and the age-adjusted results of the current meta-analysis, our estimates show that cigarette smoking might account for 35% of all female urinary tract cancer, whereas in men 50% of incidences of the disease may be associated with cigarette smoking.

DISCUSSION

The possible association between cigarette smoking and cancer of the urinary tract has been extensively investigated in 43 epidemiologic studies. These primary studies can be considered as the best available evidence. The summarized findings suggest a substantial increase of risk of urinary tract cancer for cigarette smokers. Smoking amount and smoking duration were positively associated with urinary tract cancer risk. For age at first exposure and smoking cessation, a negative association was found.

Unfortunately, the included studies did not provide sufficient information to estimate adjusted summary odds ratios for all smoking characteristics. For smoking duration, smoking cessation, and age at first exposure of smoking only unadjusted summary odds ratios could be calculated. The summary odds ratios for smoking status and smoking amount increased after adjustment for age and gender. Therefore, we expect the crude estimates for smoking duration, smoking cessation, and age at first exposure to be underestimated.

We did not attempt to uncover unpublished observations and excluded studies that did not meet the predetermined criteria. Publication bias might arise by excluding these studies. However, we could not identify funnel plot heterogeneity in our meta-analysis, either visually or in terms of statistical significance.

Because of potential heterogeneity in populations, designs, and analyses of various studies, we assumed that the true effects being estimated would vary between the studies in addition to the usual sampling variation in the estimates (within studies). To account for both sources of variation, we used random effects meta regression analysis to combine the results from the primary studies.¹⁸ The random effect approach provides some allowance for heterogeneity in studies beyond sampling error.

The epidemiology of urinary tract cancer is rather complex. For example: substantial differences exist in urinary tract cancer rates between white and black people; urinary tract cancer is considerably more common in men than in women; and the incidence of this cancer varies between North America and Europe.⁷⁹ The race of the study population in almost all component studies was white. Therefore, the influence of race on the association between cigarette smoking and urinary tract cancer could not be investigated in the current meta-analysis. For both men and women, we found similarly increased urinary tract cancer risks for cigarette smoking. Furthermore, the summary odds ratios were similar for different geographic areas.

Results from sensitivity analyses suggested that the summary odds ratios were comparable for different types of exposure measurement, for different tumor sites, and for different sources of the cases and controls in the case-control studies. The summary estimates were also similar between the different years of publication, although studies published before 1980 yielded to a lower summary odds ratio than studies published

after 1980. This difference could not be explained by diversity in population or methodology between studies published before and after 1980 and is probably an artifact of chance. It appeared that the summary estimates of case-control studies were somewhat higher than for follow-up studies. This contrast, although not statistically significant, might be a consequence of differential recall bias in case-control studies because patients with bladder cancer are possibly more sensitized toward recalling smoking habits than noncases.

The precise mechanism by which cigarette smoking causes urinary tract cancer has yet to be determined. The finding that both age at first exposure and cessation of exposure have an influence in modifying the summary odds ratio of urinary tract cancer might suggest that two stages in the mechanism of urinary tract carcinogenicity are involved, one early and one late.⁸⁰ However, few studies simultaneously included different smoking characteristics in a regression model to estimate the independent contribution of these smoking characteristics. It seems most likely that the risk of urinary tract cancer is related to some of the large number of chemicals present in smoke. 2-Naphthylamine and 4-aminobiphenyl are the leading candidates as the specific etiologic agents.^{3,8,9,81,82} Several nitrosamines have been shown to produce bladder cancer in animal models.⁹ Tars might induce bladder papillomas and carcinomas in mice.⁸ To our knowledge, no epidemiologic study on the association between tars and nicotine exposure and bladder cancer risk has been conducted, although one case-control study reported a diminution of risk from the smoking of light tobacco.⁵³ Molecular studies have suggested that exposure to certain carcinogens in cigarette smoke may contribute to DNA damage, i.e., chromosome 9 defects⁸³ and TP53 mutations.⁸⁴ These alterations are the most frequently known molecular abnormalities in the etiology of bladder cancer.⁸³ Furthermore, genetic polymorphisms, e.g., of the arylamine *N*-acetyltransferase or glutathione *S*-transferase Mu1 (GSTM1), may alter metabolism of tobacco carcinogens. Slow acetylation or lack of GSTM1 activity (which is present in 50% of whites) might result in a higher concentration of tobacco carcinogens in the bladder and consequently enhance the risk of bladder cancer among cigarette smokers.⁸⁵⁻⁸⁷ Unfortunately, the data of the current meta-analysis could not be stratified upon these or other polymorphisms. Besides the effect that many compounds in cigarettes can cause genotoxic events in the urothelium, cigarette smoking have been found to increase proliferation, as evidenced by hyperplasia of the urinary tract epithelium.⁹

In accordance with earlier reviews, it can be concluded that cigarette smoking is an important cause of urinary tract cancer for both men and women. Current cigarette smokers have an approximately threefold higher risk of urinary tract cancer than nonsmokers. This risk increases with the number of cigarettes smoked per day and the number of years smoked. Both age at first exposure and cessation of cigarette smoking have an influence on modifying the risk of urinary tract cancer. Approximately half of male urinary tract cancer and one-third of female urinary tract cancer might be attributable to cigarette smoking.

REFERENCES

1. Morrison AS. Advances in the etiology of urothelial cancer. *Urol Clin North Am.* 1984;11:557-66.
2. Matanoski GM and Elliott EA. Bladder cancer epidemiology. *Epidemiol Rev.* 1981;3:203-29.
3. Dolin PJ. An epidemiological review of tobacco use and bladder cancer. *J Smoking Related Dis.* 1991;2:129-43.
4. Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am.* 1992;6:1-30.
5. Shirai T. Etiology of bladder cancer. *Semin Urol.* 1993;11:113-26.
6. Shirai T, Fradet Y, Huland H, Bollack C, Droller M, Janknegt R, et al. The etiology of bladder cancer: are there any new clues or predictors of behavior? *Int J Urol.* 1995;2 (3 Suppl):64-75.
7. Silverman DT, Morrison AS, Devesa SS. Bladder cancer. In: Schottenfeld D and Fraumeni JF. *Cancer epidemiology and prevention.* New York and Oxford: Oxford University Press, 1996:1156-79.
8. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol.* 1996;23:536-45.
9. Johansson SL and Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol.* 1997;13:291-8.

10. World Health Organization. IARC monographs on the evaluation of carcinogenic risks to humans: tobacco smoking. Lyon: World Health Organization, 1986.
11. van der Meijden APM. Bladder cancer. *Br Med J*. 1998;317:1366-9.
12. Zeegers MPA, Tan FES, Verhagen AP, Weijnenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.
13. Woolf B. On estimation the relation between blood group and disease. *Ann Hum Genet*. 1954;19:251-3.
14. Tan FES and Zeegers MPA An alternative method of combining exposure specific odds ratios in epidemiologic studies. Research report. Maastricht: Maastricht University, Department Methodology and Statistics, 1999.
15. Light RJ and Pillemer DB. Quantitative procedures. In: Summing up: the science of reviewing research. Cambridge, MA: Harvard University Press, 1984.
16. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *Br Med J*. 1997;315:629-34.
17. StataCorp Stata Statistical Software: Release 6.0. . College Station, TX: Stata Corporation, 1999.
18. Berkey CS, Hoaglin DC, Mosteller F, Colditz GA. A random-effects regression model for meta-analysis. *Stat Med*. 1995;14:395-411.
19. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in New Orleans, Louisiana. *J Natl Cancer Inst*. 1968;41:683-709.
20. Anthony HM and Thomas GM. Bladder tumours and smoking. *Int J Cancer*. 1970;5:266-72.
21. Tyrrell AB, MacAirt JG, McCaughey WT. Occupational and non-occupational factors associated with vesical neoplasm in Ireland. *J Ir Med Assoc*. 1971;64:213-7.
22. Armstrong B, Garrod A, Doll R. A retrospective study of renal cancer with special reference to coffee and animal protein consumption. *Br J Cancer*. 1976;33:127-36.
23. Miller AB and Howe GR. Artificial sweeteners and bladder cancer. *Lancet*. 1977;10:1221-2.
24. Wynder EL and Goldsmith R. The epidemiology of bladder cancer: a second look. *Cancer*. 1977;40:1246-68.
25. Tola S, Tenho M, Korkala ML, Jarvinen E. Cancer of the urinary bladder in Finland: association with occupation. *Int Arch Occup Environ Health*. 1980;46:43-51.
26. Vineis P, Segnan N, Costa G, Terracini B. Evidence of a multiplicative effect between cigarette smoking and occupational exposures in the aetiology of bladder cancer. *Cancer Lett*. 1981;14:285-90.
27. Vineis P, Frea B, Uberti E, Ghisetti V, Terracini B. Bladder cancer and cigarette smoking in males: a case-control study. *Tumori*. 1983;69:17-22.
28. Vineis P, Esteve J, Terracini B. Bladder cancer and smoking in males: types of cigarettes, age at start, effect of stopping and interaction with occupation. *Int J Cancer*. 1984;34:165-70.
29. Morrison AS, Buring JE, Verhoek WG, Aoki K, Leck I, Ohno Y, et al. An international study of smoking and bladder cancer. *J Urol*. 1984;131:650-4.
30. Hartge P, Hoover R, Kantor A. Bladder cancer risk and pipes, cigars and smokeless tobacco. *Cancer*. 1985;15:901-6.
31. Marrett LD, Meigs JW, Flannery JT. Bladder cancer in Connecticut: the role of cigarette smoking and other risk factors. *Conn Med*. 1985;49:718-26.
32. Rebekalos A, Trichopoulos D, Tzonou A, Zavitsanos X, Velonakis E, Trichopoulos A. Tobacco smoking, coffee drinking, and occupation as risk factors for bladder cancer in Greece. *J Natl Cancer Inst*. 1985;75:455-61.
33. Vineis P, Ciccone G, Ghisetti V, Terracini B. Cigarette smoking and bladder cancer in females. *Cancer Lett*. 1985;26:61-6.
34. Wynder EL, Dieck GS, Hall NE, Lahti H. A case-control study of diesel exhaust exposure and bladder cancer. *Environ Res*. 1985;37:475-89.
35. Bravo P, del Rey Calero J, Sanchez J, Conde M. El tabaco como factor de riesgo del cancer de vejiga. *Arch Esp Urol*. 1986;39:237-40.
36. Brownson RC, Chang JC, Davis JR. Occupation, smoking, and alcohol in the epidemiology of bladder cancer. *Am J Public Health*. 1987;77:1298-300.
37. Hartge P, Silverman D, Hoover R, Schairer C, Altman R, Austin D, et al. Changing cigarette habits and bladder cancer risk: a case-control study. *J Natl Cancer Inst*. 1987;78:1119-25.
38. Vineis P, Esteve J, Hartge P, Hoover R, Silverman DT, Terracini B. Effects of timing and type of tobacco in cigarette-induced bladder cancer. *Cancer Res*. 1988;48:3849-52.
39. Slattery ML, Schumacher MC, West DW, Robison LM. Smoking and bladder cancer. The modifying effect of cigarettes on other factors. *Cancer*. 1988;61:402-8.
40. Steineck G, Norell SE, Feychting M. Diet, tobacco and urothelial cancer: a 14-year follow-up of 16,477 subjects. *Acta Oncol*. 1988;27:323-7.
41. Augustine A, Hebert JR, Kabat GC, Wynder EL. Bladder cancer in relation to cigarette smoking. *Cancer Res*. 1988;48:4405-8.
42. La Vecchia C, Negri E, Decarli A, B. DA, Liberati C, Franceschi S. Dietary factors in the risk of bladder cancer. *Nutr Cancer*. 1989;12:93-101.
43. Burch JD, Rohan TE, Howe GR, Risch HA, Hill GB, Steele R, et al. Risk of bladder cancer by source and type of tobacco exposure: a case-control study. *Int J Cancer*. 1989;44:622-8.
44. Helzlsouer KJ, Comstock GW, Morris JS. Selenium, lycopene, alpha-tocopherol, beta-carotene, retinol, and subsequent bladder cancer. *Cancer Res*. 1989;49:6144-8.
45. Clavel J, Cordier S, Boccon Gibod L, Hemon D. Tobacco and bladder cancer in males: increased risk for inhalers and smokers of black tobacco. *Int J Cancer*. 1989;44:605-10.
46. Ross RK, Paganini Hill A, Landolph J, Gerkins V, Henderson BE. Analgesics, cigarette smoking, and

- other risk factors for cancer of the renal pelvis and ureter. *Cancer Res.* 1989;49:1913-6.
47. D'Avanzo B, Negri E, La Vecchia C, Gramenzi A, Bianchi C, Franceschi S, et al. Cigarette smoking and bladder cancer. *Eur J Cancer.* 1990;26:714-8.
48. Hartge P, Harvey EB, Marston Linchan W, Silverman DT, Sullivan JW, Hoover RN, et al. Unexplained excess risk of bladder cancer in men. *J Natl Cancer Inst.* 1990;82:1636-40.
49. Iyer V, Harris RE, Wynder EL. Diesel exhaust exposure and bladder cancer risk. *Eur J Epidemiol.* 1990;6:49-54.
50. Harris RE, Chen Backlund JY, Wynder EL. Cancer of the urinary bladder in blacks and whites: a case-control study. *Cancer.* 1990;66:2673-80.
51. La Vecchia C, Negri E, B DA, Savoldelli R, Franceschi S. Genital and urinary tract diseases and bladder cancer. *Cancer Res.* 1991;51:629-31.
52. Mills PK, Beeson WL, Phillips RL, Fraser GE. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol.* 1991;133:230-9.
53. Lopez Abente G, Gonzalez CA, Errezola M, Escolar A, Izarzugaza I, Nebot M, et al. Tobacco smoke inhalation pattern, tobacco type, and bladder cancer in Spain. *Am J Epidemiol.* 1991;134:830-9.
54. De Stefani E, Correa P, Fierro L, Fontham E, Chen V, Zavala D. Black tobacco, mate, and bladder cancer: a case-control study from Uruguay. *Cancer.* 1991;67:536-40.
55. Burns PB and Swanson GM. Risk of urinary bladder cancer among blacks and whites: the role of cigarette use and occupation. *Cancer Causes Control.* 1991;2:371-9.
56. D'Avanzo B, La Vecchia C, Franceschi S, Negri E, Talamini R, Buttino I. Coffee consumption and bladder cancer risk. *Eur J Cancer.* 1992;28a:1480-4.
57. Kunze E, Chang Claude J, Frentzel Beyme R. Life style and occupational risk factors for bladder cancer in Germany. A case-control study. *Cancer.* 1992;69:1776-90.
58. McLaughlin JK, Silverman DT, Hsing AW, Ross RK, Schoenberg JB, Yu MC, et al. Cigarette smoking and cancers of the renal pelvis and ureter. *Cancer Res.* 1992;52:254-7.
59. Cordier S, Clavel J, Limasset JC, Boccon Gibod L, Le Moual N, Mandereau L, et al. Occupational risks of bladder cancer in France: a multicentre case-control study. *Int J Epidemiol.* 1993;22:403-11.
60. Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol.* 1993;3:211-6.
61. Hayes RB, Friedell GH, Zahm SH, Cole P. Are the known bladder cancer risk-factors associated with more advanced bladder cancer? *Cancer Causes Control.* 1993;4:157-62.
62. Sorahan T, Lancashire RJ, Sole G. Urothelial cancer and cigarette smoking: findings from a regional case-controlled study. *Br J Urol.* 1994;74:753-6.
63. Barbone F, Franceschi S, Talamini R, Bidoli E, La Vecchia C. Occupation and bladder cancer in Pordenone (north-east Italy): a case-control study. *Int J Epidemiol.* 1994;23:58-65.
64. Vizzaino AP, Parkin DM, Boffetta P, Skinner MEG. Bladder cancer: epidemiology and risk factors in Bulawayo, Zimbabwe. *Cancer Causes Control.* 1994;5:517-22.
65. Momas I, Daures JP, Festy B, Bontoux J, Gremy F. Bladder cancer and black tobacco cigarette smoking. Some results from a French case-control study. *Eur J Epidemiol.* 1994;10:599-604.
66. Sturgeon SR, Hartge P, Silverman DT, Kantor AF, Linehan WM, Lynch C, et al. Associations between bladder cancer risk factors and tumor stage and grade at diagnosis. *Epidemiology.* 1994;5:218-25.
67. Tremblay C, Armstrong B, Theriault G, Brodeur J. Estimation of risk of developing bladder cancer among workers exposed to coal tar pitch volatiles in the primary aluminum industry. *Am J Ind Med.* 1995;27:335-48.
68. D'Avanzo B, La Vecchia C, Negri E, Decarli A, Benichou J. Attributable risks for bladder cancer in northern Italy. *Ann Epidemiol.* 1995;5:427-31.
69. McCarthy PV, Bhatia AJ, Saw SM, Mosley JD, Vega-Quinones A. Cigarette smoking and bladder cancer: in Washington County, Maryland: ammunition for health educators. *Md Med J.* 1995;44:1039-42.
70. Murata M, Takayama K, Choi BC, Pak AW. A nested case-control study on alcohol drinking, tobacco smoking, and cancer. *Cancer Detect Prev.* 1996;20:557-65.
71. Bruemmer B, White E, Vaughan TL, Cheney CL. Nutrient intake in relation to bladder cancer among middle-aged men and women. *Am J Epidemiol.* 1996;144:485-95.
72. Engeland A, Andersen A, Haldorsen T, Tretli S. Smoking habits and risk of cancers other than lung cancer: 28 years' follow-up of 26,000 Norwegian men and women. *Cancer Causes Control.* 1996;7:497-506.
73. Bedwani R, el Khwsky F, Renganathan E, Braga C, Abu Seif HH, Abul Azm T, et al. Epidemiology of bladder cancer in Alexandria, Egypt: tobacco smoking. *Int J Cancer.* 1997;73:64-7.
74. Donato F, Boffetta P, Fazioli R, Aulenti V, Gelatti U, Porru S. Bladder cancer, tobacco smoking, coffee and alcohol drinking in Brescia northern Italy. *Eur J Epidemiol.* 1997;13:795-800.
75. Teschke K, Morgan MS, Checkoway H, Franklin G, Spinelli JJ, Van belle G, et al. Surveillance of nasal and bladder cancer to locate sources of exposure to occupational carcinogens. *Occup Environ Med.* 1997;54:443-51.
76. Sorahan T, Hamilton L, Wallace DM, Bathers S, Gardiner K, Harrington JM. Occupational urothelial tumours: a regional case-control study. *Br J Urol.* 1998;82:25-32.
77. Koivusalo M, Hakulinen T, Vartiainen T, Pukkala E, Jaakkola JJK, Tuomisto J. Drinking water mutagenicity and urinary tract cancers: a population based cancer control study in Finland. *Am J Epidemiol.* 1998;148:704-12.
78. Statistical Office of the European Communities. Social portrait of Europe. Luxembourg: Office for Official Publications of the European Communities, 1996.
79. Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J*

- Cancer. 1999;80:827-41.
80. Vineis P. Tobacco and cancer: an update. *Crit Rev Oncol Hematol*. 1995;18:103-10.
 81. Morrison AS and Cole P. Epidemiology of bladder cancer. *Urol Clin North Am*. 1976;3:13-29.
 82. Vineis P. Epidemiological models of carcinogenesis: the example of bladder cancer. *Cancer Epidemiol Biomarkers Prev*. 1992;1:149-53.
 83. Zhang ZF, Shu XM, Cordon Cardo C, Orloff I, Lu ML, Millon TV, et al. Cigarette smoking and chromosome 9 alterations in bladder cancer. *Cancer Epidemiol Biomarkers Prev*. 1997;6:321-6.
 84. Spruck CHd, Rideout WMd, Olumi AF, Ohnescit PF, Yang AS, Tsai YC, et al. Distinct pattern of p53 mutations in bladder cancer: relationship to tobacco usage. *Cancer Res*. 1993;53:1162-6.
 85. Brockmoller J, Cascorbi I, Kerb R, Roots I. Combined analysis of inherited polymorphisms in arylamine N-acetyltransferase 2, glutathione S-transferases M1 and T1, microsomal epoxide hydrolase, and cytochrome P450 enzymes as modulators of bladder cancer risk. *Cancer Res*. 1996;56:3915-25.
 86. Brockmoller J, Kerb R, Drakouliis N, Staffeldt B, Roots I. Glutathione S-transferase M1 and its variants A and B as host factors of bladder cancer susceptibility: a case-control study. *Cancer Res*. 1994;54:4103-11.
 87. Bartsch H, Malaveille C, Friesen M, Kadlubar FF, Vineis P. Black (air-cured) and blond (flue-cured) tobacco cancer risk. IV: molecular dosimetry studies implicate aromatic amines as bladder carcinogens. *Eur J Cancer*. 1993;29a:1199-207.

4

Elevated Risk of Cancer of the Urinary Tract for Alcohol Drinkers: a Meta-analysis

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ABSTRACT

Objectives: Recent narrative reviews have concluded that there is no support for an association between alcohol consumption and urinary tract cancer. Many individual studies, however, have reported positive associations, although rarely statistically significant. The purpose of this meta-analysis is to summarize and quantify this relationship with more statistical power and to perform a sensitivity analysis on the study characteristics.

Methods: We included 16 epidemiological studies published up to April 1999 and calculated summary odds ratios (SORs), both unadjusted and adjusted for age, sex and smoking by meta-regression analyses.

Results: The age- and smoking-adjusted SORs (current alcohol drinking *vs.* non-drinking) were 1.3 (95%CI 0.9-2.0) for six studies with men and 1.0 (95%CI 0.6-1.7) for four studies with women. The age-, sex- and smoking-adjusted SOR was 1.2 (95% CI 0.9-1.7) for seven studies with men and women combined. Even though studies differed in methodology, the results were rather consistent. Subgroup analyses by type or amount of alcohol were not possible due to sparse data.

Conclusion: We conclude that the available data suggest a slightly increased risk of urinary tract cancer from alcohol consumption for men. The risk related to alcohol consumption for women and the influence of the amount and type of alcohol remains unclear.

Over the last four decades many epidemiological studies have been conducted to investigate determinants of cancer of the urinary tract.^{1,2} These studies suggested that cancer of the urinary tract is influenced by environmental factors, such as cigarette smoking and occupational exposure to aromatic amines, and by chronic infections with *schistosoma haematobium*. The impact of alcohol consumption on the risk of cancer of the urinary tract is less clear. Although some epidemiological studies have reported an elevated risk of cancer of the urinary tract for alcohol drinkers compared to non-drinkers, recent narrative reviews have concluded that there is no support for such an association.^{1,3,6,7} The results of follow-up and case-control studies which have examined this association were rarely statistically significant. However, these non-significant results might also be explained by lack of statistical power in the individual studies. The purpose of the present study was to review the epidemiological literature systematically, by means of a meta-analysis with more statistical power, and to provide quantitative summary estimates of the risk of cancer of the urinary tract with emphasis on current alcohol use *vs.* non-use.

MATERIALS AND METHODS

Search strategy. Epidemiological studies were identified through a computerized Medline and Cancerlit search on follow-up and case-control studies published up to April 1999 using Medical Subjects Headings and free text words. The keywords used were urolo*, bladder, cyst*, vesic*, kidney, glomerul*, nephro*, pyel*, renal, ureteral, urethral, transitional cell, cancer, carcino*, tumor*, neoplasm*, onco*, risk, etiology, epidemiology, and caus*. References in relevant publications were examined further. For inclusion in this analysis, the articles had to provide sufficient information to estimate a summary odds ratio of newly diagnosed primary cancer of the urinary tract for current alcohol consumers compared to non-drinkers, i.e., exposure frequency distribution, exposure specific odds ratios or exposure specific incidence rate ratios. Alcohol consumption is defined as current consumption of beer, wine or spirits at baseline (follow-up studies) or in the reference period (case-control studies).

Data collection. For each study, information was collected on study design (follow-up or case-control study), measuring instrument (interview or questionnaire) and anatomical site of the neoplasm (total urinary tract, bladder, renal pelvis or ureter). For case-control studies, additional information was gathered on the sources of the cases and controls (population-based or hospital-based). Case-control studies which used controls obtained from the general population or from hospitals were defined as population-based or hospital-based case-control studies, respectively. The study characteristics were used to evaluate sources of variation in effect sizes. We extracted data allowing us to calculate both unadjusted and adjusted odds ratios to estimate the relationship between current alcohol consumption and the risk of cancer of the urinary tract. We constructed 2 x 2 tables for each study, based on exposure frequency distributions, in order to calculate the unadjusted odds ratios. The method of Woolf⁸ was used to estimate the variance of the unadjusted odds ratios. Adjusted odds ratios were extracted directly from the original reports. Because we considered age, sex and smoking to be the most important confounding variables, the authors of the original articles had to have adjusted for at least these three variables. If studies reported sex-stratified age- and smoking-adjusted odds ratios, we combined these estimates by calculating an age-, smoking- and sex-adjusted weighted average of the stratum specific odds ratios. The inverse of the standard error was used as weight. For studies that reported separate adjusted odds ratios for several consumption strata, we estimated a total odds ratio for "any use" using the exposure specific prevalences of the non-cases as weight.⁹

Statistical analysis. To detect publication bias, we explored heterogeneity in funnel plots, plots of effect estimates against their estimated precision.¹⁰ The funnel plot is based on the fact that the precision of effect estimates increases with sample size. Results from

Table 1. Study characteristics of published epidemiological studies concerning current alcohol consumption and cancer of the urinary tract, ordered by year of publication

Ref.	First author	Year	Country	Anatomical site of urinary tract	Study design			Alcohol assessment
					follow-up study	case-control study		
						case source	control source	
28	Morgan	1974	Canada	bladder	-	hospital	hospital	questionnaire*
29	Najem	1982	US	bladder	-	hospital	hospital	interview
30-32	MommSEN'	1982-83	Denmark	bladder	-	hospital	population	both techniques
34	Bravo	1987	Spain	urinary tract	-	hospital	hospital	questionnaire*
35	Iscovich	1987	US	bladder	-	hospital	hospital	interview
36	Brownson	1987	US	bladder	-	population	hospital	questionnaire*
46	Risch	1988	Canada	urinary tract	-	hospital	population	interview
37,38	Slatterly'	1983-88	US	bladder	-	population	population	interview
39	Nomura	1989	US	urinary tract	-	hospital	population	interview
40	Ross	1989	US	renal pelvis	-	population	population	interview
47	Mills	1991	US	bladder	yes	-	-	questionnaire*
41	D'Avanzo	1992	Italy	bladder	-	hospital	hospital	interview
42	Chyou	1993	Hawaii	urinary tract	yes	-	-	both techniques
43	Murata	1996	Japan	bladder	yes	-	-	questionnaire*
44	Donato	1997	Italy	bladder	-	hospital	hospital	interview
45	Brummer	1997	US	bladder	-	population	population	interview

*Self-administered questionnaire
'Same study had appeared in more than one publication

small studies will, scatter on the left side of the graph, with the spread narrowing among larger studies on the right side of the graph. In the absence of bias the plot will resemble a symmetrical inverted funnel.¹¹ We examined funnel plot asymmetry visually and measured the degree of asymmetry using Egger's unweighted regression asymmetry test.¹¹ If a study had appeared in more than one publication, data from the last publication were used for statistical analysis. We estimated the summary odds ratios with random effects meta regression analysis using the STATA statistical software package.¹² The between-study variance was estimated iteratively, using the empirical Bayes method.¹³ The summary odds ratios were only calculated for total current alcohol consumption. Subgroup analyses by type or amount of alcohol were not possible due to sparse data. The calculation of summary estimates for men or women combined or separately was based on different study sets. Therefore, we analyzed the results for men and women both separately and combined, depending on available data in the original studies. To explore reasons for the observed heterogeneity we performed sensitivity analyses on study characteristics such as study design, measuring instrument, anatomical site, and source of cases and controls and tested their effects on the relationship between alcohol consumption and cancer of the urinary tract.

RESULTS

Study characteristics. We identified 34 articles on alcohol consumption and the incidence of cancer of the urinary tract published between 1974 and 1997. Usually, the relationship between alcohol consumption and cancer of the urinary tract was not the main research hypothesis. Four studies presented relative risks for ever alcohol consumption with no information on current consumption.¹⁴⁻¹⁷ Ten other articles with mixed results, did not provide sufficient information to estimate a summary odds ratio.¹⁸⁻²⁷ The remaining 20 articles described 16 observational epidemiological studies, which are presented in table 1.²⁸⁻⁴⁷ Most studies were carried out in North America or Europe. We included three follow-up studies,^{34,42,47} six population-based case-control

studies^{30-33,37-40,45,46} and seven hospital-based case-control studies^{28,29,34-36,41,44} in the analyses. The case-control studies also varied with regard to their criteria of case selection. Four case-control studies identified the cancer cases in defined populations,^{33,36-38,40,45} while nine case-control studies selected cases from hospitals.^{28-32,34,35,39,41,44,46} Information on alcohol consumption was obtained by interview,^{29,33,35,37-41,44,46} self-administered questionnaire^{28,34,43,47} or both techniques.^{30-32,42} Some studies included all neoplasms of the urinary tract as cases, of which more than 90% were found to involve bladder cancer,^{34,39,42,46} while others selected only bladder carcinomas^{28-33,35-38,41,43-45,47} or carcinomas of the renal pelvis and ureter.⁴⁰ Most studies used histologically confirmed cases with urothelial cell cancer^{28-33,35-42,44,46,47} (Table 1).

Risk estimation. We could not identify heterogeneity in funnel plots, either visually (Figure 1) or in terms of statistical significance (all p -values ≥ 0.18). Unfortunately, half of the included studies did not provide sufficient information to estimate an adjusted summary odds ratio. The calculation of unadjusted summary odds ratios of cancer of the urinary tract for current alcohol consumers compared to non-drinkers was based on 13 studies. Nine studies allowed calculation of unadjusted odds ratios for men and women combined,^{28,29,33,35,37,39-41,44,45} The odds ratios ranged from 1.0 to 2.4. Some of these studies also presented odds ratios for men and women separately.^{28,39,44,45} The remaining studies only presented sex-specific odds ratios.^{34,36,38,42,43,48} The unadjusted odds ratios for men and women ranged from 0.8 to 2.0 and from 0.5 to 3.1, respectively. The unadjusted summary odds ratio for nine studies combining men and women was 1.4 (95% CI 1.1-1.6). The sex-stratified unadjusted summary odds ratios were 1.2 (95% CI 1.0-1.6) for nine studies with men and 1.1 (95% CI 0.7-1.8) for four studies with women (Figure 2). We did not find a statistically significant interaction between alcohol consumption and sex regarding unadjusted odds ratios ($p=0.77$).

Figure 2 summarizes the adjusted results of observational studies reporting the association between alcohol consumption and the risk of cancer of the urinary tract. Adjusted odds ratios could only be calculated for eight studies, because the exposure specific prevalence of the non-cases or individual age-, sex- and smoking-adjusted odds ratios were not always available.^{29-32,39,42,44-47} Most of the excluded studies did not provide adjusted summary odds ratios with corresponding confidence intervals for current drinkers compared to non-current drinkers. Only one study was excluded because of missing information on the distribution of controls.²⁰ Frequently used confounders, in addition to age, sex and smoking, were residence^{29-32,39,42,45} and race.^{29,39} The adjusted odds ratios of the individual studies ranged from 1.0 to 2.2 and from 0.5 to 3.4 for men and women, respectively. Seven studies contributed to the estimation of a summary odds ratio for men and women combined.^{29,31,39,44-47} The adjusted odds ratios ranged from 0.8 to 2.6. The age- and smoking-adjusted summary odds ratios for current alcohol consumption were 1.3 (95% CI 0.9-2.0) for six studies with men and 1.0 (95% CI 0.6-1.7) for four studies with women. For seven studies with men and women combined, the age-, sex- and smoking-adjusted summary odds ratio was 1.2 (95%-CI 0.9-1.7) (Figure 2). Sex did not seem to be a statistically significant effect modifier for the adjusted (age and smoking) association between alcohol consumption and cancer of the urinary tract ($p=0.42$).

Sensitivity analysis. We further examined the crude relationship of alcohol consumption and cancer of the urinary tract by study design, measuring instrument, anatomical site and sources of cases and controls (only for case-control studies) to explore their influence on the outcome estimates (Figure 3). All tests for interaction were statistically non-significant. The summary odds ratios for alcohol consumption and cancer of the urinary tract were 1.3 (95% CI 0.9-2.0) for follow-up studies and 1.2 (95% CI 1.0-1.4) for case-control studies. The summary odds ratio for studies that used self-administered questionnaires to obtain information on alcohol consumption was 1.1 (95% CI 0.7-1.8). Studies that used interview techniques had a summary odds ratio of 1.3 (95% CI 0.8-2.2). Summary odds ratios were calculated for studies classified according to the anatomical site of the tumor. The summary odds ratio for the relationship between

Figure 1. Funnel plot for current alcohol drinkers versus non-drinkers, unadjusted. * Interrupted and uninterrupted reference lines indicates no effect and total summary odds ratio, respectively

alcohol consumption and cancer of the total urinary tract was 1.0 (95% CI 0.8-1.3). For studies that selected only bladder carcinomas, the summary odds ratio was 1.3 (95% CI 1.1-1.5). Case-control studies that selected cases or controls from defined populations had summary odds ratios of 1.2 (95% CI 0.9-1.6) and 1.2 (95% CI 0.9-1.7), respectively. The summary odds ratios for hospital-based case-control studies were 1.2 (95% CI 0.9-1.5) for studies that selected cases from hospitals and 1.2 (95% CI 0.9-1.5) for studies selected controls from hospitals.

DISCUSSION

We were able to retrieve 30 articles reporting epidemiological studies on current alcohol consumption and human cancer of the urinary tract. These primary studies can be considered as the best available evidence. Our meta-analysis revealed an approximately 30% elevated risk of cancer of the urinary tract for male alcohol drinkers compared to non-drinkers, although not statistically significant. The risk of cancer of the urinary tract for women in relation to alcohol consumption remains unclear. We expect, however, a similar pattern if more women were included in the original studies. We did not attempt to uncover unpublished observations and could not include studies with insufficient information to estimate a summary odds ratio. The excluded studies showed mixed results. The estimated effects of alcohol consumption ranged from protective,^{15,17,24} through no effect^{16,25-27} to harmful.^{14,18-20,23} Three studies did not indicate a direction of the estimated effect.^{22,49,50} Although publication bias might arise by excluding these studies, we could not identify it in our meta-analysis, neither visually nor in terms of statistical significance. The definition of “current drinker” at baseline (follow-up studies) or in the reference period (case-control studies) might have caused heterogeneity between studies, because the follow-up period and the reference date

Figure 2. Adjusted (age, sex, smoking) odds ratios and 95% confidence intervals of cancer of the urinary tract for current alcohol consumers compared to non-drinkers, published in epidemiological studies. * Same study had appeared in more than one publication; reference line indicates no effect, box sizes are proportional to weight study

varied between the included studies. Therefore, results have to be interpreted with caution. Because of potential additional heterogeneity in populations, designs and analyses of various studies, we assumed that the true effects being estimated would vary between the studies in addition to the usual sampling variation in the estimates (within studies). To account for both sources of variation we used random effects meta regression analysis to combine the results from the primary studies.¹³ The random effect approach provides some allowance for heterogeneity in studies beyond sampling error. The summary odds ratios were similar across study designs and source of the cases and controls in case-control studies. Although we could not find statistically significant interactions, it appears that the weak elevated risk of cancer for alcohol consumers is

Figure 3. Crude summary odds ratios and 95% confidence intervals of cancer of the urinary tract for current alcohol consumers compared to non-drinkers by study design, measuring instrument, tumor site and source of cases and controls for nine studies with men. * Only for case-control studies; interrupted and uninterrupted reference lines indicates no effect and total summary odds ratio respectively

mainly confined to cancer of the urinary bladder. The summary odds ratio was higher for studies that used interviewing techniques than for studies that used self-administered questionnaires. This contrast can be a consequence of response bias due to different assessment techniques or to chance alone.

Although 16 studies contributed to this meta-analysis, only eight provided sufficient information to calculate odds ratios, adjusted for at least age, smoking and sex. It was not possible to adjust for a specific set of confounders. Because of the unavailability of adjusted odds ratios in some of the included studies, we also calculated unadjusted odds ratios. Focussing entirely on adjusted odds ratios would have lead to the exclusion of 50% of the studies which also have (crude) information on the association between alcohol consumption and cancer of the urinary tract.

Some authors suggested that residual confounding due to tobacco smoking could explain an increased risk for alcohol drinking found in some studies.^{7,44} The adjusted and unadjusted estimates, however, were comparable. Furthermore, a small increased risk was also found among those who stopped smoking⁴⁴ and those who claimed to have never smoked.^{34,37,44} Another explanation for an association between alcohol consumption and cancer of the urinary tract might be that non-drinkers are a rather selected population. In the European Union 89% of the male population drinks alcohol.⁵¹ Non-drinkers may differ also in occupation; the consumption of coffee, tea, vegetables or fruit; and other lifestyle habits from the general population of drinkers. Age and smoking are currently known to be the most important risk factors for cancer of the urinary tract. However, it remains possible that confounding explains the weak association found in this meta-analysis. consumption of coffee, tea, vegetables or fruit; and other lifestyle habits from the general population of drinkers. Age and smoking are currently known to be the most important risk factors for cancer of the urinary tract. However, it remains possible that confounding explains the weak association found in this meta-analysis. consumption of coffee, tea, vegetables or fruit; and other lifestyle habits from the general population of drinkers. Age and smoking are currently known to be the most important risk factors for cancer of the urinary tract. However, it remains possible that confounding explains the weak association found in this meta-analysis.

The remaining articles provided data within different exposure categories.^{33-37,39,40,42-45} The content of these categories, however, differed substantially between the studies. In this meta-analysis we therefore compared current alcohol consumption with no alcohol consumption. The associations between the consumption of specific alcoholic beverages (beer, wine or spirits) and the risk cancer of the urinary tract were reported in nine studies with no consistent results.^{14,20,24,33,35,40-42,46}

We conclude that current alcohol consumption slightly increases the risk of male cancer of the urinary tract with approximately 30%. The risk of cancer of the urinary tract related to alcohol consumption for women and the influence of the amount and type of alcohol remains unclear.

REFERENCES

1. Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am.* 1992;6:1-30.
2. Longnecker MP. Alcohol consumption and risk of cancer in humans: an overview. *Alcohol.* 1995;12:87-96.
3. Longnecker MP, Enger SM. Epidemiologic data on alcoholic beverage consumption and risk of cancer. *Clin Chim Acta.* 1996;246:121-41.
4. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol.* 1997;13:291-8.
5. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol.* 1996;23:536-45.
6. World Cancer Research Fund, American Institute for Cancer Research. In: *Food, Nutrition and the Prevention of Cancer: a global perspective.* Menasha: Banta Book Group, 1997.

7. Silverman DT, Morrison AS, Devesa SS. In: Schottenfeld D, Fraumeni JF. Schottenfeld D, Fraumeni JFs. Cancer epidemiology and prevention. New York Oxford: Oxford university press, 1996.
8. Woolf B. On estimation the relation between blood group and disease. *Ann Hum Genet.* 1954;19:251-3.
9. Tan FES, Zeegers MPA: An alternative method of combining exposure specific odds ratios in epidemiologic studies. In Research Report. Maastricht: Maastricht University, Department Methodology and Statistics, 1999.
10. Light RJ, Pillemer DB. In: Summing up: the science of reviewing research. Harvard: Harvard University Press, 1984.
11. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *Br Med J.* 1997;315:629-34.
12. StataCorp: Stata Statistical Software: Release 6.0. College Station, Texas: Stata Corporation, 1999
13. Berkey CS, Hoaglin DC, Mosteller F, Colditz GA. A random-effects regression model for meta-analysis. *Stat Med.* 1995;14:395-411.
14. Jensen OM, Wahrendorf J, Knudsen JB, Sorensen BL. The Copenhagen case-control study of bladder cancer. II. Effect of coffee and other beverages. *Int J Cancer.* 1986;37:651-7.
15. Ohno Y, Aoki K, Obata K, Morrison AS. Case-control study of urinary bladder cancer in metropolitan Nagoya. *Natl Cancer Inst Monogr.* 1985;69:229-34.
16. Cartwright RA, Adib R, Glashan R, Gray BK. The epidemiology of bladder cancer in west yorkshire: a preliminary report on non-occupational aetiologies. *Carcinogenesis.* 1981;2:343-7.
17. Howe GR, Burch JD, Miller AB, Cook GM, Esteve J, Morrison B, et al. Tobacco use, occupation, coffee, various nutrients, and bladder cancer. *J Natl Cancer Inst.* 1980;64:701-13.
18. Hakulinen T, Lehtimäki L, Lehtonen M, Teppo L. Cancer morbidity among two male cohorts with increased alcohol consumption in Finland. *J Natl Cancer Inst.* 1974;52:1711-4.
19. Akdas A, Kirkali Z, Bilir N. Epidemiological case-control study on the etiology of bladder cancer in Turkey. *Eur Urol.* 1990;17:23-6.
20. Kunze E, Claude J, Frentzel Beyme R, Paczkowski K, Schneider G, Schubert H. Association of cancer of the lower urinary tract with consumption of alcoholic beverages. A case-control study. *Carcinogenesis.* 1986;7:163-5.
21. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in new orleans, Louisiana. *J Natl Cancer Inst.* 1968;41:683-709.
22. Yu Y, Hu J, Wang PP, Zou Y, Qi Y, Zhao P, et al. Risk factors for bladder cancer: a case-control study in northeast China. *Eur J Cancer Prev.* 1997;6:363-9.
23. Momas I, Daures JP, Festy B, Bontoux J, Gremy F. Relative importance of risk factors in bladder carcinogenesis: some new results about Mediterranean habits. *Cancer Causes Control.* 1994;5:326-32.
24. Williams RR, Horm JW. Association of cancer sites with tobacco and alcohol consumption and socioeconomic status of patients: interview study from the third national cancer survey. *J Natl Cancer Inst.* 1977;58:525-47.
25. La Vecchia C, Negri E, Decarli A, B DA, Liberati C, Franceschi S. Dietary factors in the risk of bladder cancer. *Nutr Cancer.* 1989;12:93-101.
26. Wynder EL, Dieck GS, Hall NE, Lahti H. A case-control study of diesel exhaust exposure and bladder cancer. *Environ Res.* 1985;37:475-89.
27. Wynder EL, Goldsmith R. The epidemiology of bladder cancer: a second look. *Cancer.* 1977;40:1246-68.
28. Morgan RW, Meera GJ. Bladder cancer: smoking, beverages and artificial sweeteners. *Can Med Assoc J.* 1974;111:1067-70.
29. Najem GR, Louria DB, Seebode JJ, Thind IS, Prusakowski JM, Ambrose RB, et al. Life time occupation, smoking, caffeine, saccharine, hair dyes and bladder carcinogenesis. *Int J Epidemiol.* 1982;11:212-7.
30. Mommensen S, Aagaard J, Sell A. An epidemiological case-control study of bladder cancer in males from a predominantly rural district. *Eur J Cancer Clin Oncol.* 1982;18:1205-10.
31. Mommensen S, Aagaard J. A case-control study of bladder-cancer: a multivariate, stratified analysis of a low-risk population. *Dan Med Bull.* 1983;30:427-32.
32. Mommensen S, Aagaard J, Sell A. An epidemiological study of bladder cancer in a predominantly rural district. *Scand J Urol Nephrol.* 1983;17:307-12.
33. Thomas DB, Uhl CN, Hartge P. Bladder cancer and alcoholic beverage consumption. *Am J Epidemiol.* 1983;118:720-7.
34. Bravo MP, Del Rey Calero J, Conde M. Bladder cancer and the consumption of alcoholic beverages in Spain. *Eur J Epidemiol.* 1987;3:365-9.
35. Iscovich J, Castelletto R, Esteve J, Munoz N, Colanzi R, Coronel A, et al. Tobacco smoking, occupational exposure and bladder cancer in Argentina. *Int J Cancer.* 1987;40:734-40.
36. Brownson RC, Chang JC, Davis JR. Occupation, smoking, and alcohol in the epidemiology of bladder cancer. *Am J Public Health.* 1987;77:1298-300.
37. Slattery ML, West DW, Robison LM. Fluid intake and bladder cancer in Utah. *Int J Cancer.* 1988;42:17-22.
38. Slattery ML, Schumacher MC, West DW, Robison LM. Smoking and bladder cancer. The modifying effect of cigarettes on other factors. *Cancer.* 1988;61:402-8.
39. Nomura A, Kolonel LN, Yoshizawa CN. Smoking, alcohol, occupation, and hair dye use in cancer of the lower urinary tract. *Am J Epidemiol.* 1989;130:1159-63.
40. Ross RK, Paganini Hill A, Landolph J, Gerkins V, Henderson BE. Analgesics, cigarette smoking, and other risk factors for cancer of the renal pelvis and ureter. *Cancer Res.* 1989;49:1045-8.
41. D'Avanzo B, La Vecchia C, Franceschi S, Negri E, Talamini R, Buttino I. Coffee consumption and bladder cancer risk. *Eur J Cancer.* 1992;28a:1480-4.
42. Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol.* 1993;3:211-6.
43. Murata M, Takayama K, Choi BC, Pak AW. A nested case-control study on alcohol drinking, tobacco smoking, and cancer. *Cancer Detect Prev.* 1996;20:557-65.
44. Donato F, Boffetta P, Fazioli R, Aulenti V, Gelatti U, Porru S. Bladder cancer, tobacco smoking, coffee and alcohol drinking in Brescia northern Italy. *Eur J Epidemiol.* 1997;13:795-800.

45. Bruemmer B, White E, Vaughan TL, Cheney CL. Fluid intake and the incidence of bladder cancer among middle-aged men and women in a three-county area of western Washington. *Nutr Cancer*. 1997;29:163-8.
46. Risch HA, Burch JD, Miller AB, Hill GB, Steele R, Howe GR. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol*. 1988;127:1179-91.
47. Mills PK, Beeson WL, Phillips RL, Fraser GE. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol*. 1991;133:230-9.
48. Adami HO, McLaughlin JK, Hsing AW, Wolk A, Ekblom A, Holmberg L, et al. Alcoholism and cancer risk: a population-based cohort study. *Cancer Cause Control*. 1992;3:419-25.
49. Block G, Patterson B, Subar A. Fruit, vegetables, and cancer prevention: a review of the epidemiological evidence. *Nutr Cancer*. 1992;18:1-29.
50. Sullivan JW. Epidemiologic survey of bladder cancer in greater New Orleans. *J Urol*. 1982;128:281-3.
51. Hupkens CLH, Knibbe RA, Drop MJ. Alcohol consumption in the European Community: uniformity and diversity in drinking patterns. *Addiction*. 1993;88:1391-404.

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Are Coffee and Tea Consumption associated with Urinary Tract Cancer Risk? A Systematic Review and Meta-analysis

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ABSTRACT

Background: Narrative reviews have concluded that there is a small association between coffee consumption and an increased risk of urinary tract cancer, possibly due to confounding by smoking. No association for tea consumption has been indicated. This systematic review attempts to summarize and quantify these associations both unadjusted and adjusted for age, smoking and sex.

Methods: Thirty-four case-control and three follow-up studies were included in this systematic review. Summary odds ratios (OR) were calculated by meta-regression analyses.

Results: The unadjusted summary OR indicated a small increased risk of urinary tract cancer for current coffee consumers versus non-drinkers. The adjusted summary OR were: 1.26 (95% CI: 1.09-1.46) for studies with only men, 1.08 (95% CI: 0.79-1.46) for studies with only women and 1.18 (95% CI: 1.01-1.38) for studies with men and women combined. Neither unadjusted nor adjusted summary OR provided evidence for a positive association between tea consumption and urinary tract cancer. Even though studies differed in methodology, the results were rather consistent. We did not perform dose-response analyses for coffee and tea consumption due to sparse data.

Conclusions: In accordance with earlier reviews, we found that coffee consumption increases the risk of urinary tract cancer by approximately 20%. The consumption of tea seems not to be related to an increased risk of urinary tract cancer.

Over the last four decades, a number of epidemiological studies have been conducted to investigate determinants of urinary tract cancer.¹⁻⁵ These studies suggested that bladder cancer is influenced by environmental factors, such as cigarette smoking and exposure to industrial chemicals (e.g., aromatic amines), and by chronic infections with *Schistosoma Haematobium*. The impact of coffee and tea consumption on the risk of urinary tract cancer is less clear, although many case-control studies and several follow-up studies on these associations have been published. Because coffee and tea are two of the most popular beverages consumed worldwide, the relationship between coffee and tea consumption and the incidence of bladder cancer is an important concern. Earlier narrative reviews on coffee consumption concluded that the evidence was consistent with a small positive association.^{4,6-8} Viscoli *et al.*⁹ quantified this association in 1993 in a meta-analysis based on seven studies and concluded that the best available evidence does not suggest a clinically important association between coffee consumption and cancer of the lower urinary tract. According to four narrative reviews, there is no positive association between tea consumption and urinary tract cancer, although a summary association has never been quantified.^{4,6,10,11} The present study aims at reviewing all epidemiological studies up to December 2000 more systematically, to provide quantitative summary estimates of the risk of urinary tract cancer associated with both coffee and tea consumption and to evaluate changes in summary estimates after adjustment for cigarette smoking.

MATERIALS AND METHODS

Search strategy. The study design has been published previously.¹² Epidemiological studies were identified by the principal investigator through a computerized Medline, Cancerlit and Current Contents search on follow-up and case-control studies of all languages published until December 2000 using Medical Subjects Headings and free text words. The keywords used were urolo*, bladder, cyst*, vesic*, kidney, glomerul*, nephro*, pyel*, renal, ureteral, urethral, transitional cell, cancer, carcino*, tumor*, neoplasm*, onco*, risk, etiology, epidemiology, and caus*. Citation tracking was performed until no new studies were found. For inclusion in this analysis, the articles had to provide sufficient information to estimate a summary odds ratio (OR) and the associated standard error of incident primary urinary tract cancer for coffee or tea consumers compared to non-drinkers, i.e. exposure frequency distribution, exposure-specific OR or exposure-specific incidence rate ratios. Coffee and tea consumption were defined as ever consumption or current consumption at baseline (for follow-up studies) or in the reference period (for case-control studies).

Data collection. A criteria list for the assessment of quality items (study characteristics) in observational cancer research was developed. The criteria list was used to provide covariables for meta-regression to explore reasons for observed heterogeneity in results and is available upon request. The criteria list was developed through consensus meetings with experts in the fields of cancer and meta-analysis. The list comprised items felt to be important for the quality of observational research, including study design (follow-up or case-control study), measuring instrument (interview or self-administered questionnaire) and anatomical site of the neoplasm (total urinary tract, bladder, renal pelvis or ureter). For case-control studies, additional information was gathered on the sources of the cases and controls. Case-control studies that used controls obtained from the general population or from hospitals were defined as population-based or hospital-based case-control studies, respectively. If case-control studies used both population- and hospital-based controls, population-based controls were used for further analysis. Data were extracted to permit the calculation of both unadjusted and adjusted OR to estimate the association between coffee and tea consumption and the risk of urinary tract cancer. Two-way contingency tables for each study were constructed, based on exposure frequency distributions, in order to calculate the unadjusted OR. Adjusted OR were extracted directly from the original reports. Because we considered age, smoking

and sex to be the most important confounding variables, the authors of the original articles had to have adjusted for at least these three variables for inclusion in the calculation of adjusted summary estimates. If studies reported sex stratified age- and smoking-adjusted OR, we combined these estimates by calculating age-, smoking- and sex-adjusted OR.¹³ In such calculations, the inverse of the variance was used to weight the reported sex-stratified estimates. For studies that reported separate adjusted OR for several consumption strata, we estimated a total OR for current or ever use weighted by the exposure-specific prevalence of the non-cases¹⁴.

Statistical analysis. To detect publication or related biases, we explored asymmetry in funnel plots, i.e., plots of effect estimates against their estimated precision.¹⁵ This is examined visually and the degree of asymmetry is measured using Egger's unweighted regression asymmetry test.^{16,17} If a study appeared in more than one publication, data from the last publication were used for statistical analysis. We estimated the summary OR and corresponding 95% confidence intervals (CI) with random effects meta regression analysis using the Stata statistical software.^{18,19} The between-study variance was estimated iteratively, using the empirical Bayes method.^{19,20} Summary OR were calculated for total coffee consumption (current drinker versus non-drinker and ever drinker versus never-drinker), decaffeinated coffee (drinker versus non-drinker) and total tea consumption (current drinker versus non-drinker and ever drinker versus never-drinker). We did not perform subgroup analyses by beverage type other than decaffeinated coffee or by the number of cups consumed per day due to sparse data. Data were analysed separately or combined for men and women, depending on data availability in the original studies. Furthermore, we evaluated the influence of the type of adjustment for cigarette smoking in the component studies on the summary association between current coffee and tea consumption and bladder cancer risk. This analysis might give some insight in the existence of residual confounding from inadequate adjustment of cigarette smoking in the component studies. To explore reasons for observed heterogeneity, sensitivity analyses were performed on the study characteristics measuring instrument, anatomical site of the neoplasm and source of cases and controls, and their influence on the association between coffee or tea consumption and urinary tract cancer was tested.

Study characteristics. We identified 69 articles reporting follow-up or case-control studies on coffee and tea consumption and urinary tract cancer published between 1968 and 1999.²¹⁻⁸⁹ Generally, the association between coffee or tea consumption and urinary tract cancer was not the main research hypothesis. Twenty articles were excluded because they did not provide sufficient information to estimate a summary OR and corresponding standard error^{21-29,33-39} or did not use incident cases.^{30-32,87} The remaining 49 articles described three follow-up studies^{40,41,84,88} and 34 case-control studies with incident cases.^{42-83,85,86,89} One case-control study that provided separate associations for parts of the study performed in the US, the UK and Japan was considered as three separate studies.⁴⁵ The case-control studies were population based (n=12),^{42-56,83,89} hospital based (n=18)^{57-78,85,86} or neighbourhood based (n=3).⁷⁹⁻⁸¹ One case-control study used both population- and hospital-based controls⁸². The hospital controls in most case-control studies did not have any diet- or smoking-related disease. The case-control studies also varied with regard to their criteria of case selection. Seven case-control studies identified cancer cases in defined populations.^{44,46,47,49,51,52,55,56,79,80, 83,89} 22 Case-control studies selected cases from hospitals^{42,43,45,53,58-63,65-78,81,82,85,86} and five case-control studies used both sources.^{48,50, 54, 57,64} Information on coffee and tea consumption was obtained by interview,^{40,42-58,61,62,64-83,86,89} self-administered questionnaire^{41,59,60,85,88} or both techniques.^{63,84} Some studies included all neoplasms of the urinary tract as cases, of which more than 90% were found to involve bladder cancer,^{42,45,48,53,60,73,84,85} while others selected only bladder carcinomas,^{40,41,44,46,47,49-52,54-59,62-72,74-79,81-83,86,88} carcinomas of the renal pelvis^{61,89} or carcinomas of renal pelvis and ureter combined.⁸⁰ Most studies used histologically confirmed cases with urothelial cell cancer (Table 1).

RESULTS

Table 1. Study characteristics of published epidemiological studies concerning coffee and tea consumption and cancer of the urinary tract, ordered by year of publication

Ref.	First author	Year	Country	Anatomical site of urinary tract	Study design follow-up study	Case-control study		Coffee/Tea assessment
						Case source	control source	
57	Dunham	1968	US	bladder	-	Both	hospital	interview
42	Cole*	1971	US	urinary tract	-	hospital	population	interview
58	Bross	1973	US	bladder	-	hospital	hospital	interview
59	Morgan	1974	Canada	bladder	-	hospital	hospital	questionnaire
43	Schmauz*	1974	US	urinary tract	-	hospital	population	interview
60	Simon	1975	US	urinary tract	-	hospital	hospital	questionnaire
61	Armstrong	1976	UK	renal pelvis	-	hospital	hospital	interview
44	Miller†	1977	Canada	bladder	-	population	population	interview
62	Wynder§	1977	US	bladder	-	hospital	hospital	interview
63	Metlin	1979	US	bladder	-	hospital	hospital	both
79	Howe†	1980	Canada	bladder	-	population	neighbor	interview
64	Cartwright	1981	UK	bladder	-	Both	hospital	interview
45	Morrison	1982	US	urinary tract	-	hospital	population	interview
45	Morrison	1982	UK	urinary tract	-	hospital	population	interview
45	Morrison	1982	Japan	urinary tract	-	hospital	population	interview
65	Najem	1982	US	bladder	-	hospital	hospital	interview
46	Hartge†	1983	US	bladder	-	population	population	interview
47	Marrett†	1983	US	bladder	-	population	population	interview
89	Mc Laughlin	1983	US	renal pelvis	-	population	population	interview
48	Ohno	1985	Japan	urinary tract	-	Both	population	interview
66	Rebekalos	1985	Greece	bladder	-	hospital	hospital	interview
85	Bravo	1986	Spain	urinary tract	-	hospital	hospital	questionnaire
40	Heilbrun†	1986	US	bladder	yes	-	-	interview
49	Jensen	1986	Denmark	bladder	-	population	population	interview
67	Kabat§	1986	US	bladder	-	hospital	hospital	interview
83	Piper	1986	US	bladder	-	population	population	interview
68	Iscovich	1987	US	bladder	-	hospital	hospital	interview
69	Ciccone	1988	Italy	bladder	-	hospital	hospital	interview
50	Risch	1988	Canada	bladder	-	Both	population	interview
52	Slattery†	1988	US	bladder	-	population	population	interview
51	Slattery†	1988	US	bladder	-	population	population	interview
70	LaVecchia**	1989	Italy	bladder	-	hospital	hospital	interview
80	Ross	1989	US	renal pelvis††	-	population	neighbor	interview
71	Clavel	1991	France	bladder	-	hospital	hospital	interview
41	Mills	1991	US	bladder	yes	-	-	questionnaire
53	Nomura	1991	US	urinary tract	-	hospital	population	interview
72	d'Avanzo**	1992	Italy	bladder	-	hospital	hospital	interview
73	Kunze	1992	Germany	urinary tract	-	hospital	hospital	interview
74	LaVecchia**	1992	Italy	bladder	-	hospital	hospital	interview
84	Chyou†	1993	US	urinary tract	yes	-	-	both
82	Pujolar	1993	Spain	bladder	-	hospital	both	interview
81	Vena	1993	US	bladder	-	hospital	neighbor	interview
75	Barbone**	1994	Italy	bladder	-	hospital	hospital	interview
76	Hours	1994	France	bladder	-	hospital	hospital	interview
54	Momas	1994	France	bladder	-	Both	population	interview
55	Sturgeon†	1994	US	bladder	-	population	population	interview
77	d'Avanzo**	1995	Italy	bladder	-	hospital	hospital	interview
56	Bruemmer	1997	US	bladder	-	population	population	interview
78	Donato††	1997	Italy	bladder	-	hospital	hospital	interview
86	Donato††	1998	Italy	bladder	-	hospital	hospital	interview
88	Michaud	1999	US	bladder	yes	-	-	questionnaire

*,†,§,||,||| same study has appeared in more than one publication

†† and ureter

Risk estimation. Funnel plots showed no asymmetry, neither visually nor in terms of statistical significance (p -values ≥ 0.51 and ≥ 0.12 for current coffee and tea consumption, respectively)(Figure 1).

Table 2 summarizes the unadjusted and adjusted results of observational studies reporting the association between coffee and tea consumption. Almost all included articles provided some information to estimate an association between coffee consumption and urinary tract cancer.^{41,73,75,86,88,89} The study-specific effect estimates are available upon request. The unadjusted summary OR for current coffee drinkers compared to non-drinkers were 1.30 (95% CI: 1.15-1.47) for 25 studies with only men, 1.18 (95% CI: 0.96-1.44) for 23 studies with only women and 1.26 (95% CI: 1.11-1.43) for 25 studies with men and women combined. The adjusted summary OR for current coffee consumption were somewhat lower after adjustment for age, smoking and sex. The adjusted summary OR for current coffee consumption were 1.26 (95% CI: 1.09-1.46), 1.08 (95% CI: 0.79-1.46) and 1.18 (95% CI: 1.01-1.38) for 16 studies with men, 12 studies with women and 14 studies with men and women combined. The adjusted summary associations between ever versus never coffee consumption and urinary tract cancer also showed small increased risks, although the unadjusted summary estimates were higher. The consumption of decaffeinated coffee also appeared to be associated with bladder cancer risk, although not statistically significant (Table 2).

Twenty articles provided sufficient information to estimate a summary association of tea consumption.^{40,46,48-53,56,59,60,68,71-74,79,84,88,89} Neither the unadjusted nor the age-, smoking-, and sex-adjusted summary associations provided evidence for a positive association between current tea consumption and the risk of urinary tract cancer (Table 2). The sex-specific adjusted summary OR for current tea consumers compared to non-drinkers were 1.08 (95% CI: 0.94-1.24) for seven studies with men and 0.99 (95% CI: 0.81-1.20) for six studies with women. Seven studies allowed calculation of adjusted summary OR for men and women combined, 1.01 (95% CI: 0.92-1.10). Very few studies reported an effect estimate for ever versus never tea consumption. Both unadjusted and adjusted summary associations for ever tea consumption did not indicate a positive association between tea consumption and bladder cancer risk. All summary associations were around unity (Table 2).

Confounding by cigarette smoking. Seventeen studies presented adjusted OR for current coffee and tea consumption for men and women combined. Of these, seven studies adjusted for cigarette smoking by incorporating one smoking variable (mostly smoking status) in a regression model.^{48,56,62,65,69,77,82} Seven other studies used a combination of cigarette smoking features (mostly amount and duration) to adjust for cigarette smoking.^{49,50,52,53,55,73,78} The type of adjustment for three studies remains unclear.^{41,66,74} The summary OR for current coffee consumption compared to non-drinkers was larger in studies in which several cigarette smoking features were used to adjust for cigarette smoking compared to studies in which only one smoking variable was used (Table 3). For tea consumption, the type of adjustment for cigarette smoking did not influence the summary associations substantially (Table 3).

Sensitivity analysis. We further examined the crude association of current coffee and tea consumption and urinary tract cancer by measuring instrument, sources of cases and controls and anatomical site of the tumour to explore their influence on the outcome estimates in case-control studies that provided information for men and women combined (Figure 2). Unfortunately, none of the follow-up studies provided sufficient information for men and women combined to contribute to this sensitivity analysis. All tests for multiplicative interaction were statistically non-significant. Most subset-specific summary OR did not differ substantially. Neither selection on anatomical site of the tumour nor selection on the source of cases and controls altered the summary OR. It appeared, however, that the summary associations for studies that used interviewing techniques were higher than for studies that used self-administered questionnaires (Figure 2).

Table 2. Summary odds ratios (OR) for coffee and tea consumption both unadjusted and adjusted for age, smoking and sex calculated from epidemiological studies

Coffee/Tea consumption	Male			Female			Male and female		
	n*	OR (95% CI)	Q† (p)	n*	OR (95% CI)	Q† (p)	n*	OR (95% CI)	Q† (p)
Unadjusted summary odds ratios									
Coffee consumption									
current versus non-drinker	25	1.30 (1.15-1.47)	45.94 (<0.01)	23	1.18 (0.96-1.44)	35.00 (0.04)	25	1.26 (1.11-1.43)	56.36 (<0.01)
ever versus never drinker	7	1.92 (1.37-2.70)	13.57 (0.04)	6	1.66 (0.81-3.38)	15.47 (<0.01)	7	1.85 (1.21-2.82)	22.43 (<0.01)
Decaffeinated coffee consumption									
drinker versus non-drinker	4	1.08 (0.88-1.32)	3.79 (0.29)	2	0.56 (0.35-0.91)	0.01 (0.91)	4	1.01 (0.74-1.37)	6.52 (0.09)
Tea consumption									
current versus non-drinker	8	1.04 (0.89-1.21)	10.11 (0.18)	8	0.97 (0.78-1.20)	9.17 (0.24)	9	0.94 (0.82-1.08)	14.59 (0.07)
ever versus never drinker	3	0.94 (0.66-1.35)	3.69 (0.16)	2	1.36 (0.18-10.11)	14.90 (<0.01)	3	0.77 (0.47-1.25)	7.16 (0.03)
Adjusted summary odds ratios									
Coffee consumption									
current versus non-drinker	16	1.26 (1.09-1.46)	40.15 (<0.01)	12	1.08 (0.79-1.46)	26.68 (<0.01)	14	1.18 (1.01-1.38)	30.03 (<0.01)
ever versus never drinker	6	1.19 (1.00-1.41)	6.03 (0.30)	5	1.16 (0.86-1.56)	4.25 (0.37)	5	1.18 (1.03-1.36)	1.06 (0.90)
Decaffeinated coffee consumption									
drinker versus non-drinker	5	1.04 (0.86-1.25)	4.47 (0.35)	3	1.18 (0.63-2.22)	5.76 (0.06)	4	1.18 (0.99-1.40)	2.40 (0.49)
Tea consumption									
current versus non-drinker	7	1.08 (0.94-1.24)	9.07 (0.17)	6	0.99 (0.81-1.20)	6.96 (0.22)	7	1.01 (0.92-1.10)	5.47 (0.49)
ever versus never drinker	4	1.02 (0.84-1.25)	2.97 (0.40)	3	1.00 (0.27-3.73)	13.81 (<0.01)	3	1.09 (0.54-2.17)	9.60 (0.15)

*number of analysed studies
†X²-test for heterogeneity

Table 3. Summary odds ratios (OR) for current coffee and tea consumption compared to non-drinkers according to the type of adjustment for cigarette smoking made in the component studies, for men and women combined

Coffee/tea consumption	Adjusted for one cigarette smoking variable*			Adjusted for several cigarette smoking features†		
	n‡	OR (95% CI)	Q† (p)	n‡	OR (95% CI)	Q† (p)
Coffee consumption						
current versus non-drinker	7	1.04 (0.89-1.22)	8.90 (0.18)	5	1.36 (1.08-2.01)	11.74 (0.02)
Tea consumption						
current versus non-drinker	1	1.02 (0.77-1.33)	N/A	5	1.09 (0.96-1.25)	3.25 (0.52)

*adjusted for cigarette smoking status or amount only
†adjusted for any combination of cigarette smoking status, amount or duration
‡number of analysed studies
§X²-test for heterogeneity

DISCUSSION

We were able to retrieve 49 articles reporting follow-up or case-control studies on coffee and tea consumption and the risk of human urinary tract cancer in the present meta-analysis. These observational studies can be considered as the best available epidemiological evidence, because no human experimental studies have been conducted. The available evidence suggests a small increased risk for coffee consumers remaining after adjustment for age, smoking and sex. The consumption of tea seems not to be related to an increased risk of urinary tract cancer.

We did not attempt to uncover unpublished observations and excluded studies that did not meet the predetermined criteria. The excluded studies showed inconsistent results for coffee consumption. The estimated associations of coffee consumption ranged from moderately inverse,^{27,32,38} through no association^{24-26,28,29,31,90} to moderately positive.^{22,23,30,33-37,39,91} Although three of the excluded studies on tea consumption have presented positive associations,^{31,37} most studies could not confirm these findings.^{21,23,27,33,35,90} Publication bias might arise by excluding these studies. However, we did not identify funnel plot asymmetry in our meta-analysis, either visually or in terms of statistical significance.

The definition of 'current drinker' at baseline (follow-up studies) or in the reference period (case-control studies) might have caused heterogeneity between studies, because the follow-up period and the reference date varied between the included studies. Therefore, results have to be interpreted with caution. Because of potential additional heterogeneity in populations, designs and analyses of various studies, we assumed that the true effects being estimated would vary between the studies in addition to the usual sampling variation in the estimates (within studies) even if the test for heterogeneity could not identify heterogeneity. To account for both sources of variation we used a random effects meta regression analysis to combine the results from the primary studies.²⁰ The random effect approach provides some allowance for heterogeneity in studies beyond sampling error.

The summary OR were similar across sources of the cases and controls in the case-control studies. It appeared that the anatomical site of the tumour did not modify the association between coffee/tea consumption and urinary tract cancer risk. Because the overwhelming majority of urinary tract tumours occurred in the urinary bladder and, since the renal pelvis and urether are covered by the same urothelium as the urinary bladder, no biologically plausible difference would be expected. Although we could not find statistically significant interaction effects, it appeared that the summary estimates of coffee and tea consumption were higher for studies that used interviewing techniques than for studies that used self-administered questionnaires. This contrast can be a consequence of response bias due to different assessment techniques or to chance alone.

Limitations in reported data meant that the summary OR could not be quantified by amount or type of tea or coffee, other than decaffeinated coffee. The included studies provided data on coffee or tea consumption within exposure categories with different cut-off points. Therefore, we had to dichotomise coffee and tea consumption in current consumption versus no consumption as well as ever consumption versus never consumption. It should be noted, that a dose-response analysis would give more information about the underlying association of coffee and tea consumption^{92,93} with urinary tract cancer. However, the result of a dose-response analysis appears to depend heavily on the choice of assigned dose of the different exposure categories.⁹² Hence, for a proper dose-response analysis, the number of exposure levels should be at least equal to four. More than 25% of the component studies reported three exposure levels at the most for coffee^{55,57,63-66,73,76,77,84,85,87} and tea consumption.^{48,50,52,53,73,74,79} Moreover, for more than 25% of the component studies, no dose-response analysis for the adjusted OR was possible, because no exposure-specific confidence intervals were reported. In fact, in only 14 out of 46 studies on coffee consumption and in two out of 14 on tea consumption, was a dose response analysis tolerable. With the above-mentioned considerations in mind, we have decided not to conduct dose-response analyses at all.

Coffee

Tea

Interrupted and uninterrupted reference lines indicates total summary odds and null effect

Figure 1. Funnel plots for current coffee and tea drinkers versus non drinkers, unadjusted

Interrupted and uninterrupted reference lines indicates total summary odds and null effect

Figure 2. Forest plots of crude summary odds ratios and 95% confidence intervals of urinary tract cancer for current coffee and tea consumers compared to non-drinkers, by measuring instrument, source of cases and controls and tumour site for case-control studies with men and women combined

Although 49 articles contributed to this meta-analysis, only 30 provided sufficient information to permit calculation of OR, adjusted for at least age, smoking and sex. It was not possible to adjust for a specific set of confounders. The decision of authors to report adjusted effect estimates or not, may have been influenced by the results of the component studies. Similarly, the type of confounders that authors chose to adjust for varied and may to some extent have been driven by the data. Therefore, we also calculated unadjusted OR. Furthermore, focussing entirely on adjusted OR would have resulted in the exclusion of 39% of the studies that also had (crude) information on the association between coffee or tea consumption and cancer of the urinary tract. The presentation of both unadjusted and adjusted OR might give some insight into the potential confounding effect of especially that of cigarette smoking. It remains possible that adjustment for smoking in the component studies, and therefore in our summary estimates, may have been inadequate if smoking was not carefully adjusted for. Yet, it appeared that adjustment for several cigarette smoking features in the component studies, compared to adjustment for one smoking variable alone, was not associated with a decreased risk of bladder cancer, as would be expected in the case of residual confounding. However, these results should be carefully interpreted since the comparisons were based on different subsets of studies. To avoid residual confounding, the impact of coffee drinking on bladder cancer risk can be evaluated in lifelong non-smokers.⁷ Of all identified articles, four reported no increased risk associated with coffee drinking among non-smokers,^{23,58,67,79} whereas 11 studies suggested a moderately increased risk.^{41,46,50,51,58,60,66,69,71,72,82}

In addition to residual confounding by cigarette smoking, another explanation for a small positive association between coffee consumption and cancer of the urinary tract might be that non-coffee-drinkers are a rather selected population. Non-coffee drinkers may differ also in occupation, the consumption of alcohol, tea, vegetables or fruit, or other lifestyle habits from the general population of coffee drinkers.

The consumption of tea seems not to be related to an increased risk of urinary tract cancer, although the results are based on only a few studies. Evidence from animal experimental studies shows that it is plausible that certain compounds present in tea (e.g., flavonoids) may even protect against the development of cancer.⁹⁴ If an inverse association between tea consumption and urinary tract cancer does exist, it is likely to be small. Further research from large epidemiological studies is needed in this area.

In accordance with earlier reviews and based on predominantly case-control data, we found that coffee consumption increases the risk of urinary tract cancer by approximately 20%. The consumption of tea seems not to be related to an increased risk of urinary tract cancer. The risk of urinary tract cancer related to the amount or type of coffee and tea needs further investigation.

REFERENCES

1. Silverman DT, Hartge P, Morrison AS et al. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am.* 1992;6:1-30.
2. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol.* 1997;13:291-8.
3. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol.* 1996;23:536-45.
4. World Cancer Research Fund, American Institute for Cancer Research. Bladder. In: *Food, Nutrition and the Prevention of Cancer: a global perspective.* Menasha: Banta Book Group, 1997:338-61.
5. van der Meijden APM. Bladder cancer. *Br Med J.* 1998;317: 366-9.
6. WHO, IARC. IARC monographs on the evaluation of carcinogenic risks to humans: coffee, tea, mate, methylxanthines and methylglyoxal, Vol. 51. Lyon: World Health Organization, 1991.
7. Silverman DT, Morrison AS, Devesa SS. Bladder cancer. In: *Cancer epidemiology and prevention* (Schottenfeld D, Fraumeni JF, eds). New York Oxford: Oxford university press, 1996:1156-79.
8. Nehlig A, Debry G. Coffee and cancer: a review of human and animal data. *World Rev Nutr Diet.* 1996;79:185-221.
9. Viscoli CM, Lachs MS, Horwitz RI. Bladder cancer and coffee drinking: a summary of case-control research. *Lancet.* 1993;341:1432-7.

10. Bloi WJ, Chow WH, McLaughlin JK. Tea and cancer: a review of the epidemiological evidence. *Eur J Cancer Prev.* 1996;5:425-38.
11. Katiyar SK, Mukhtar H. Tea consumption and cancer. *World Rev Nutr Diet.* 1996;79:154-84.
12. Zeegers MPA, Tan FES, Verhagen AP et al. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control.* 1999;10:445-51.
13. Woolf B. On estimation the relation between blood group and disease. *Ann Hum Genet.* 1954;19:251-3.
14. Tan FES, Zeegers MPA. An alternative method of combining exposure specific odds ratios in epidemiologic studies. In: Research Report. Maastricht: Maastricht University, Department Methodology and Statistics, 1999.
15. Light RJ, Pillemer DB. Quantitative procedures. In: Summing up: the science of reviewing research. Harvard: Harvard University Press, 1984.
16. Egger M, Davey Smith G, Schneider M et al. Bias in meta-analysis detected by a simple, graphical test. *Br Med J.* 1997;315:629-34.
17. Steichen TJ, Egger M, Sterne J. Tests for publication bias in meta-analysis. *Stata Technical Bulletin.* 1998;STB-44:sbe19.1.
18. StataCorp. Stata Statistical Software: Release 6.0. In: College Station, Texas: Stata Corporation, 1999.
19. Sharp SJ. Meta-analysis regression. *Stata Technical Bulletin* 1998;STB-42:sbe23.
20. Berkey CS, Hoaglin DC, Mosteller F et al. A random-effects regression model for meta-analysis. *Stat Med.* 1995;14:395-411.
21. Zheng W, Doyle TJ, Kushi LH et al. Tea consumption and cancer incidence in a prospective cohort study of postmenopausal women. *Am J Epidemiol.* 1996;144:175-82.
22. Bravo MP, Del Rey Calero J, Conde M. Risk factors of bladder cancer in Spain. *Neoplasma.* 1987;34:633-7.
23. Akdas A, Kirkali Z, Bilir N. Epidemiological case-control study on the etiology of bladder cancer in Turkey. *Eur Urol.* 1990;17:23-6.
24. Mommsen S, Aagaard J, Sell A. An epidemiological study of bladder cancer in a predominantly rural district. *Scand J Urol Nephrol.* 1983;17:307-12.
25. Mommsen S, Aagaard J, Sell A. An epidemiological case-control study of bladder cancer in males from a predominantly rural district. *Eur J Cancer Clin Oncol.* 1982;18:1205-10.
26. Mommsen S, Aagaard J. A case-control study of bladder-cancer: a multivariate, stratified analysis of a low-risk population. *Dan Med Bull.* 1983;30:427-32.
27. Whittemore AS, Paffenbarger RS, Jr., Anderson K et al. Early precursors of urogenital cancers in former college men. *J Urol.* 1984;132:1256-61.
28. Wynder EL, Dieck GS, Hall NE et al. A case-control study of diesel exhaust exposure and bladder cancer. *Environ Res.* 1985;37:475-89.
29. Whittemore AS, Paffenbarger RS, Jr., Anderson K et al. Early precursors of site-specific cancers in college men and women. *J Natl Cancer Inst.* 1985;74:43-51.
30. Snowdon DA, Phillips RL. Coffee consumption and risk of fatal cancers. *Am J Public Health.* 1984;74:820-3.
31. Kinlen LJ, Willows AN, Goldblatt P et al. Tea consumption and cancer. *Br J Cancer* 1988;58:397-401.
32. Gonzalez CA, Lopez Abente G, Errezola M et al. Occupation, tobacco use, coffee, and bladder cancer in the county of Mataro (Spain). *Cancer.* 1985;55:2031-4.
33. Miller AB. The etiology of bladder cancer from the epidemiological viewpoint. *Cancer Res.* 1977;37:2939-42.
34. Miller CT, Neutel CI, Nair RC et al. Relative importance of risk factors in bladder carcinogenesis. *J Chron Dis.* 1978;31:51-6.
35. Sullivan JW. Epidemiologic survey of bladder cancer in greater New Orleans. *J Urol.* 1982;128:281-3.
36. Mommsen S, Aagaard J, Sell A. A case-control study of female bladder cancer. *Eur J Cancer Clin Oncol.* 1983;19:725-9.
37. Claude J, Kunze E, Frentzel Beyme R et al. Life-style and occupational risk factors in cancer of the lower urinary tract. *Am J Epidemiol.* 1986;124:578-89.
38. Jacobsen BK, Bjelke E, Kvale G et al. Coffee drinking, mortality, and cancer incidence: results from a Norwegian prospective study. *J Natl Cancer Inst.* 1986;76:823-31.
39. Fraumeni JF, Jr., Scotto J, Dunham LJ. Coffee-drinking and bladder cancer. *Lancet.* 1971;2:1204.
40. Heilbrun LK, Nomura A, Stemmermann GN. Black tea consumption and cancer risk: a prospective study. *Br J Cancer.* 1986;54:677-83.
41. Mills PK, Beeson WL, Phillips RL et al. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol.* 1991;133:230-9.
42. Cole P. Coffee-drinking and cancer of the lower urinary tract. *Lancet.* 1971;26:1335-7.
43. Schmauz R, Cole P. Epidemiology of cancer of the renal pelvis and urether. *J Natl Cancer Inst.* 1974;52:1431-4.
44. Miller AB, Howe GR. Artificial sweeteners and bladder cancer. *Lancet.* 1977;10:1221-2.
45. Morrison AS, Buring JE, Verhoek WG et al. Coffee drinking and cancer of the lower urinary tract. *J Natl Cancer Inst.* 1982;68:91-4.
46. Hartge P, Hoover R, West DW et al. Coffee drinking and risk of bladder cancer. *J Natl Cancer Inst.* 1983;70:1621-6.

47. Marrett LD, Walter SD, Meigs JW. Coffee drinking and bladder cancer in Connecticut. *Am J Epidemiol*. 1983;117:113-27.
48. Ohno Y, Aoki K, Obata K et al. Case-control study of urinary bladder cancer in metropolitan Nagoya. *Natl Cancer Inst Monogr*. 1985;69:229-34.
49. Jensen OM, Wahrendorf J, Knudsen JB et al. The Copenhagen case-control study of bladder cancer. II. Effect of coffee and other beverages. *Int J Cancer*. 1986;37:651-7.
50. Risch HA, Burch JD, Miller AB et al. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol*. 1988;127:1179-91.
51. Slattery ML, Schumacher MC, West DW et al. Smoking and bladder cancer. The modifying effect of cigarettes on other factors. *Cancer*. 1988;61:402-8.
52. Slattery ML, West DW, Robison LM. Fluid intake and bladder cancer in Utah. *Int J Cancer*. 1988;42:17-22.
53. Nomura AMY, Kolonel LN, Hankin JH et al. Dietary factors in cancer of the urinary tract. *Int J Cancer*. 1991;48:199-205.
54. Momas I, Daures JP, Festy B et al. Relative importance of risk factors in bladder carcinogenesis: some new results about Mediterranean habits. *Cancer Causes Control*. 1994;5:326-32.
55. Sturgeon SR, Hartge P, Silverman DT et al. Associations between bladder cancer risk factors and tumor stage and grade at diagnosis. *Epidemiology*. 1994;5:218-25.
56. Brummer B, White E, Vaughan TL et al. Fluid intake and the incidence of bladder cancer among middle-aged men and women in a three-county area of western Washington. *Nutr Cancer*. 1997;29:163-8.
57. Dunham LJ, Rabson AS, Steward HS et al. Rates, interview, and pathology study of cancer of the urinary bladder in new orleans, Louisiana. *J Natl Cancer Inst*. 1968;41:683-709.
58. Bross IDJ, Tindings J. Another look at coffee drinking and cancer of the urinary bladder. *Prev Med*. 1973;2:445-51.
59. Morgan RW, Meera GJ. Bladder cancer: smoking, beverages and artificial sweeteners. *Can Med Assoc J*. 1974;111:1067-70.
60. Simon D, Yen S, Cole P. Coffee drinking and cancer of the lower urinary tract. *J Natl Cancer Inst*. 1975;54:587-91.
61. Armstrong B, Garrod A, Doll R. A retrospective study of renal cancer with special reference to coffee and animal protein consumption. *Br J Cancer*. 1976;33:127-36.
62. Wynder EL, Goldsmith R. The epidemiology of bladder cancer: a second look. *Cancer*. 1977;40:1246-68.
63. Mettlin C, Graham S. Dietary risk factors in human bladder cancer. *Am J Epidemiol*. 1979;110:255-63.
64. Cartwright RA, Adib R, Glashan R et al. The epidemiology of bladder cancer in west yorkshire: a preliminary report on non-occupational aetiologies. *Carcinogenesis*. 1981;2:343-7.
65. Najem GR, Louria DB, Seebode JJ et al. Life time occupation, smoking, caffeine, saccharine, hair dyes and bladder carcinogenesis. *Int J Epidemiol*. 1982;11:212-7.
66. Rebekalos A, Trichopoulos D, Tzonou A et al. Tobacco smoking, coffee drinking, and occupation as risk factors for bladder cancer in greece. *J Natl Cancer Inst*. 1985;75:455-61.
67. Kabat GC, Dieck GS, Wynder EL. Bladder cancer in nonsmokers. *Cancer*. 1986;57:362-7.
68. Iscovich J, Castelletto R, Esteve J et al. Tobacco smoking, occupational exposure and bladder cancer in Argentina. *Int J Cancer*. 1987;40:734-40.
69. Ciccone G, Vincis P. Coffee drinking and bladder cancer. *Cancer Lett*. 1988;41:45-52.
70. La Vecchia C, Negri E, Decarli A et al. Dietary factors in the risk of bladder cancer. *Nutr Cancer*. 1989;12:93-101.
71. Clavel J, Cordier S. Coffee consumption and bladder cancer risk. *Int J Cancer*. 1991;47:207-12.
72. D'Avanzo B, La Vecchia C, Franceschi S et al. Coffee consumption and bladder cancer risk. *Eur J Cancer*. 1992;28a:1480-4.
73. Kunze E, Chang Claude J, Frentzel Beyme R. Life style and occupational risk factors for bladder cancer in Germany. A case-control study. *Cancer*. 1992;69:1776-90.
74. La Vecchia C, Negri E, Franceschi S et al. Tea consumption and cancer risk. *Nutr Cancer*. 1992;17:27-31.
75. Barbone F, Franceschi S, Talamini R et al. Occupation and bladder cancer in Pordenone (north-east Italy): a case-control study. *Int J Epidemiol*. 1994;23:58-65.
76. Hours M, Danache B, Fevotte J et al. Bladder cancer and occupational exposures. *Scan J Work Environ Health*. 1994;20:322-30.
77. D'Avanzo B, La Vecchia C, Negri E et al. Attributable risks for bladder cancer in northern Italy. *Ann Epidemiol*. 1995;5:427-31.
78. Donato F, Boffetta P, Fazioli R et al. Bladder cancer, tobacco smoking, coffee and alcohol drinking in Brescia northern Italy. *Eur J Epidemiol*. 1997;13:795-800.
79. Howe GR, Burch JD, Miller AB et al. Tobacco use, occupation, coffee, various nutrients, and bladder cancer. *J Natl Cancer Inst*. 1980;64:701-13.
80. Ross RK, Paganini Hill A, Landolph J et al. Analgesics, cigarette smoking, and other risk factors for cancer of the renal pelvis and ureter. *Cancer Res*. 1989;49:1045-8.
81. Vena JE, Freudenheim J, Graham S et al. Coffee, cigarette smoking, and bladder cancer in western New York. *Ann Epidemiol*. 1993;3:586-91.

82. Pujolar EA, Gonzalez CA, Lopez Abente G et al. Bladder cancer and coffee consumption in smokers and non-smokers in Spain. *Int J Epidemiol.* 1993;22:38-44.
83. Piper JM, Matanoski GM, Tonascia J. Bladder cancer in young women. *Am J Epidemiol.* 1986;123:1033-42.
84. Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol.* 1993;3:211-6.
85. Bravo P, del Rey J, Sanchez J et al. Cafe y analgesicos como factores de riesgo del cancer de vejiga. *Arch Esp Urol.* 1986;39:337-41.
86. Donato F, Boffetta P, Fazioli R et al. Reliability of data on smoking habit and coffee drinking collected by personal interview in a hospital-based case-control study. *Eur J Epidemiol.* 1998;14:259-67.
87. Stensvold I, Jacobsen BK. Coffee and cancer: a prospective study of 43,000 Norwegian men and women. *Cancer Causes Control.* 1994;5:401-8.
88. Michaud DS, Spiegelman D, Clinton SK et al. Fluid intake and the risk of bladder cancer in men. *N Engl J Med.* 1999;340:1390-7.
89. McLaughlin JK, Blot WJ, Mandel JS et al. Etiology of cancer of the renal pelvis. *J Natl Cancer Inst.* 1983;71:287-91.
90. Armstrong B, Doll R. Environmental factors and cancer incidence and mortality in different countries, with special reference to dietary practices. *Int J Cancer.* 1975;15:617-31.
91. Morrison AS. Geographic and time trends of coffee imports and bladder cancer. *Eur J Cancer.* 1978;14:51-4.
92. Berlin JA, Longnecker MP, Greenland S. Meta-analysis of epidemiologic dose-response data. *Epidemiology.* 1993;4:218-28.
93. Greenland S, Longnecker MP. Methods for trend estimation from summarized dose-response data, with applications to meta-analysis. *Am J Epidemiol.* 1992;135:1301-9.
94. Goldbohm RA, Hertog MG, Brants HA et al. Consumption of black tea and cancer risk: a prospective cohort study. *J Natl Cancer Inst.* 1996;88:93-100.

6

A Prospective Study on Active and Environmental Tobacco Smoking and Bladder Cancer Risk

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ABSTRACT

The authors investigated in a prospective cohort study among 120,852 adult subjects, the associations between cigarette, cigar, pipe, environmental tobacco smoking (ETS) and bladder cancer. All subjects completed in 1986 a questionnaire on cancer risk factors. Follow-up for incident bladder cancer was established by linkage to cancer registries until 1992. The case-cohort analysis was based on 619 cases and 3,346 subcohort members.

Compared with never smokers, the age- and sex-adjusted incidence rate ratios (RR) for ex- and current cigarette smokers were 2.11 (95%CI 1.50-2.95) and 3.32 (95%CI 2.39-4.61), respectively. The RR for smoking duration was 1.03 (95%CI 1.02-1.04) per one-year-increment. The RR per 10 cigarettes/day was 1.27 (95%CI 1.17-1.37). Associations of cigarette smoking with bladder cancer were largely attributable to smoking duration. Tar and nicotine exposure weakly increased bladder cancer risk. Smoking cessation, age at first exposure, filter-tip usage, cigar and pipe smoking and ETS were not associated with bladder cancer.

The authors conclude that current cigarette smokers have a threefold higher bladder cancer risk than non-smokers. Ex-smokers experience a twofold increased risk. About half of male bladder cancer and one fifth of female bladder cancer was attributable to cigarette smoking. Other smoking types (cigar, pipe, or ETS) were not associated with increased risks.

Bladder cancer is the most common urological cancer and is the seventh most common cancer among men, accounting for approximately 200,000 new cases per year worldwide. Bladder cancer is considerably more common in men than women (ratio worldwide is about 3.5:1). The incidence of this cancer varies worldwide; in general, the highest incidence is observed in North America and Europe.¹ Narrative reviews, based on many case control and follow-up studies, concluded that there is strong support for a positive association between various cigarette smoking characteristics and bladder cancer.²⁻¹¹ Cigarette smoking explains the occurrence of a greater proportion of bladder cancer than does any other known environmental factor.² In a recent systematic review of 43 epidemiological studies, based on predominantly case-control data, we concluded that current cigarette smokers may have an approximately threefold risk of bladder cancer compared with lifelong non-smokers.¹²

However, the majority of earlier studies on cigarette smoking have presented the association between individual cigarette smoking characteristics on bladder cancer adjusted for only other environmental factors (e.g., diet, occupation). Although of interest, few studies simultaneously included different smoking characteristics in a regression model to estimate the individual impact of cigarette smoking features. The impact of active cigar and pipe smoking and environmental tobacco smoking (ETS) has also been evaluated less often than active cigarette smoking.¹³⁻²⁵

These findings prompted us to investigate the influence of cigarette, cigar, pipe and ETS on the risk of bladder cancer in more detail in the ongoing Netherlands Cohort Study. In this follow-up study among 120,852 men and women, which started in 1986, detailed information has been collected on life time smoking history, including smoking status, frequency, age at first exposure, duration, cessation, inhalation, filter usage, tobacco brand and ETS, by means of a self-administered questionnaire.

MATERIALS AND METHODS

Cohort. This population based prospective cohort study on diet and cancer started in the Netherlands in September 1986. The cohort includes 58,279 men and 62,573 women aged 55 to 69 years at baseline. The study population originated from 204 municipal population registries throughout the country. The case cohort approach was used for data processing and analysis.²⁶ Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after the baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up, during the follow-up period. The study design, including data collection strategies, has been described in detail previously.²⁷

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.²⁸ The completeness of cancer follow-up was estimated to be over 96 percent.²⁹ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 3,346 subcohort members (1,630 men and 1,716 women) and 619 incident cases (532 men and 87 women) with microscopically confirmed, incident carcinomas of the urinary bladder, ureters, renal pelvis or urethra were identified. Of these cases, 584 (94.3%) were diagnosed with bladder cancer of which 559 (95.7%) were transitional cell carcinomas. The morphology of the remaining carcinomas were squamous cell (n=3), adeno (n=6), mixed (n=1) or not specified (n=15). Because the overwhelming majority of tumors occurred in the urinary bladder and the ureters, renal pelvis and urethra are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Questionnaire. At baseline, all cohort members completed a self-administered questionnaire on risk factors for cancer. Tobacco smoking was addressed by questions on age at first exposure to smoking, age at last exposure to smoking, smoking frequency, inhalation and smoking duration for cigarette, cigar and pipe smokers. Furthermore, questions were asked about the cigarette brand most commonly smoked with or without filter-tip and the proportion of a cigarette actually smoked (using a Visual Analogue Scale). ETS was inquired by questions on smoking habits of parents and spouses, exposure to environmental smoke at work and duration of exposure to ETS in private and occupational setting combined. The questionnaire data were key entered twice and processed in a standardized manner blinded with respect to case/sub-cohort status in order to minimize observer bias in coding and interpretation of the data.

Statistical analysis. Based on the questionnaire data we constructed the following variables on active cigarette, cigar and pipe smoking: status (never, ex, current), frequency (cigarettes, cigars or pipes per day), age at first exposure (years), duration (years), years since cessation (years), inhalation (yes/no), filter usage (filter, non-filter) and ETS: spouse smoking status (never, ex, current), parental smoking (yes/no), exposure to environmental smoke at work (high/low) and duration of exposure to ETS (hours/day). The continuous tobacco smoking characteristics were categorized into strata based on the distribution in the sub-cohort. We calculated the daily exposure to tar and nicotine for cigarette smokers from cigarette brand specific information obtained from the Dutch Inspectorate for Health Protection, the Dutch Foundation on Smoking and Health and the Dutch Foundation of Tobacco Industry in combination with daily cigarette smoking frequency and the percentage of a cigarette actually smoked. Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models³⁰ with the Stata statistical software package.³¹ Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow³² and Lin and Ying.³³ Tests for dose response trends in risk for bladder cancer over multiple categories were assessed by fitting ordinal exposure variables as continuous terms and performing likelihood-ratio tests between regression models with and without these variables. We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against failure time.

The effect of active tobacco smoking and ETS was first evaluated by regression analyses after adjustment for age and sex and list wise deletion of missing smoking data. Based on earlier analyses,³⁴⁻³⁶ the following variables were subsequently considered as potential additional confounders: intake of alcohol (g/day), coffee (cups/day), tea (cups/day) water (ml/day), vegetables (g/day) and fruit (g/day), occupational exposure to dye, rubber, leather or vehicle fumes (ever/never), and first degree family history of bladder cancer (yes/no). Those variables that showed a more than ten percent influence on the risk of bladder cancer when considered in a multivariable model were included as covariates in multivariable analyses. The adjusted incidence rate ratios of bladder cancer for cigarette, cigar and pipe smokers compared with non-smokers are presented according to the different smoking features. Furthermore, we compared different regression models to identify the main predictors of bladder cancer, taking into account the changes in effect sizes and the contribution to the log likelihood of the regression model. To prevent redundancy and collinearity, smoking status (current smoking *vs.* ex/never smoking), smoking inhalation (yes *vs.* no/never smoking) and filter usage (non-filter *vs.* filter/never smoker) were recoded as indicator variables when they were simultaneous incorporated in one regression model with other smoking covariables. Based on the proportion of current cigarette smokers in the sub-cohort and the sex specific age-adjusted incidence rate ratios we estimated the population attributable risk of bladder cancer for men and women. To ensure that the results were not influenced by changes in exposure by subjects with preclinical bladder cancer, we conducted analyses

with and without cases diagnosed in the first one or two years of follow-up. The results from these analyses were similar and therefore analyses with all cases were presented.

RESULTS

The distribution of bladder cancer cases and subcohort members according to age, sex and tobacco product ever smoked is presented in table 1. The age distribution among cases and subcohort members was comparable. Almost all men in our study population (96% of the cases, 90% of the subcohort members) and approximately half of the women (60% of the cases, 41% of the subcohort members) reported ever smoking of cigarettes (Table 1). Among these, the prevalence of the ex-smokers was equal to the prevalence of the current smokers. Most men and almost all women never smoked cigar or pipe during their life. If cigar or pipe were smoked, than it was most often combined with cigarette smoking. Cigarettes were more often smoked solely than in combination with other types of tobacco (Table 1).

Table 1. Distribution of bladder cancer cases and subcohort members according to age, sex and tobacco product ever smoked, Netherlands Cohort Study (1986-1992)

	Cases		Subcohort	
	Men (n=532)	Women (n=87)	Men (n=1630)	Women (n=1716)
	number (%)			
Age (years)				
55-59	119 (22.4)	29 (33.3)	620 (38.0)	651 (37.9)
60-64	207 (38.9)	29 (33.3)	568 (34.9)	583 (34.0)
65-69	206 (38.7)	29 (33.3)	442 (27.1)	482 (28.1)
Tobacco product				
Never smokers	20 (3.8)	35 (40.2)	159 (9.8)	1019 (59.4)
Cigarette	334 (62.8)	51 (58.6)	977 (59.9)	689 (40.2)
Cigar	7 (1.3)	0 (0.0)	37 (2.3)	0 (0.0)
Pipe	3 (0.6)	0 (0.0)	5 (0.3)	0 (0.0)
Cigarette, Pipe	26 (4.9)	0 (0.0)	73 (4.5)	0 (0.0)
Cigarette, Cigar	74 (13.9)	1 (1.2)	196 (12.0)	4 (0.2)
Cigar, Pipe	6 (1.1)	0 (0.0)	18 (1.1)	0 (0.0)
All three products	59 (11.1)	0 (0.0)	162 (9.9)	0 (0.0)

The association between cigarette smoking and bladder cancer was similar in men and women for all cigarette smoking characteristics. Therefore, the results are presented for men and women combined. As is shown in table 2, cigarette smoking clearly appeared to be associated with bladder cancer risk. The incidence rate ratio of bladder cancer was 3.32 (CI 2.39-4.61) for current cigarette smokers compared with never smokers. The sex-specific RRs were 3.71 (CI 2.27-6.05) and 2.42 (CI 1.44-4.05) for men and women, respectively. Stopping smoking appeared beneficial, although ex-smokers still experienced an increased RR of bladder cancer compared with lifelong non-cigarette smokers: 2.24 (CI 1.37-3.65) and 2.29 (CI 1.34-3.90) for men and women, respectively. Both the number of cigarettes smoked per day and the years of cigarettes smoked during lifetime were associated with an increased risk of bladder cancer with statistically significant positive trends in risk. Age at first exposure and years since smoking cessation were negatively associated with the risk of bladder cancer (*p*-trend: 0.05 and <0.01, respectively) and inhalation increased this risk. The incidence rate ratios associated with an increase in age at first exposure or years of smoking cessation were 0.98 (95%CI 0.95-1.00) and 0.96 (95%CI 0.95-0.98), respectively. It appeared that the use of filter-tipped cigarettes did not lower the risk of bladder cancer substantially compared with the use of non filter-tipped cigarettes (Table 2). Additional adjustment for other non-smoking factors (i.e., alcohol, tea, coffee, water, vegetables, fruit, occupation,

and family history) or active cigar and pipe smoking did not change the risk estimates for cigarette smoking. Based on the results of this study, we estimated that cigarette smoking might account for 23 percent of all female bladder cancer, whereas in men 50 percent of the disease may be associated with cigarette smoking.

Table 2. Incidence rate ratios (RR) for bladder cancer according to cigarette smoking features for men and women in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Cigarette smoking Features	Cases in cohort	Person years in subcohort	RR (95% CI)*
Never tobacco smoker†	55	7276	1.00 (referent)
Status			
Ex	263	7001	2.11 (1.50-2.95)
Current	282	5664	3.32 (2.39-4.61)
Frequency (cig/day)			
<5	30	1488	1.77 (1.08-2.89)
5-<10	59	1826	2.40 (1.57-3.67)
10-<15	87	2463	2.23 (1.50-3.30)
15-<20	93	1780	3.38 (2.28-5.01)
20-<25	120	2329	3.22 (2.21-4.69)
25+	115	1900	3.65 (2.48-5.38)
p-value for linear trend			<0.01
Increment, 10 cig/day			1.27 (1.17-1.37)
Duration (years)			
<10	10	632	1.41 (0.68-2.94)
10-<20	39	1592	1.78 (1.12-2.84)
20-<30	63	2506	1.72 (1.14-2.60)
30-<40	125	3213	2.69 (1.87-3.87)
40-<50	220	3807	3.40 (2.40-4.80)
50+	79	565	5.40 (3.45-8.45)
p-value for linear trend			<0.01
Increment, 1 year			1.03 (1.02-1.04)
Age at first exposure (years)			
<15	140	2059	3.51 (0.39-5.17)
15-<17	143	2796	2.90 (0.98-4.25)
17-<19	108	3056	2.26 (1.55-3.31)
19-<21	79	1806	3.07 (2.04-4.60)
21-<25	33	1012	2.47 (1.52-4.03)
25+	33	1666	2.04 (1.28-3.25)
p-value for linear trend			0.05
Increment, 1 year‡			0.98 (0.95-1.00)
Years since cessation			
<1	295	5821	3.40 (2.45-4.71)
1-<10	112	2240	2.94 (2.02-4.29)
10-<20	71	2324	1.69 (1.13-2.52)
20-<30	54	1527	1.85 (1.20-2.86)
30+	11	723	0.81 (0.40-1.63)
p-value for linear trend			<0.01
Increment, 1 year‡			0.96 (0.95-0.98)
Inhalation			
No	95	3659	1.93 (1.33-2.80)
Yes	428	8610	3.06 (2.19-4.27)
Filter usage			
Filter-tipped	85	3374	2.63 (1.82-3.80)
Non-filter-tipped	314	6063	2.64 (1.85-3.77)

*adjusted for age (years) and sex

†never smoked cigars, pipe, or cigarettes

‡lifelong non-smokers were excluded from the continuous analysis

Table 3 presents different regression models to identify the main smoking-related predictors of bladder cancer among ever cigarette smokers. Incorporation in the regression equation of age at first exposure and years since cessation instead of smoking duration did not change the log likelihood of the age- and sex-adjusted model. The magnitude of the reciprocal incidence rate ratios for age at first exposure (RR=1.02) and years since cessation (RR=1.04) were similar to the incidence rate ratio for smoking duration (RR=1.04).

Table 3. Adjusted* incidence rate ratios (RR) for bladder cancer according to four different regression models including cigarette smoking frequency, cigarette smoking duration, age at first exposure and years since cessation in continuous analyses, Netherlands Cohort Study (1986-1992)

	Model 1 RR (95% CI)	Model 2 RR (95% CI)	Model 3 RR (95% CI)	Model 4 RR (95% CI)
Frequency (increment, 10cig/day)	-	1.17 (1.06-1.29)	-	1.19 (1.08-1.31)
Duration (increment 1 year)	1.04 (1.02-1.05)	1.03 (1.02-1.05)	-	-
Age at first exposure (increment 1 year)	-	-	0.98 (0.95-1.00)	0.98 (0.96-1.00)
Years since cessation (increment 1 year)	-	-	0.96 (0.95-0.98)	0.96 (0.95-0.98)
Log likelihood	-7482.78	-7475.71	-7485.88	-7477.08

*additionally adjusted for age (years) and sex; lifelong non-smokers were excluded from the analysis

Cigarette smoking frequency, however, still contributed significantly to the log likelihood of the model even after cigarette smoking duration was incorporated ($p<0.01$) (Table 3).

The incidence rate ratios for current *vs.* ex-smokers, smoke inhalers *vs.* smoke non-inhalers and users of non-filter-tipped cigarettes *vs.* users of filter-tipped cigarettes dropped towards unity after adjustment for cigarette smoking frequency and duration (table 4). The incidence rate ratios for men and women smoking <10, 10-<20, 20-<25 or more 25 cigarettes per day were 0.75, 0.88, 0.73, 1.07, 1.01, and 1.18 compared with non-smokers after adjustment for age, sex and smoking duration, respectively (p -trend=0.02).

The incidence rate ratios for subjects smoking for a period of <10, 10-<20, 20-<30, 30-<40, 40-<50, or more than 50 years were 1.33, 1.45, 1.31, 2.12, 2.74, and 4.62 compared with non-smokers after adjustment for age, sex, and smoking frequency, respectively (p -trend<0.01). Further adjustment for cigarette smoking status, inhalation, or filter usage did not change the results substantially (Table 4).

Increasing daily tar and nicotine exposure were strong positively associated with bladder cancer risk in age- and sex-adjusted analyses, but weakly associated after adjustment for cigarette smoking frequency and cigarette smoking duration (Table 5).

Because almost no women reported ever smoking cigars or pipes and men most commonly smoked cigars and pipes in combination with cigarettes, the incidence rate ratios from cigar and pipe smoking compared with non-tobacco smoking were estimated for men only, adjusted for cigarette smoking. Analyses for cigar and pipe smoking solely were not possible due to sparse data. Age confounded the association between cigar and pipe smoking and the risk of bladder cancer (Table 6). The incidence rate ratio for cigar and pipe smoking indicated a statistically significant increased risk for cigar and pipe smokers. However, after additional adjustment for the cigarette smoking frequency and duration, almost none of the estimated incidence rate ratios for different cigar or pipe smoking characteristics remained statistically significant. The point estimates suggested that cigar and pipe smoking are not associated with an increased bladder cancer risk. Only the number of pipes smoked per day was positively associated with bladder cancer risk, although not statistically significant (p -trend=0.17) (Table 6).

Table 4. Incidence rate ratios (RR) for bladder cancer according to cigarette smoking features for men and women after simultaneous inclusion of cigarette smoking frequency and duration in the regression model, Netherlands Cohort Study (1986-1992)

Cigarette smoking Features	RR (95% CI)
Status*	
Ex	1.00 (referent)
Current	1.07 (0.82-1.39)
Frequency (cig/day)†	
Increment, 10 cig/day	1.14 (1.04-1.26)
Duration (years)‡	
Increment, 1 year	1.03 (1.02-1.04)
Inhalation*	
No	1.00 (referent)
Yes	1.15 (0.89-1.50)
Filter usage*	
Filter-tipped	1.00 (referent)
Non-filter-tipped	0.95 (0.70-1.28)

*adjusted for age (years), sex, cigarette smoking frequency (cigarettes/day), and cigarette smoking duration (years)

†adjusted for age (years), sex, and cigarette smoking duration (years)

‡adjusted for age (years), sex, and cigarette smoking frequency (cigarettes/day)

Table 5. Adjusted incidence rate ratios (RR) for bladder cancer according to tar and nicotine exposure from ever cigarette smoking for men and women in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Tar / Nicotine	Cases in cohort	Person years in subcohort	RR (95% CI)*	RR (95% CI)†
Never tobacco smoker‡	55	7276	1.00 (referent)	1.00 (referent)
Tar (mg/day)				
<100	45	1959	2.25 (1.44-3.51)	0.80 (0.40-1.60)
100-<200	68	1480	3.22 (2.11-4.90)	1.09 (0.53-2.25)
200-<300	73	1400	3.02 (1.99-4.58)	1.00 (0.46-2.16)
300-<400	46	902	3.05 (1.89-4.91)	1.01 (0.44-2.35)
400-<500	37	379	4.93 (2.86-8.49)	1.52 (0.61-3.83)
500+	52	632	4.53 (2.83-7.24)	1.66 (0.65-4.21)
p-value for linear trend			<0.01	0.02
Increment, 100 mg/day			1.15 (1.09-1.22)	1.08 (0.93-1.25)
Nicotine (mg/day)				
<10	59	2417	2.23 (1.47-3.39)	0.78 (0.39-1.54)
10-<20	89	1700	3.48 (2.34-5.17)	1.12 (0.54-2.33)
20-<30	68	1278	3.13 (2.04-4.82)	0.94 (0.42-2.13)
30-<40	36	516	3.54 (2.10-5.97)	1.10 (0.47-2.54)
40-<50	18	267	3.40 (1.79-6.44)	1.02 (0.39-2.72)
50+	52	590	4.74 (2.95-7.61)	1.43 (0.57-3.62)
p-value for linear trend			<0.01	0.08
Increment, 10 mg/day			1.12 (1.06-1.18)	1.03 (0.94-1.13)

*adjusted for age (years) and sex

†adjusted for age (years), sex, cigarette smoking frequency (cigarettes/day), and cigarette smoking duration (years)

‡never smoked cigars, pipe, or cigarettes

Table 6. Adjusted incidence rate ratios (RR) for bladder cancer according to cigar and pipe smoking characteristics for men in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Cigar/Pipe smoking features	Cigar smoking		Pipe smoking	
	RR (95%CI)*	RR (95%CI)†	RR (95%CI)*	RR (95%CI)†
Never smoker‡	1.00 (referent)	1.00 (referent)	1.00(referent)	1.00 (referent)
Status				
Ex	2.15 (1.23-3.76)	0.44 (0.14-1.35)	2.39 (1.34-4.28)	1.00 (0.32-3.16)
Current	2.88 (1.68-4.93)	0.61 (0.17-2.16)	3.03 (1.67-5.50)	1.00 (0.25-3.93)
Frequency (cig/day)				
<2	2.11 (1.15-3.85)	0.48 (0.13-1.77)	1.73 (0.74-4.03)	1.28 (0.30-5.51)
2-<5	1.89 (1.03-3.47)	0.45 (0.13-1.57)	2.19 (1.14-4.20)	1.45 (0.44-4.78)
5+	4.01 (2.22-7.22)	0.86 (0.23-3.18)	3.04 (1.61-5.73)	1.69 (0.44-6.47)
p-trend	<0.01	0.23	<0.01	0.26
Increment, 1/day	1.13 (1.07-1.19)	1.09 (1.00-1.18)	1.10 (1.03-1.17)	1.02 (0.92-1.12)
Duration (years)				
<20	2.60 (1.46-4.64)	0.52 (0.17-1.64)	2.86 (1.58-5.19)	0.97 (0.27-3.51)
20-<40	2.41 (1.36-4.25)	0.42 (0.13-1.36)	2.11 (1.08-4.12)	0.70 (0.20-2.47)
40+	3.28 (1.74-6.18)	0.65 (0.18-2.40)	3.12 (1.51-6.44)	0.44 (0.09-2.21)
p-trend	<0.01	0.89	<0.01	0.03
Increment, 1 year	1.02 (1.01-1.03)	1.00 (0.98-1.02)	1.01 (1.00-1.03)	0.98 (0.96-1.00)
Age at first exposure (years)				
<20	3.12 (1.68-5.81)	0.40 (0.10-1.61)	2.66 (1.26-5.62)	1.00 (0.24-4.11)
20-<30	2.00 (1.11-3.60)	0.33 (0.10-1.12)	1.87 (0.92-3.81)	1.05 (0.31-3.50)
30+	2.76 (1.53-4.98)	0.37 (0.10-1.38)	3.18 (1.77-5.73)	1.24 (0.38-4.06)
p-trend	<0.01	0.37	<0.01	0.59
Increment, 1 year§	1.00 (0.98-1.02)	1.00 (0.98-1.02)	0.99 (0.97-1.01)	0.99 (0.97-1.02)
Years since cessation				
<10	1.85 (0.98-3.47)	0.42 (0.13-1.33)	2.54 (1.36-4.74)	1.07 (0.33-3.44)
10-<20	2.79 (1.46-5.34)	0.66 (0.19-2.36)	2.51 (1.18-5.35)	0.72 (0.18-2.88)
20+	2.88 (1.68-4.93)	0.71 (0.20-2.50)	2.89 (1.59-5.24)	0.81 (0.20-3.29)
p-trend	0.18	0.01	<0.01	0.44
Increment, 1 year§	0.98 (0.96-1.00)	0.98 (0.94-1.02)	1.00 (0.98-1.02)	1.03 (1.00-1.07)
Inhalation				
No	2.10 (1.22-3.62)	0.36 (0.11-1.22)	2.65 (1.50-4.68)	0.87 (0.24-3.14)
Yes	3.46 (1.94-6.17)	0.47 (0.12-1.90)	2.34 (1.18-4.63)	0.70 (0.15-3.29)

* adjusted for age (years)

† adjusted for age (years), cigarette smoking frequency (cigarettes/day), and cigarette smoking duration (years)

‡ never smoked cigars, pipe, or cigarettes

§ lifelong non-smokers were excluded from the analysis

For passive smokers, age and sex appeared to be influential confounders. Exposure to ETS from spouses, parents or work was not associated with an increased risk of bladder cancer among never tobacco smokers (Table 7). Furthermore, duration of exposure to environmental smoking in private and occupational settings combined did not appear to increase the risk of bladder cancer. Where the point estimates for parental and work related ETS were above unity, the estimates for ETS from spouses and duration of exposure were lower. However, none of the presented incidence rate ratios for ETS were statistically significant. All estimates were based on small numbers (Table 7).

Table 7. Adjusted incidence rate ratios (RR) for bladder cancer according to environmental tobacco smoking among never tobacco smokers, Netherlands Cohort Study (1986-1992)

Environmental Tobacco Smoking (ETS)	Cases in cohort	Person years in subcohort	RR (95% CI) ^a
Partner smoking status			
Never	19	1484	1.00 (referent)
Ex	18	2709	0.95 (0.46-1.97)
Current	11	2116	0.74 (0.29-1.87)
Parents smoking			
No	10	1212	1.00 (referent)
Yes	42	5760	1.15 (0.56-2.36)
Work related ETS			
Low	19	3097	1.00 (referent)
High	21	2233	1.35 (0.70-2.60)
Duration exposure ETS (hours/day)^b			
no exposure	18	1868	1.00 (referent)
1-3	13	1930	0.69 (0.33-1.44)
3+	10	1720	0.64 (0.29-1.42)
p-value for linear trend			0.24
Increment, 1 hour/day			0.94 (0.86-1.04)

^aadjusted for age (years) and sex

^bin private and occupational settings combined

DISCUSSION

The results of this prospective cohort study strongly supports a positive association between active cigarette smoking and bladder cancer risk in both men and women. Current cigarette smokers have an approximately threefold higher risk of bladder cancer and ex-smokers experience a twofold increased risk. It appeared that associations of cigarette smoking characteristics with bladder cancer risk were largely attributable to cigarette smoking duration only; that cigarette smoking cessation, age at first exposure and usage of filter-tips did not influence bladder cancer risk; and that exposure to tar and nicotine weakly increased the risk of bladder cancer. Other types of smoking (i.e., cigar, pipe or passive smoking) were not associated with an increased risk.

One strength of this and other prospective studies is that exposure was assessed before diagnosis of bladder cancer. Therefore, recall bias is not likely to have influenced our results. Furthermore, selection bias is not likely, because the follow-up of cases and subcohort members was almost complete.^{29,37} The relatively large number of bladder cancer cases in this study was an important strength.

Even though previous epidemiological studies on cigarette smoking associated with bladder cancer (8 cohort and 35 case control studies) differed in methodology, they consistently suggested a substantial increase in risk of bladder cancer for cigarette smokers.¹² The precise mechanism by which cigarette smoking causes bladder cancer has yet to be determined. It seems most likely that the risk of bladder cancer is related to some of the large number of compounds present in smoke. 2-Naphtylamine and 4-aminobiphenyl are the leading candidates as the specific etiological agents.^{4,9,10,38,39} Several nitrosamines have also been shown to produce bladder cancer in animal models.¹⁰ Tars induce bladder papillomas and carcinomas in mice.⁹ However, this study suggests that smoking of low-tar and nicotine cigarettes with or without filter-tips does not reduce the risk of bladder cancer substantially after controlling for cigarette smoking status, frequency and duration. To our knowledge, no other epidemiological study on the association between tars and nicotine exposure and bladder cancer risk has been conducted. One case control study reported a diminution of risk from the smoking of light tobacco.⁴⁰ However, this risk estimate was not adjusted for cigarette smoking duration.⁴⁰ The calculation of tar and nicotine exposure from cigarettes might have been confounded by misclassification of the cigarette brand most commonly smoked if the

validity of self-reporting for ex-smokers is lower than for current smokers. This might be possible because of recall bias.⁴¹ However, the incidence rate ratios for tar and nicotine exposure from cigarettes were comparable between ex and current smokers. An alternative explanation for the reported weak association between exposure to tars and nicotine and bladder cancer risk is that the smoking of light and filter-tipped cigarettes is compensated with an increased puff volume, frequency and duration, inter-puff interval, or volume and duration of inhalation.^{42,43} In the present study, both frequency and duration of cigarette smoking were statistically significantly higher among smokers of filter-tip cigarettes.

The fact that cigarette smoking frequency and duration were consistently associated with an increased risk of bladder cancer in both categorical and continuous models suggests a direct positive dose response association. In our dataset, the men *vs.* women incidence rate ratio decreased from 6.70 (CI 5.28-8.50) to 3.51 (CI 2.67-4.62) after adjustment for these smoking factors. Furthermore, the fact that both age at first exposure and years since cessation of exposure have an influence in modifying the risk of bladder cancer might suggest that cigarette smoke contains both initiators and promoters for bladder carcinogenesis. However, it appeared that the associations of these cigarette smoking characteristics with bladder cancer risk could largely be explained by cigarette smoking duration. The statistically significant incidence rate ratios for all cigarette smoking characteristics decreased substantially towards unity (and beyond) after adjustment for cigarette smoking duration. This would suggest that the promoting activity of cigarette smoke is of more importance than the initiating activity.

The evidence of an association between bladder cancer risk and other forms of tobacco use (*i.e.*, cigar and pipe) is inconsistent and, when found, generally weaker than the association with cigarette smoking.^{4,8,10,44} Despite the plausibility of associations between cigar and pipe smoking and the risk of bladder cancer (since the smoke of both types of tobacco contains many of the same substances as that of cigarette tobacco), it has been suggested that their effect is likely to be small.^{14,17,45} In the present study, most incidence rate ratio for cigar and pipe smoking were found to be lower than one after adjustment for cigarette smoking, which is probably an artefact of chance, since the confidence intervals were rather large.

Data from experimental studies support the hypothesis that non-smokers might be exposed to potential carcinogens through the cigarette smoking of others.⁴⁶ Mutagens have been detected in the blood, urine of passive smokers.^{46,47} Therefore, it seems plausible to suggest that individuals exposed to ETS might be at increased risk of bladder cancer.¹⁷ However, in the present follow-up study and in two previous case-control studies no excess risk of bladder cancer due to ETS was demonstrated.^{17,48} According to Burch *et al.*,¹⁷ it is not likely that any association between passive smoking and bladder cancer will be strong, considering that the association between passive smoking and lung cancer appears to be relatively weak as well. However, these findings are preliminary and must be confirmed with other studies.

We conclude that active cigarette smoking is strongly associated with bladder cancer risk. Current cigarette smokers have an approximately threefold higher risk of bladder cancer than non-smokers. Stopping smoking might be beneficial, although ex-smokers still experience a twofold increased risk. In this study, about half of male bladder cancer and one fifth of female bladder cancer was attributable to cigarette smoking. Other types of smoking (*i.e.*, cigar, pipe or passive smoking) were not associated with an increased risk.

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REFERENCES

1. Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer*. 1999;80:827-41.
2. Morrison AS. Advances in the etiology of urothelial cancer. *Urol Clin North Am*. 1984;11:557-66.
3. World Health Organization. IARC monographs on the evaluation of carcinogenic risks to humans: tobacco smoking. Vol. 38. Lyon: World Health Organization, 1986.
4. Dolin PJ. An epidemiological review of tobacco use and bladder cancer. *J Smoking Related Dis*. 1991;2:129-143.
5. Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am*. 1992;6:1-30.
6. Shirai T. Etiology of bladder cancer. *Semin Urol*. 1993;11:113-26.
7. Shirai T, Fradet Y, Huland H, Bollack C, Droller M, Janknegt R, Jones P, Kagawa S. The etiology of bladder cancer: are there any new clues or predictors of behavior? *Int J Urol*. 1995;2:64-75.
8. Silverman DT, Morrison AS, Devesa SS. Bladder cancer. In: Schottenfeld D, Fraumeni JF, eds. *Cancer epidemiology and prevention*. New York and Oxford: Oxford University Press, 1996;1156-79.
9. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol*. 1996;23:536-45.
10. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol*. 1997;13:291-8.
11. van der Meijden APM. Bladder cancer. *British Medical Journal*. 1998;317:1366-1369.
12. Zeegers MP, Tan FE, Dorant E, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer*. 2000;89:630-9.
13. Sorahan T, Sole G. Coarse fishing and urothelial cancer: a regional case-control study. *Br J Cancer*. 1990;62:138-41.
14. Hartge P, Hoover R, Kantor A. Bladder cancer risk and pipes, cigars and smokeless tobacco. *Cancer*. 1985;15:901-6.
15. Slattery ML, Schumacher MC, West DW, Robison LM. Smoking and bladder cancer. The modifying effect of cigarettes on other factors. *Cancer*. 1988;61:402-8.
16. Jensen OM, Knudsen JB, McLaughlin JK, Sorensen BL. The Copenhagen case-control study of renal pelvis and ureter cancer: role of smoking and occupational exposures. *Int J Cancer*. 1988;41:557-61.
17. Burch JD, Rohan TE, Howe GR, Risch HA, Hill GB, Steele R, Miller AB. Risk of bladder cancer by source and type of tobacco exposure: a case-control study. *Int J Cancer*. 1989;44:622-8.
18. Armstrong B, Garrod A, Doll R. A retrospective study of renal cancer with special reference to coffee and animal protein consumption. *British Journal of Cancer*. 1976;33:127-36.
19. Wynder EL, Goldsmith R. The epidemiology of bladder cancer: a second look. *Cancer*. 1977;40:1246-68.
20. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in New Orleans, Louisiana. *J natl cancer inst*. 1968;41:683-709.
21. Tyrrell AB, MacAirt JG, McCaughey WT. Occupational and non-occupational factors associated with vesical neoplasm in Ireland. *J Ir Med Assoc*. 1971;64:213-7.
22. Sorahan T, Hamilton L, Wallace DM, Bathers S, Gardiner K, Harrington JM. Occupational urothelial tumours: a regional case-control study. *Br J Urol*. 1998;82:25-32.
23. Mommsen S, Aagaard J. A case-control study of bladder-cancer: a multivariate, stratified analysis of a low-risk population. *Dan Med Bull*. 1983;30:427-32.
24. Mommsen S, Aagaard J. Tobacco as a risk factor in bladder cancer. *Carcinogenesis*. 1983;4:335-8.
25. Moller-Jensen O, Wahrendorf J, Blettner M, Knudsen JB, Sorensen BL. The copenhagen case-control study of bladder cancer: role of smoking in invasive and non-invasive bladder tumours. *Journal of Epidemiology and Community Health*. 1987;41:30-6.
26. Prentice RL. A case-cohort design for epidemiologic cohort studies and disease prevention trials. *Biometrika*. 1986;73:1-11.
27. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol*. 1990;43:285-95.
28. van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, Hunen PHM. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol*. 1990;19:553-8.
29. Goldbohm RA, van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond*. 1994;72:80-4.
30. Volovics A, van den Brandt PA. Methods for the analyses of case-cohort studies. *Biometrical Journal*. 1997;2:195-214.
31. StataCorp. *Stata Statistical Software: Release 6.0*. College Station, TX: Stata Corporation, 1999.
32. Barlow WE. Robust variance estimation for the case-cohort design. *Biometrics*. 1994;50:1064-72.
33. Lin DY, Ying Z. Cox regression with incomplete covariate measurements. *JASA*. 1993;88:1341-9.
34. Zeegers MP, Tan FES, Goldbohm RA, van den Brandt PA. Are coffee and tea consumption associated with urinary tract cancer risk: a systematic review and meta-analysis. *Int J Epidemiol*. 2001;in press.
35. Zeegers MP, Tan FE, Verhagen AP, Weijenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.

36. Steinmaus CM, Nunez S, Smith AH. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol*. 2000;151:693-702.
37. van den Brandt PA, van het Veer P, Goldbohm RA. A prospective cohort study on dietary fat and the risk of postmenopausal breast cancer. *Cancer Res*. 1993;53:75-82.
38. Morrison AS, Cole P. Epidemiology of bladder cancer. *Urol Clin North Am*. 1976;3:13-29.
39. Vincis P. Epidemiological models of carcinogenesis: the example of bladder cancer. *Cancer Epidemiol, Biomarkers Prev*. 1992;1:149-153.
40. Lopez Abente G, Gonzalez CA, Errezola M, Escolar A, Izarzugaza I, Nebot M, Riboli E. Tobacco smoke inhalation pattern, tobacco type, and bladder cancer in Spain. *Am J Epidemiol*. 1991;134:830-9.
41. Peach H, Shah D, Morris RW. Validity of smokers' information about present and past cigarette brands-implications for studies of the effects of falling tar yields of cigarettes on health. *Thorax*. 1986;41:203-7.
42. Herning RI, Jones RT, Benowitz NL, Mines AH. How a cigarette is smoked determines blood nicotine levels. *Clin Pharmacol Ther*. 1983;33:84-90.
43. Tobacco use - United States, 1900-1999. *JAMA*. 1999;282:2202-2204.
44. Matanoski GM, Elliott EA. Bladder cancer epidemiology. *Epidemiol Rev*. 1981;3:203-29.
45. Morrison AS, Buring JE, Verhock WG, Aoki K, Leck I, Ohno Y, Obata K. An international study of smoking and bladder cancer. *Journal d Urologie*. 1984;131:650-4.
46. Sandler DP, Wilcox AJ, Everson RB. Cumulative effects of lifetime passive smoking on cancer risk. *Lancet*. 1985;1:312-5.
47. Bos RP, Theuvs JL, Henderson PT. Excretion of mutagens in human urine after passive smoking. *Cancer Lett*. 1983;19:85-90.
48. Sandler DP, Everson RB, Wilcox AJ. Passive smoking in adulthood and cancer risk. *Am J Epidemiol*. 1985;121:37-48.

7 Alcohol Consumption and Bladder Cancer Risk: Results from the Netherlands Cohort Study

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ABSTRACT

Although several epidemiologic studies have been conducted on alcohol consumption and bladder cancer risk, the risk according to quantity and type of alcohol consumed is not clear. The authors investigated these associations in a large prospective cohort study on diet and cancer among 120,852 subjects in the Netherlands aged 55-69 years at baseline (1986). Subjects completed a questionnaire on risk factors for cancer, including alcohol consumption. Follow-up for incident cancer was established by record linkage to cancer registries. The case-cohort analysis was restricted to a follow-up period of 6.3 years and was based on 594 cases with bladder cancer and 3,170 subcohort members. The authors corrected for age and smoking in multivariable analyses. The incidence rate ratios for men who consumed <5, 5-<15, 15-<30 and ≥ 30 grams of alcohol per day were 1.49, 1.52, 1.16, and 1.63 compared with nondrinkers, respectively (p for trend=0.13). Alcohol consumed from beer, wine, and liquor was associated with moderately elevated risks, although most were not statistically significant. The incidence rate ratios for women varied around unity. The results of this study do not suggest an important association between alcohol consumption and bladder cancer risk.

Although several epidemiologic studies have been conducted to investigate the association between alcohol consumption and bladder cancer risk, the risk according to quantity and type of alcohol consumed is still not clear. In a recent meta-analysis of these studies, a slightly elevated risk of bladder cancer was demonstrated for men currently drinking alcohol compared with abstainers.¹ In this meta-analysis, however, no data were available to explore the influence of quantity and types of alcoholic beverages consumed. These results prompted us to investigate these associations in more detail in the ongoing Netherlands Cohort Study.

MATERIALS AND METHODS

Cohort. The study design has been described in detail previously.² The study population originated from 204 municipal population registries throughout the Netherlands, and the cohort includes 58,279 men and 62,573 women who were 55-69 years at baseline (1986). We used the case-cohort approach for data processing and analysis.³ A subcohort of 3,500 subjects was randomly sampled from the cohort after baseline exposure measurement and was followed up to obtain vital status information. No subcohort members were lost to follow-up during the follow-up period.

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch database of pathology reports⁴ and follow-up was more than 95 percent complete.⁵ The present analysis was restricted to 6.3 years of follow-up. After we excluded prevalent cases, 3,346 subcohort members and 619 incident cases with microscopically confirmed carcinomas of the bladder, ureters, renal pelvis, or urethra were identified. Because the overwhelming majority of tumors occurred in the bladder, and because the renal pelvis and ureter are covered by the same urothelium, the term bladder cancer was used as a synonym for these neoplasms.

Questionnaire. All subjects completed a self-administered questionnaire on risk factors for cancer. The food-frequency section concentrated on habitual consumption during the year before the study began. Consumption of alcoholic beverages was addressed by questions on beer, red wine, white wine, sherry, and other fortified wines, liqueur, and liquor. The questionnaire data were keyed twice and were processed in a standardized manner blinded with respect to case-subcohort status. The questionnaire has been proven to be valid and reproducible.^{6,7}

Data-analysis. People who drank alcoholic beverages less than once a month were considered nondrinkers. Four items from the questionnaire (i.e., red wine, white wine, sherry, and liqueur) were combined into a wine variable, since these items were substantially correlated and separate treatment would have resulted in a sparsity of data. Mean daily alcohol consumption was calculated by using the computerized Dutch food composition table⁸. On the basis of pilot study data, standard glass sizes were defined as 200 ml for beer, 105 ml for wine, 80 ml for sherry, and 45 ml for both liqueur and liquor, corresponding to 8, 10, 11, 7, and 13 grams of alcohol, respectively.

The following variables were considered as potential confounders: age (years); consumption of coffee (ml/day), water (ml/day), vegetables (g/day), and fruit (g/day); cigarette smoking status (never/former/current), amount (cigarettes/day), and duration (years of cigarette smoking); occupational exposure to dye, rubber, leather, or vehicle fumes (ever/never); and first-degree family history of bladder cancer (yes/no). Incidence rate ratios and corresponding 95% confidence intervals for bladder cancer were estimated by using exponentially distributed failure time regression models.⁹

To ensure that the results were not influenced by changes in exposure for subjects with preclinical disease, we conducted analyses with and without cases diagnosed in the first 1 or 2 years of follow-up. The results from these analyses were similar; therefore, this paper presents results from the analyses in which all cases were included.

Table 1. Age- and smoking-adjusted incidence rate ratios (categorical and continuous analyses) for bladder cancer in men and women, according to total alcohol consumption from alcoholic beverages, Netherlands Cohort Study (1986-1992)

Alcohol consumption	No.of Cases in cohort	No.of Person-years in subcohort	Adjusted for age		Adjusted for age and smoking*	
			RR ^a	95% CI ^a	RR ^a	95% CI ^a
Men						
no alcohol intake	62	1446	1.00	Reference	1.00	Reference
<5 g/day	108	1947	1.43	1.00, 2.05	1.49	1.00, 2.21
5-<15 g/day	136	2646	1.33	0.94, 1.88	1.52	1.04, 2.21
15-<30 g/day	109	2192	1.24	0.86, 1.77	1.16	0.78, 1.71
30+ g/day	102	1324	1.98	1.37, 2.88	1.63	1.08, 2.47
p value for linear trend			<0.01		0.13	
alcohol increment 10g/day			1.09	1.03, 1.15	1.04	0.98, 1.10
Women						
no alcohol intake	25	3147	1.00	Reference	1.00	Reference
<5 g/day	29	3622	1.01	0.59, 1.76	0.97	0.56, 1.69
5+ g/day	23	2979	0.95	0.55, 1.76	0.75	0.41, 1.37
alcohol increment 10g/day			0.97	0.72, 1.32	0.85	0.60, 1.20

*Smoking status, amount, and duration

^aRR, incidence rate ratio; CI, confidence interval

This study was based on 594 cases (517 men and 77 women) and 3,170 subcohort members (1,591 men and 1,579 women) for whom data on alcohol consumption were complete.

RESULTS

Fifteen percent of the men and almost one third of the women were nondrinkers. Most men consumed all types of alcoholic beverages, whereas almost 50% of the women drank wine exclusively.

Of the potential risk factors, sex modified the association between alcohol consumption and bladder cancer incidence. Therefore, the analyses were stratified on sex. The age-adjusted risk for men was higher for alcohol drinkers than for nondrinkers (incidence rate ratio=1.43, 95% confidence interval (CI):1.06, 1.95). As shown in table 1, this risk increased according to the quantity of alcohol consumed (p for trend<0.01). After additional adjustment for cigarette smoking status, amount, and duration, the risk estimates remained practically stable for alcohol drinkers compared with nondrinkers (incidence rate ratio=1.43, 95% CI:1.02, 1.99), although the dose-response relation was found to be lower and not statistically significant (p for trend=0.13). The association between alcohol consumption and bladder cancer risk did not differ within strata of smoking status, amount, or smoking duration (data not shown). There was no indication of an association between alcohol consumption and risk of bladder cancer for women (Table 1). Additional correction for other potential confounders did not change the risk estimates substantially.

Further analyses for specific alcoholic beverages were restricted to men only (Table 2). When nondrinkers were used as the reference, the age- and smoking- adjusted incidence rate ratio for beer drinkers consuming 15-<30g/day of alcohol increased slightly, to 1.70 (95% CI:0.90-3.23), with the quantity of alcohol consumed. Higher intakes from beer were not related to an increased risk. Consumption of wine was not related to an increased risk for men consuming <30 g/day, but men who drank ≥ 30 g/day of alcohol from wine had an increased incidence rate ratio of 1.73 (95% CI:0.74, 4.05) compared with nondrinkers. The highest risk was found for men who drank ≥ 30 g/day from liquor compared with nondrinkers. The corresponding incidence rate ratio was 1.94 (95% CI:1.17, 3.22). Lower intakes from liquor did not seem to be related to an elevated risk.

Table 2. Adjusted incidence rate ratios (age and dose-response analyses) for bladder cancer in men, according to alcohol consumption from 1960 to 1992 (Netherlands Cohort Study (1986-1992))

Alcohol consumption	No. of Cases in cohort	No. of Person-years in subcohort	Adjusted for age		Adjusted for age and smoking*	
			RR [†]	95% CI [†]	RR [†]	95% CI [†]
no alcohol intake	62	1446	1.00	Reference	1.00	Reference
Alcohol from beer						
<5 g/day	174	3383	1.32	0.95, 1.85	1.35	0.94, 1.95
5-<15 g/day	89	1607	1.49	1.02, 2.18	1.44	0.95, 2.18
15-<30 g/day	22	331	1.94	1.07, 3.52	1.70	0.90, 3.23
30+ g/day	10	170	1.45	0.66, 3.20	1.09	0.46, 2.57
p value for linear trend			<0.01		0.12	
alcohol increment 10g/day			1.07	0.96, 1.19	1.03	0.91, 1.16
Alcohol from wine[‡]						
<5 g/day	151	2686	1.38	0.98, 1.94	1.54	1.06, 2.23
5-<15 g/day	67	1417	1.19	0.80, 1.77	1.23	0.80, 1.90
15-<30 g/day	25	581	1.13	0.66, 1.93	1.14	0.65, 2.00
30+ g/day	11	135	2.08	0.94, 4.61	1.73	0.74, 4.05
p value for linear trend			0.24		0.46	
alcohol increment 10g/day			1.00	0.88, 1.15	0.99	0.87, 1.13
Alcohol from liquor						
<5 g/day	114	2207	1.30	0.91, 1.86	1.44	0.98, 2.11
5-<15 g/day	89	1603	1.37	0.94, 2.00	1.38	0.92, 2.08
15-<30 g/day	70	1155	1.41	0.95, 2.10	1.25	0.81, 1.91
30+ g/day	50	512	2.48	1.57, 3.92	1.94	1.17, 3.22
p value for linear trend			<0.01		0.03	
alcohol increment 10g/day			1.14	1.05, 1.23	1.06	0.97, 1.16

*Smoking status, amount, and duration

[†]RR, incidence rate ratio; CI confidence interval

[‡]Among male wine drinkers in the subcohort 63% drank red wine, 58% white wine, 48% sherry, and 16% liqueur

Adjustment for age only or simultaneous inclusion of alcohol consumption from beer, wine, and liquor in one regression model did not change the results substantially. Although some point estimates suggested increased risks for alcohol consumers, practically none of the incidence rate ratios or dose-response trends was statistically significant (Table 2).

DISCUSSION

The results of this prospective study do not suggest an important association between alcohol consumption and bladder cancer risk. If any, the association between alcohol consumption and male bladder cancer is probably small.

Some authors have suggested that residual confounding due to tobacco smoking could explain an increased risk as a result of alcohol drinking.^{10,11} We attempted to model cigarette smoking habits such that they best explained bladder cancer by using smoking status, amount, and duration. However, correction for smoking did not change the incidence rate ratios substantially. Therefore, the association observed between alcohol consumption and bladder cancer risk did not seem to be entirely due to residual confounding by smoking, although some influence cannot be excluded.

A recent meta-analysis based on predominantly case-control studies concluded that alcohol consumption slightly increases male bladder cancer risk (summary odds ratio=1.35 (95% CI:0.91, 2.02)), an estimate that might not be of practical importance.¹ We repeated this meta-analysis to evaluate whether the summary odds ratio for male alcohol consumers compared with nondrinkers remained stable after the present study

was included.¹ We found that the new age- and smoking-adjusted summary odds ratio was 1.35 (95% CI:0.96, 1.91). Associations between specific alcoholic beverages and bladder cancer risk were reported in nine studies without consistent results.¹²⁻²⁰

The literature to date does not support a causal role for alcohol consumption in bladder cancer etiology. Several mechanisms have been postulated to explain ethanol-related carcinogenesis. Ethanol slows down protein synthesis. One consequence is that cell repair mechanisms might be inhibited, which could lead to malignant changes.^{21,22} Furthermore, ethanol might improve permeability of membranes to carcinogens and might enhance carcinogenic activity.²²⁻²⁴ Other explanations include the effect of ethanol on cell proliferation, possibly caused by acetaldehyde.^{22,23,25} Animal experiments have shown that nitrosamines might be carcinogenic in the bladder.^{21,26}

However, the quantities of these carcinogens in alcohol are small,¹⁷ and certain nitrosamines, upon direct instillation into the bladder, do not cause bladder cancer.²⁵ The urogenous-contact hypothesis associates development of bladder cancer with prolonged exposure to carcinogens in urine.^{27,28} High consumption of fluids may reduce this exposure. One prospective study demonstrated this negative association,²⁸ whereas other studies reported positive associations,^{13,29-33} or no association.^{15,34} Correction for total water intake did not change the results substantially.

In accordance with earlier studies, the results of this prospective study do not suggest an important increased risk of bladder cancer for male alcohol consumers. If any such association exists, it is probably small. In our study, we found that the risk of bladder cancer increased slightly according to the quantity of alcohol consumed, irrespective of the type of alcoholic beverage, although no statistically significant dose-response trends were identified. We found no association between alcohol consumption and bladder cancer risk for women.

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REFERENCES

1. Zeegers MPA, Tan FES, Verhagen AP, Weijnenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.
2. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in The Netherlands. *J Clin Epidemiol*. 1990;43:285-95.
3. Barlow WE, Ichikawa L, Rosner D, Izumi S. Analysis of case-cohort designs. *J Clin Epidemiol*. 1999;52:1165-72.
4. van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, Hunen PHM. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol*. 1990;19:553-8.
5. Goldbohm RA, Van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond*. 1994;72:80-4.
6. Goldbohm RA, van den Brandt PA, Brants HA, et al. Validation of a dietary questionnaire used in large-scale prospective cohort study on diet and cancer. *Eur J Clin Nutr*. 1994;48:253-65.
7. Goldbohm RA, van 't Veer P, van den Brandt PA, et al. Reproducibility of a food frequency questionnaire and stability of dietary habits determined from five annually repeated measurements. *Eur J Clin Nutr*. 1995;49:420-9.
8. NEVO-table, Dutch food composition table 1986-1987. The Hague, The Netherlands: Voorlichtingsbureau voor de Voeding. 1986.
9. Volovics A, Van den Brandt PA. Methods for the analyses of case-cohort studies. *Biomet. J*. 1997;2:195-214.
10. Donato F, Boffetta P, Fazioli R, Aulenti V, Gelatti U, Porru S. Bladder cancer, tobacco smoking, coffee and alcohol drinking in Brescia northern Italy. *Eur J Epidemiol*. 1997;13:795-800.

11. Silverman DT, Morrison AS, Devesa SS. Bladder cancer. In: Schottenfeld D, Fraumeni JF, eds. *Cancer epidemiology and prevention*. New York, NY: Oxford University Press, 1996:1156-79.
12. Iscovich J, Castelletto R, Esteve J, et al. Tobacco smoking, occupational exposure and bladder cancer in Argentina. *Int J Cancer*. 1987;40:734-40.
13. Jensen OM, Wahrendorf J, Knudsen JB, Sorensen BL. The Copenhagen case-control study of bladder cancer. II. Effect of coffee and other beverages. *Int J Cancer*. 1986;37:651-7.
14. D'Avanzo B, La Vecchia C, Franceschi S, Negri E, Talamini R, Buttinio I. Coffee consumption and bladder cancer risk. *Eur J Cancer*. 1992;28a:1480-4.
15. Risch HA, Burch JD, Miller AB, Hill GB, Steele R, Howe GR. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol*. 1988;127:1179-91.
16. Thomas DB, Uhl CN, Hartge P. Bladder cancer and alcoholic beverage consumption. *Am J Epidemiol*. 1983;118:720-7.
17. Kunze E, Claude J, Frentzel Beyme R, Paczkowski K, Schneider G, Schubert H. Association of cancer of the lower urinary tract with consumption of alcoholic beverages. A case-control study. *Carcinogenesis*. 1986;7:163-5.
18. Williams RR, Horm JW. Association of cancer sites with tobacco and alcohol consumption and socioeconomic status of patients: interview study from the Third National Cancer Survey. *J Natl Cancer Inst*. 1977;58:525-47.
19. Ross RK, Paganini Hill A, Landolph J, Gerkins V, Henderson BE. Analgesics, cigarette smoking, and other risk factors for cancer of the renal pelvis and ureter. *Cancer Res*. 1989;49:1045-8.
20. Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol*. 1993;3:211-6.
21. Berger A. Science commentary: Why wine might be less harmful than beer and spirits (editorial). *Br Med J*. 1998;317:848.
22. Scitz HK, Simanowski UA. Alcohol and carcinogenesis. *Annu Rev Nutr*. 1988;8:99-119.
23. Garro AJ, Lieber CS. Alcohol and cancer. *Annu Rev Pharmacol Toxicol*. 1990;30:219-49.
24. Freund G. Possible relationships of alcohol in membranes to cancer. *Cancer Res*. 1979;39:2899-901.
25. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol*. 1997;13:291-8.
26. Shirai T. Etiology of bladder cancer. *Semin Urol*. 1993;11:113-26.
27. Jones PA, Ross RK. Prevention of bladder cancer. *N Engl J Med*. 1999;340:1424-26.
28. Michaud DS, Spiegelman D, Clinton SK, et al. Fluid intake and the risk of bladder cancer in men. *N Engl J Med*. 1999;340:1390-97.
29. Kunze E, Chang Claude J, Frentzel Beyme R. Life style and occupational risk factors for bladder cancer in Germany. A case-control study. *Cancer*. 1992;69:1776-90.
30. Claude J, Kunze E, Frentzel Beyme R, Paczkowski K, Schneider J, Schubert H. Life-style and occupational risk factors in cancer of the lower urinary tract. *Am J Epidemiol*. 1986;124:578-89.
31. Slattery ML, West DW, Robison LM. Fluid intake and bladder cancer in Utah. *Int J Cancer*. 1988;42:17-22.
32. Vena JE, Graham S, Freudenheim J, et al. Drinking water, fluid intake and bladder cancer in western New York. *Arch Environ Health*. 1993;48:191-8.
33. Cantor KP, Hoover R, Hartge P, et al. Bladder cancer, drinking water source, and tap water consumption: a case-control study. *J Natl Cancer Inst*. 1987;79:1269-79.
34. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in New Orleans, Louisiana. *J Natl Cancer Inst*. 1968;41:683-709.

8

Are Coffee, Tea and Fluid Consumption associated with Bladder Cancer Risk? Results from the Netherlands Cohort Study

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ABSTRACT

Objectives: Coffee, tea and fluid consumption have been thought to influence bladder cancer incidence. In a large prospective study, these associations were investigated.

Methods: In 1986, cohort members (55-69 years) completed a questionnaire on cancer risk factors. Follow-up was established by linkage to cancer registries until 1992. The multivariable case-cohort analysis was based on 569 bladder cancer cases and 3,123 subcohort members.

Results: The incidence rate ratios (RR) for men consuming <2 cups of coffee/day was 0.89 (95%CI 0.51-1.5) using the median consumption category (4-<5 cups/day) as reference. This RR increased to 1.3 (95%CI 0.94-1.9) for men consuming ≥ 7 cups/day, although no clear dose-response association was found. The RRs decreased from 1.2 (95%CI 0.56-2.7) for women consuming <2 cups coffee/day to 0.36 (95%CI 0.18-0.72) for women consuming ≥ 5 cups/day compared to the median consumption category (3-<4 cups/day). Men and women who abstained from drinking tea had a RR of 1.3 (95%CI 0.97-1.8) compared to those consuming 2-<3 cups of tea per day (median consumption category). The RR for men and women comparing highest to lowest quintile of total fluid consumption was 0.87 (95%CI 0.63-1.2).

Conclusion: The data suggest a possible positive association between coffee consumption and bladder cancer risk in men and a probable inverse association in women. Tea consumption was inversely associated with bladder cancer. Total fluid consumption did not appear to be associated with bladder cancer.

INTRODUCTION

Over the last four decades, several risk factors for bladder cancer, the fourth most common cancer in man, have been investigated. Many epidemiological studies and several reviews have been conducted to investigate determinants of bladder cancer.^{1,2} These studies suggested that bladder cancer is influenced by environmental factors, such as cigarette smoking and exposure to industrial chemicals, and by chronic infections with *Schistosoma Haematobium*. In 1993, Viscoli et al concluded in a meta-analysis of seven studies that coffee consumption might not have a clinically important impact on bladder cancer risk. Recently, an updated meta-analysis on both coffee and tea consumption demonstrated a small elevated risk of bladder cancer for current coffee drinkers and did not identify an association for tea drinkers compared with non-drinkers.³ The results were rather consistent across study design characteristics. In the meta-analysis, however, insufficient data were available to explore the influence of the amount of coffee and tea on bladder cancer risk. Because coffee and tea are so widely used, the association between coffee and tea consumption and the bladder cancer incidence is an important concern. Furthermore, because coffee and tea consumption are accounting for a considerable amount of daily fluid consumption total fluid consumption is of interest, particularly given a recent analysis, which reported a negative association.⁴ These results prompted us to investigate the association between coffee, tea and total fluid consumption and bladder cancer risk in the ongoing Netherlands Cohort Study. In this follow-up study among 120,852 men and women, which started in 1986, detailed information has been collected on dietary habits, including drinking habits, by means of a self-administered questionnaire.

METHODS

Cohort. This population based prospective cohort study on diet and cancer started in the Netherlands in September 1986. The cohort includes 58,279 men and 62,573 women aged 55 to 69 years at baseline. The study population originated from 204 municipal population registries throughout the country. The case cohort approach was used for data processing and analysis.⁵ Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after the baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up, during the follow-up period. The study design has been described in detail previously.⁶

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.⁷ The completeness of cancer follow-up was estimated to be over 96 percent.⁸ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 3,346 subcohort members (1,630 men and 1,716 women) and 619 incident cases (532 men and 87 women) with microscopically confirmed, incident carcinomas of the urinary bladder, ureters, renal pelvis and urethra were identified. Of these cases, 584 (94.3%) were diagnosed with bladder cancer. The morphology of the bladder carcinomas was transitional cell (n=559), squamous cell (n=3), adeno (n=6), mixed (n=1) or not specified (n=15). The morphology of the carcinomas of ureters, renal pelvis and urethra (n=35) were not specified but most probable transitional cell. Because the overwhelming majority of tumours occurred in the urinary bladder and the renal pelvis and ureter are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Questionnaire. At baseline, all cohort members completed a self-administered questionnaire on risk factors for cancer. The semi-quantitative food-frequency section of the questionnaire concentrated on habitual consumption of food and beverages during the year preceding the start of the study. Use of beverages was addressed by questions on the consumption of water (i.e., tap water, mineral water), milk (i.e., milk, buttermilk, cocoa), juice (i.e., tomato, orange, others), soda and lemonade (i.e., cola, others), alcoholic beverages (i.e., beer, red wine, white wine, sherry, port, liqueur, spirits), coffee and tea. The questionnaire covered almost all beverages consumed regularly. The questionnaire data were key entered twice and processed in a standardized manner blinded with respect to case/subcohort status in order to minimize observer bias in coding and data interpretation.

Data-analysis. 50 Cases and 223 subcohort members with incomplete or inconsistent dietary data were excluded, according to criteria described previously.⁹ Therefore, the analyses presented are based on 569 cases (491 men and 78 women) and 3,123 subcohort members (1,525 men and 1,598 women). Among these remaining subjects, questions on whether subjects drank coffee or tea were left blank by 0.5% (coffee) and 1.2% (tea); they were considered to be non-users. Of all included subjects, 0.8% and 2.1% reported drinking coffee or tea, respectively, but did not report how much. They were assumed to drink 4.5 cups of coffee or 3.2 cups of tea per day (i.e., the mean number of cups consumed by the drinkers). The standard size of a cup of coffee or tea was assessed from a pilot study to be 125 ml. The consumption of coffee and tea were categorized into strata based on the distribution in the case and subcohort groups. For categorical analyses, the median consumption stratum was chosen as reference category. We evaluated the total intake of caffeine from caffeine containing beverages (i.e., coffee, tea and cola). A cup of coffee or tea or a glass of cola (175 ml) was assumed to contain respectively 81, 15 and 23 milligrams of caffeine. No questions were asked on decaffeinated coffee consumption, as in the pilot study this item did not appear to be consumed much. Based on questionnaire data we calculated total fluid consumption using information on frequency and beverage specific serving size of the 19 specific beverages. Furthermore, total intake of water from beverages and food was calculated using the computerized Dutch food composition table.¹⁰

Cigarette smoking was considered as a potential confounding factor in the association between coffee/tea consumption and bladder cancer risk since cigarette smoking is one of the most important environmental risk factors for bladder cancer.¹¹ Cigarette smoking was operationalised through questions on current and former cigarette smoking status at baseline (yes *vs.* no), smoking amount (cigarettes/day) and smoking duration (years of cigarette smoking) for current and former smokers. Other variables that were considered as potential confounders in multivariable analysis were: age (years), alcohol intake (g/day), coffee consumption (ml/day), tea consumption (ml/day), total water intake (ml/day), vegetables consumption (g/day), fruit consumption (g/day), current cigarette smoking (yes *vs.* no), smoking amount (cigarettes/day), smoking duration (years of cigarette smoking), occupational exposure to dye, rubber, leather or vehicle fumes (ever *vs.* never), and first degree family history of bladder cancer (yes *vs.* no). Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models¹² with the Stata statistical software package. Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow.¹³ Tests for dose response trends in risk for bladder cancer were assessed by fitting ordinal exposure variables as continuous terms and performing likelihood-ratio tests between regression models with and without these variables. We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against failure time. List wise deletion of subjects with missing data on the primary determinants and confounders was applied in multivariable analyses. Sex modified the association between coffee consumption and the incidence of bladder cancer in multivariable analyses. The RRs concerning tea and total fluid

consumption were similar for men and women. Therefore, we conducted regression analyses for men and women separately for coffee consumption and for men and women combined for tea and total fluid consumption. Subgroup analyses conditional on smoking duration were conducted to evaluate potential coffee, tea or total fluid by smoking interaction.

RESULTS

More than 96 percent of the subjects reported drinking coffee and more than 85 percent reported drinking tea. Most subjects drank both beverages (83%). Coffee was more often consumed solely than tea (13% and 3%, respectively). The mean daily coffee consumption among coffee drinkers was higher for men (cases: 640 ml, subcohort members: 591 ml) than for women (cases: 478 ml, subcohort members: 517 ml). Conversely, tea was more consumed by female tea drinkers (cases: 474 ml, subcohort members: 431 ml) than by male tea drinkers (cases: 367 ml, subcohort members: 378 ml). The association between coffee and tea consumption was negative for cases and subcohort members ($r=-0.30$ and -0.27 , respectively). Coffee and tea consumption accounted on average for 43% and 24% of the total daily fluid consumption for men, 41%, and 30% for women, respectively (data not shown). Table 1 displays the distribution of potential risk factors for bladder cancer with respect to coffee and tea consumption among subcohort members for men and women separately. For this purpose, coffee and tea consumption were dichotomised in low and high consumption categories (lower *vs.* higher than median consumption). Number of cigarettes smoked per day, years of cigarette smoking, consumption of alcohol and total water intake increased with increasing daily coffee consumption and not with tea consumption (except for total water intake) for both men and women. Age and the consumption of vegetables were comparable between low and high coffee and tea consumers. The consumption of fruit was negatively associated with coffee consumption and positively associated with tea consumption. Few subjects (<3%) reported a high risk occupation or having first-degree family members with bladder cancer (Table 1).

Table 1. Distribution of potential risk factors for bladder cancer with respect to coffee and tea consumption and sex among subcohort members, Netherlands Cohort Study (1986-1992)

Risk factors	Coffee consumption (cups/day)				Tea consumption (cups/day)			
	Men		Women		Men		Women	
	<5 n=830	≥5 n=695	<4 n=594	≥4 n=1004	<3 n=851	≥3 n=674	<3 n=697	≥3 n=901
Men								
age (years)	62.0	60.6	61.9	61.0	60.8	62.1	60.9	61.8
alcohol intake (g/day)	13.7	15.6	5.6	5.9	15.9	12.8	5.9	5.6
total water intake (ml/day)	1982	2342	1841	2093	2041	2279	1815	2142
vegetable consumption (g/day)	189	195	196	197	188	197	192	200
fruit consumption (g/day)	160	147	205	191	144	168	187	203
smoking amount (cigt/day)*	15.2	18.8	9.9	11.8	17.8	34.3	12.7	9.8
smoking duration (years)*	32.0	35.0	28.4	27.6	15.8	32.3	29.8	26.1
coffee cons. (cups/day)	3.0	6.5	2.1	5.1	5.0	4.0	4.4	3.6
tea cons. (cups/day)	3.0	2.1	3.5	2.8	1.2	4.3	1.3	4.5
Percentage								
non-smokers	17.2	8.8	66.0	54.9	12.0	15.1	54.4	62.6
high risk occupation†	0.8	0.6	0.2	0.3	0.7	0.7	0.1	0.3
positive family history of bladder cancer	1.6	1.3	2.5	2.2	1.4	1.5	2.4	2.2

*among cigarette smokers only
†ever exposed to dye, rubber, leather, and vehicle fumes

As is shown in table 2, the age- and smoking-adjusted RRs for men consuming less than two cups of coffee per day was 0.89 (CI:0.51-1.54) compared to subjects drinking 4-5 cups of coffee per day (the median consumption category). This RR increased with increasing coffee consumption to 1.3 (CI:0.94-1.9) for men consuming seven or more cups per day. However, none of the individual RRs were statistically significant. Although no clear dose-response association was found, the test for linear trend was statistically significant (p -trend<0.01). There was no indication of a positive association between coffee consumption and bladder cancer risk for women. On the contrary, for women, the age- and smoking-adjusted RRs decreased by increasing coffee consumption from 1.2 (CI:0.56-2.7) for women drinking less than two cups of coffee per day to 0.36 (CI:0.56-2.7) for women consuming five or more cups per day, compared to the median consumption category (3-<4 cups/day) (p -trend<0.01). When coffee consumption was modelled as a continuous term in the analyses we also found a reduced risk for women per cup of coffee consumed per day. Simultaneous inclusion of both coffee and tea consumption in one regression model (Table 2) or additional correction for current cigarette smoking, alcohol intake, total water intake, vegetables consumption, fruit consumption, occupational exposure and family history of bladder cancer did not essentially change the risk estimates for coffee consumption (data not shown).

Table 2. Adjusted incidence rate ratios (RR) for bladder cancer according to coffee consumption in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Coffee consumption (cups/day)	Cases in cohort	Person years in subcohort	RR (95%CI) ^a	RR (95%CI) ^a	RR (95%CI) ^b
Men					
0-<2	23	628	0.83 (0.50-1.36)	0.89 (0.51-1.54)	0.92 (0.53-1.60)
2-<3	32	907	0.69 (0.44-1.07)	0.72 (0.45-1.13)	0.74 (0.47-1.17)
3-<4	61	961	1.24 (0.86-1.79)	1.27 (0.87-1.87)	1.27 (0.86-1.87)
4-<5	119	2463	1.00 (reference)	1.00 (reference)	1.00 (reference)
5-<6	72	1432	1.12 (0.80-1.58)	0.98 (0.68-1.40)	0.96 (0.67-1.38)
6-<7	91	1456	1.43 (1.04-1.98)	1.25 (0.89-1.76)	1.19 (0.84-1.68)
≥7	93	1302	1.76 (1.27-2.44)	1.33 (0.94-1.90)	1.25 (0.87-1.78)
p-value for linear trend			<0.01	<0.01	0.06
coffee increment, 1 cup/day			1.09 (1.05-1.13)	1.04 (1.00-1.09)	1.03 (0.98-1.08)
Women					
0-<2	11	773	1.11 (0.52-2.40)	1.23 (0.56-2.73)	1.16 (0.51-2.62)
2-<3	13	1293	0.78 (0.38-1.60)	0.84 (0.40-1.76)	0.82 (0.39-1.72)
3-<4	20	1573	1.00 (reference)	1.00 (reference)	1.00 (reference)
4-<5	17	3144	0.43 (0.22-0.83)	0.44 (0.22-0.86)	0.44 (0.22-0.86)
≥5	17	3064	0.44 (0.23-0.86)	0.36 (0.18-0.72)	0.37 (0.19-0.72)
p-value for linear trend			<0.01	<0.01	<0.01
coffee increment, 1 cup/day			0.87 (0.75-1.01)	0.83 (0.72-0.96)	0.84 (0.72-0.97)

^aadjusted for age (years)
^aadjusted for age (years), number of cigarettes/day, and years of cigarette smoking
^badjusted for age (years), number of cigarettes/day, years of cigarette smoking, and tea consumption (ml/day)

Tea consumption was inversely associated with bladder cancer risk, irrespective of the number of cups consumed per day (Table 3). Men and women consuming 0, <2, 3-<4, 4-<5 and ≥5 cups of tea per day had age-, sex-, and smoking-adjusted RRs of 1.3 (CI:0.97-1.8), 0.95 (CI:0.67-1.3), 0.81 (CI:0.58-1.1), 0.81 (CI:0.59-1.1) and 0.93 (CI:0.66-1.3) compared to the median consumption category (2-<3 cups/day), respectively (p -trend<0.01). The risk of bladder cancer decreased by five percent for every increment of one cup of tea per day in continuous analysis. Neither adjustment for coffee consumption (Table 3) nor adjustment for current cigarette smoking and other potential risk factors changed these results. Because coffee consumption for women and tea consumption for men and women appeared to be inversely associated with bladder cancer risk, we investigated the combined effect of coffee and tea consumption. We found

no significant interaction effect between coffee and tea consumption on bladder cancer risk for men and women (data not shown).

Table 3. Adjusted incidence rate ratios (RR) for bladder cancer according to tea consumption in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Tea consumption (cups/day)	Cases in cohort	Person years in subcohort	RR (95%CI) ^a	RR (95%CI) ^b	RR (95%CI) ^c
no tea consumption	115	2422	1.45 (1.09-1.94)	1.31 (0.97-1.78)	1.30 (0.96-1.78)
<2	70	2318	0.96 (0.69-1.33)	0.95 (0.67-1.34)	0.95 (0.67-1.34)
2-<3	155	4625	1.00 (reference)	1.00 (reference)	1.00 (reference)
3-<4	71	2914	0.80 (0.58-1.10)	0.81 (0.58-1.13)	0.81 (0.58-1.13)
4-<5	80	3631	0.69 (0.51-0.94)	0.81 (0.59-1.12)	0.81 (0.59-1.12)
5+	78	3086	0.88 (0.64-1.20)	0.93 (0.66-1.29)	0.93 (0.66-1.30)
p-value for linear trend			<0.01	<0.01	<0.01
tea increment, 1 cup/day			0.92 (0.87-0.97)	0.95 (0.89-1.00)	0.95 (0.90-1.00)

^aadjusted for age (years) and sex

^badjusted for age (years), sex, number of cigarettes/day, and years of cigarette smoking

^cadjusted for age (years), sex, number of cigarettes/day, years of cigarette smoking, and coffee consumption (ml/day)

Most of the daily caffeine intake was derived from coffee (85.0%) and tea (14.5%) consumption. For men, the age- and smoking-adjusted risk of bladder cancer increased with increasing caffeine intake to 1.5 (CI:1.0-2.1) comparing highest to lowest quintile of caffeine intake (p -trend=0.03) (table 4). For women, conversely, a negative dose response trend was found (p -trend<0.01). The RRs for women in the highest quintile of caffeine intake was 0.50 (CI:0.25-1.0) compared to women in the lowest consumption quintile (p -trend<0.01) (Table 4).

Table 4. Adjusted incidence rate ratios (RR) for bladder cancer according to caffeine intake in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Caffeine Quintiles	Men		Women	
	Range (Median) in mg/day	RR* (95%CI)	Range (Median) in mg/day	RR* (95%CI)
1 (low)	0-274 (208)	1.00 (reference)	0-252 (193)	1.00 (reference)
2	275-357 (340)	1.25 (0.87-1.80)	253-340 (289)	0.95 (0.51-1.76)
3	358-436 (400)	1.14 (0.79-1.63)	341-385 (370)	0.59 (0.29-1.19)
4	437-520 (488)	1.23 (0.85-1.78)	386-487 (430)	0.31 (0.12-0.79)
5 (high)	521-2438 (629)	1.47 (1.03-2.12)	488-1309 (543)	0.50 (0.25-1.00)
p-value for linear trend		0.03		<0.01
increment, 100mg/day		1.04 (0.98-1.10)		0.80 (0.67-0.96)

*adjusted for age (years), number of cigarettes/day and years of cigarette smoking

Neither total fluid consumption from beverages only nor total water intake from food and beverages combined was associated with the incidence of bladder cancer (Table 5). None of the age-, sex- and smoking-adjusted risks for subjects categorized by quintiles of consumption differed significantly from the lowest consumption quintile. The RRs per 100 ml increment per day were around unity (all p -trends>0.70). The results for total fluid consumption and total water intake persisted after additional adjustment for coffee and tea consumption (Table 5) or other potential risk factors for bladder cancer (data not shown).

Table 5 Incidence rate ratios (RR) for bladder cancer according to total fluid consumption (from beverages) and total water intake (from food and beverages) in categorical and continuous analyses, Netherlands Cohort Study (1986-1992)

Quintile	Total fluid consumption			Total water intake		
	Range in mg/day	RR (95%CI)*	RR (95%CI)†	Range in mg/day	RR (95%CI)*	RR (95%CI)†
1 (low)	12-984	1.00 (reference)	1.00 (reference)	340-1686	1.00 (reference)	1.00 (reference)
2	985-1181	0.81 (0.54-1.21)	0.83 (0.55-1.25)	1687-1916	0.78 (0.56-1.09)	0.80 (0.57-1.11)
3	1181-1372	0.70 (0.47-1.07)	0.74 (0.48-1.13)	1917-2130	0.82 (0.59-1.14)	0.86 (0.61-1.21)
4	1373-1614	0.97 (0.66-1.43)	1.04 (0.69-1.56)	2131-2435	0.86 (0.62-1.18)	0.91 (0.63-1.30)
5 (high)	1615-4647	0.87 (0.63-1.21)	0.91 (0.65-1.29)	2436-5352	0.88 (0.64-1.21)	0.94 (0.64-1.39)
p-value for linear trend		0.99	0.77		0.70	0.88
increment, 100 ml/day		1.00 (0.97-1.03)	1.01 (0.98-1.05)		0.99 (0.97-1.01)	1.00 (0.97-1.03)

*adjusted for age (years), sex, number of cigarettes/day, and years of cigarette smoking

†adjusted for age (years), sex, number of cigarettes/day, years of cigarette smoking, coffee consumption (ml/day), and tea consumption (ml/day)

Almost none of the RRs were notably altered after adjustment for smoking. Cigarette smoking duration, however, was found to be a statistically significant effect modifier in the association between bladder cancer and coffee and tea consumption, (p -interaction coffee<0.01, p -interaction tea=0.05) (data not shown). Coffee consumption was inversely associated with bladder cancer risk for men smoking cigarettes for less than 20 years, although not statistically significant. Never smokers and men who had smoked cigarettes over a longer period than 20 years had increasing risks of bladder cancer with increasing coffee consumption. An inverse association between tea consumption and bladder cancer risk was most pronounced in men and women who smoked cigarettes for less than 20 years and more than 40 years. The results for total fluid consumption were similar for different classifications of smoking duration (p -interaction=0.24). These stratum specific results, however, are based on small numbers (data not shown).

DISCUSSION

The results of this prospective cohort study support a possible positive association between coffee consumption and male bladder cancer, mainly confined to the higher consumption categories (i.e., ≥ 7 cups/day). For women, a inverse association was found. Tea consumption was inversely associated with bladder cancer for both men and women, irrespective of consumption frequency. The results do not suggest an association between total fluid consumption and bladder cancer.

One strength of this and other prospective studies is that exposure was assessed before diagnosis of bladder cancer. Therefore, recall bias is not likely to have influenced our results. Furthermore, selection bias is not likely, because the follow-up of cases and subcohort members was almost complete.^{8,14} The measurement of coffee, tea and total fluid intake has not been validated. However, subjects with incomplete or inconsistent food frequency questionnaires were excluded from analyses.⁹ The relatively large number of bladder cancer cases in this study was an other important strength. To ensure that the results were not influenced by changes in exposure by subjects with preclinical bladder cancer, we conducted analyses with and without cases diagnosed in the first one or two years of follow-up. Results from these analyses were similar.

A potentially more realistic threat to the interpretation of the observed associations is residual confounding by risk factors for bladder cancer that happen to be associated with coffee, tea or total fluid consumption. Multivariable analyses showed that the estimates of the RRs for coffee, tea and total fluid consumption were not confounded by current cigarette smoking, total consumption of alcohol, coffee (for tea and total fluid analyses), tea (for coffee and total fluid analyses), water, vegetables and fruit, high risk occupation and positive family history of bladder cancer. Age, sex, smoking amount and smoking duration were found to be important determinants. We attempted to model cigarette

smoking habits such that they best explained bladder cancer. This resulted in a model including number of years smoked and habitual number of cigarettes smoked per day, both as continuous variables. When we added the smoking variables to an age and sex (for tea and total fluid analyses)-adjusted model, the RR estimates changed only slightly. We therefore believe that the associations observed were not entirely due to residual confounding by smoking, although we cannot exclude some influence due to misclassification of the smoking variables.

The mean coffee and tea consumption measured in this study is similar to the mean intake measured in previous food consumption surveys conducted in the Netherlands.^{15,16} In accordance with previous studies,³ our results suggested a positive association between coffee consumption and male bladder cancer risk. Although no clear dose-response association was found, the test for linear trend was strongly statistically significant. Therefore, we expect the observed non-linear association to be the result of chance. For women, we found an inverse association between coffee consumption and bladder cancer risk, which was statistically significant. At present, the biological mechanism that might explain this discrepancy between men and women remains unclear and needs further investigation. Previous epidemiological studies on coffee consumption associated with bladder cancer, were primarily based on case-control data, although three prospective studies have been conducted.^{4,17,18} In a recent meta-analysis based on these studies, we concluded that coffee consumption might increase the risk of urinary tract cancer slightly.³

Tea consumption was inversely associated with bladder cancer risk for both men and women. Evidence from animal studies shows that certain compounds present in tea may have inhibitory effects on bladder tumour formation and growth. This inhibitory activity is believed to be mainly due to the antioxidative and possibly antiproliferative effects of polyphenol compounds (e.g., flavonoids).¹⁹⁻²¹ It has been pointed out that no other agent tested for possible chemoprevention effects in animal models has elicited such strong activity as tea and its components at concentrations usually consumed by humans.²¹ In the present study, we found an inverse association between tea intake and bladder cancer risk. Lack of a positive association of tea consumption is consistent with the results of two previous prospective studies^{4,22} and most case control studies.³

An inverse association between tea consumption and bladder cancer risk might be explained by the hypothesis that drinking more fluids is associated with a decreased risk of bladder cancer.^{4,24} The urogenous-contact hypothesis associates the development of bladder cancer with prolonged exposure to carcinogens in urine.^{4,25} High consumption of fluids may reduce this exposure by diluting the urine and reducing contact time through increased frequency of urination. However, total fluid consumption has also been hypothesized to increase the risk of bladder cancer by increasing the workload of the bladder or by exposing the bladder to specific fluids that contain carcinogens (e.g., in coffee or alcohol) or chemopreventive agents (e.g., in tea).^{26,27} Neither total fluid consumption nor total water intake were associated with bladder cancer risk in the present study. Previous studies showed mixed results. Some case control studies reported positive associations between total fluid consumption and bladder cancer,²⁶⁻³² whereas others found no association.^{33,34} Only one case control study reported an inverse association between total fluid consumption and bladder cancer risk in women, but not in men.³⁵ Only two follow-up studies to date were conducted to investigate this association.^{4,17} One follow-up study did not find an association between fluid consumption and bladder cancer.¹⁷ Recently, a preventive effect of total fluid consumption was presented in an other follow-up study.⁴ Although the weight of evidence is not in favour of a protective effect of fluid intake, further research is needed to evaluate this matter.

In conclusion, the results of this prospective study suggest a possible positive association between coffee consumption and bladder cancer risk for men. Men consuming more than seven cups per day were found to have the highest risk of bladder cancer. For women a probable inverse association was found. Tea was inversely associated with bladder cancer for both men and women. The authors found no evidence for an association between total fluid consumption and bladder cancer risk.

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REFERENCES

1. World Cancer Research Fund, American Institute for Cancer Research. Bladder. Food, Nutrition and the Prevention of Cancer: a global perspective. Menasha: Banta Book Group: 1997;338-361.
2. van der Meijden APM. Bladder cancer. *Br Med J*. 1998;317:1366-1369.
3. Zeegers MPA, Tan FES, Goldbohm RA, van den Brandt PA. Are coffee and tea consumption associated with urinary tract cancer risk. A systematic review and meta-analysis. *Int J Epidemiol*. 2001; in press
4. Michaud DS, Spiegelman D, Clinton SK, et al. Fluid intake and the risk of bladder cancer in men. *N Engl J Med*. 1999;340:1390-1397.
5. Barlow WE, Ichikawa L, Rosner D, Izumi S. Analysis of case-cohort designs. *J Clin Epidemiol*. 1999;52:1165-1172.
6. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol*. 1990;43:285-295.
7. van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, Hunen PHM. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol*. 1990;19:553-558.
8. Goldbohm RA, van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond*. 1994;72:80-84.
9. Goldbohm RA, van den Brandt PA, Brants HA, et al. Validation of a dietary questionnaire used in large-scale prospective cohort study on diet and cancer. *Eur J Clin Nutr*. 1994;48:253-265.
10. NEVO-table, Dutch food composition table 1986-1987. The Hague, Netherlands: Voorlichtingsbureau voor de Voeding, 1986.
11. Zeegers MPA, Tan FES, Dorant ED, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer*. 2000;89:630-639.
12. Volovics A, van den Brandt PA. Methods for the analyses of case-cohort studies. *Biom. J*. 1997;2:195-214.
13. Barlow WE. Robust variance estimation for the case-cohort design. *Biometrics*. 1994;50:1064-1072.
14. van den Brandt PA, van het Veer P, Goldbohm RA. A prospective cohort study on dietary fat and the risk of postmenopausal breast cancer. *Cancer Res*. 1993;53:75-82.
15. Aarnink EJM, Kistemaker C. Het gebruik van koffie en thee, uitgedrukt in penetratie en volume, voor verschillende groepen uit de Nederlandse bevolking: Voedselconsumptiepeiling 1987-1988. Zeist: TNO-Voeding, 1989.
16. Grundemann RWM. Het gebruik van alcohol, tabak en koffie onder de werkende bevolking in Nederland. Leiden: Nederlands Instituut voor Praeventieve Gezondheidszorg, 1990.
17. Mills PK, Beeson WL, Phillips RL, Fraser GE. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol*. 1991;133:230-239.
18. Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol*. 1993;3:211-216.
19. Yang CS, Wang ZY. Tea and cancer. *J Natl Cancer Inst*. 1993;85:1038-1049.
20. Goldbohm RA, Hertog MG, Brants HA, van Poppel G, van den Brandt PA. Consumption of black tea and cancer risk: a prospective cohort study. *J Natl Cancer Inst*. 1996;88:93-100.
21. Dreosti IE, Wargovich MJ, Yang CS. Inhibition of carcinogenesis by tea: the evidence from experimental studies. *Crit Rev Food Sci Nutr*. 1997;37:761-770.
22. Katiyar SK, Mukhtar H. Tea consumption and cancer. *World Rev Nutr Diet*. 1996;79:154-184.
23. Heilbrun LK, Nomura A, Stemmermann GN. Black tea consumption and cancer risk: a prospective study. *Br J Cancer*. 1986;54:677-683.
24. Charatan F. Fluid intake affects the risk of bladder cancer in men. *Br Med J*. 1999; 318: 1372.
25. Jones PA, Ross RK. Prevention of Bladder Cancer. *N Engl J Med*. 1999;340:1424-1426.
26. Bianchi GD, Cerhan JR, Parker AS, et al. Tea consumption and risk of bladder and kidney cancers in a population-based case-control study. *Am J Epidemiol*. 2000;151:377-383.
27. Slattery ML, West DW, Robison LM. Fluid intake and bladder cancer in Utah. *Int J Cancer*. 1988;42:17-22.
28. Jensen OM, Wahrendorf J, Knudsen JB, Sorensen BL. The Copenhagen case-control study of bladder cancer. II. Effect of coffee and other beverages. *Int J Cancer*. 1986;37:651-657.
29. Kunze E, Chang Claude J, Frentzel Beyme R. Life style and occupational risk factors for bladder cancer in Germany. A case-control study. *Cancer*. 1992;69:1776-1790.

30. Claude J, Kunze E, Frentzel Beyme R, Paczkowski K, Schneider J, Schubert H. Life-style and occupational risk factors in cancer of the lower urinary tract. *Am J Epidemiol.* 1986;124:578-589.
31. Vena JE, Graham S, Freudenheim J, et al. Drinking water, fluid intake and bladder cancer in western new york. *Arch Environ Health.* 1993;48:191-198.
32. Cantor KP, Hoover R, Hartge P, et al. Bladder cancer, drinking water source, and tap water consumption: a case-control study. *J Natl Cancer Inst.* 1987;79:1269-1279.
33. Risch HA, Burch JD, Miller AB, Hill GB, Steele R, Howe GR. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol.* 1988;127:1179-1191.
34. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in New Orleans, Louisiana. *J Natl Cancer Inst.* 1968;41:683-709.
35. Wilkens LR, Kadir MM, Kolonel LN, Nomura AM, Hankin JH. Risk factors for lower urinary tract cancer: the role of total fluid consumption, nitrites and nitrosamines, and selected foods. *Cancer Epidemiol Biomarkers Prev.* 1996;5:161-166.

9

Consumption of Vegetables and Fruits and Bladder Cancer incidence: a Prospective Study

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ABSTRACT

Introduction: It has been suggested that vegetable and fruit consumption may protect against cancer at various anatomical sites. However, few detailed results concerning bladder cancer have been reported so far.

Methods: The association between the consumption of 21 vegetables and nine fruits and bladder cancer risk was assessed in the Netherlands Cohort Study among 120,852 men and women aged 55-69 years at baseline in 1986. After 6.3 years of follow-up 538 incident cases with complete vegetable data and 569 cases with complete fruit data were available for analyses.

Results: In multivariable case-cohort analyses, the following age, sex, and smoking adjusted incidence rate ratios for groups of vegetable and fruit consumption were observed (comparing highest *vs.* lowest quintile of consumption): total vegetables, 0.91 (95% confidence interval [CI]:0.65-1.27); cooked vegetables, 0.98 (CI:0.71-1.35); raw vegetables, 1.10 (CI:0.78-1.53); cooked leafy vegetables, 0.89 (CI:0.65-1.23); raw leafy vegetables, 0.94 (CI:0.73-1.22); pulses, 1.03 (CI:0.74-1.44); brassicas, 0.75 (CI:0.54-1.04); allium vegetables 0.89 (CI:0.67-1.19); total fruit, 0.74 (CI:0.53-1.04) and citrus fruit, 0.85 (CI:0.62-1.17). For three separate items (cauliflower, cooked carrots and mandarins) a statistically significant inverse association was seen, whereas for other specific vegetables or fruit no statistically significant positive association was observed.

Conclusions: Total vegetable consumption was not associated with bladder cancer risk, whereas the consumption of brassicas, total fruit and citrus fruit might be inversely associated with bladder cancer risk.

INTRODUCTION

Bladder cancer is the most common urological cancer and is the seventh most common cancer among men, accounting for approximately 200,000 new cases per year worldwide.¹ Over the last four decades, many epidemiological studies and several reviews have been conducted to investigate determinants of bladder cancer.²⁻⁶ These studies suggested that bladder cancer is influenced by environmental factors, including cigarette smoking, fluid consumption, schistosomal infections and exposure to industrial chemicals (e.g., aromatic amines) and diet.

Among the dietary factors, it has been suggested that vegetable and fruit consumption may protect against cancer of at various anatomical sites. With respect to bladder cancer, most epidemiological studies support a possible protective effect of vegetable⁷⁻¹¹ and fruit consumption,^{7,9,12,13} although some studies have reported no association for vegetable^{13,14} or fruit consumption.^{8,14} Previous studies also have been inconsistent with regard to which vegetables and fruits may be responsible for an inverse association.

Some possible anticarcinogenic mechanisms of substances in vegetables and fruits include antioxidant effects, effects on cell differentiation, increased activity of enzymes that detoxify carcinogens, blocked formation of nitrosamines, preserved integrity of intracellular matrixes, effects on DNA methylation, maintenance of normal DNA repair, increased apoptosis of cancer cells and, decreased cell proliferation.¹⁵ From a biological viewpoint however, it is not yet clear whether the total amount of consumed vegetables and fruits is relevant for the possible protection against cancer or only the consumption of specific vegetables.

The Netherlands Cohort Study (NLCS) with 6.3 years of follow-up and with substantially more incident bladder cancer cases available than in previous studies provided the opportunity to study prospectively detailed relationships between the total amount of vegetable and fruit consumption as well as the consumption frequency of specific vegetables and fruits and bladder cancer incidence for men and women consuming a Dutch diet.

MATERIALS AND METHODS

Cohort. This population based prospective cohort study on diet and cancer started in the Netherlands in September 1986. The cohort includes 58,279 men and 62,573 women aged 55 to 69 years at baseline. The study population originated from 204 municipal population registries throughout the country. The case cohort approach was used for data processing and analysis.¹⁶ Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up, during the follow-up period. The study design, including data collection strategies, has been described in detail previously.¹⁷

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.¹⁸ The completeness of cancer follow-up was estimated to be over 95 percent.¹⁹ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 3,346 subcohort members (1,630 men and 1,716 women) and 619 incident cases (532 men and 87 women) with microscopically confirmed, incident carcinomas of the urinary bladder, ureters, renal pelvis or urethra were identified. Of these cases, 584 (94.3%) were diagnosed with bladder cancer of which 559 (95.7%) were transitional cell carcinomas. Because the overwhelming majority of tumors occurred in the urinary bladder and the ureters, renal pelvis and urethra are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Exposure status. At baseline, the cohort members completed a mailed, self-administered questionnaire on risk factors for cancer. Usual consumption of food and beverages during the year preceding the start of the study was assessed with a 150-item semi quantitative food frequency questionnaire. Participants were asked to report their consumption frequency of 21 vegetables and nine fruits (Table 1). Categories ranged from never or less than once per month to '3-7 times per week' (vegetables) or '6-7 times per week' (fruits). For onions, tomatoes, sweet peppers, and mushrooms participants were asked to report the number they usually ate per week (onions, tomatoes) or month (sweet peppers, mushrooms (in boxes of 250 g)). Frequency of vegetable consumption was asked separately for summer and winter, from which a combined consumption frequency was calculated. Usual serving sizes were asked for string beans and cooked endive only, the mean of which served as an indicator for serving sizes of all cooked vegetables. This procedure was chosen because in a pilot study it was shown that serving sizes of different types of cooked vegetables were correlated, although not strongly, within subjects. To derive an individual serving size for each type of vegetable the indicator serving size was multiplied with a type-specific factor calculated from the same pilot study data as the ratio of the means of the specific to the indicator serving sizes. The frequencies and amounts with regard to fruit consumption have been converted to consumption in grams per day using standard portion sizes. The choice of items for inclusion in the questionnaire was such that it covered almost all vegetables and fruits eaten regularly, with the exception of chicory, red cabbage and cucumber. An open-ended question on other foods eaten on a regular basis was also included. Participants could write down how many times per week they ate such a food and how much they were used eating on each occasion. The questionnaire has been validated against a 9-day diet record. The spearman correlation coefficient was 0.38 and 0.60 for total vegetable and total fruit consumption, respectively.²⁰

Statistical analyses. Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models²¹ with the Stata statistical software package.²² Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow²³ and Lin and Ying.²⁴ We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against failure time.

The following variables were subsequently considered as potential confounders based on earlier analyses²⁵⁻²⁸ age (years), consumption of alcohol (g/day), coffee (ml/day), tea (ml/day), total water (ml/day), current cigarette smoking (yes/no), cigarette smoking amount (cigarettes/day), cigarette smoking duration (years of cigarette smoking), occupational exposure to dye, rubber, leather or vehicle fumes (ever/never), and first degree family history of bladder cancer (yes/no). Those variables that showed a more than ten percent influence on the risk of bladder cancer when considered in a multivariable model were included as covariates in multivariable analyses.

Subjects with incomplete or inconsistent dietary data were excluded from analyses.²⁰ In order to check the quality and consistency of the responses on the several vegetable questions, a specific error index was computed. When the error index exceeded a certain value, i.e. more than three errors, subjects were excluded from analyses on vegetable consumption. Finally, a total of 538 bladder cancer cases (463 men and 75 women) and 2,953 subcohort members (1,456 men and 1,497 women) remained for analyses of vegetable data and 569 bladder cancer cases (491 men and 78 women) and 3,123 subcohort members (1,525 men and 1,598 women) for analyses on fruit data.

We first evaluated combined vegetable and fruit consumption, total vegetables, prepared and raw vegetables categorized in botanical groups, total fruits and citrus fruits, after categorizing subjects into quintile or tertile levels of consumption depending on the distribution in the subcohort. Cases detected during the first and the first two years of follow-up were excluded from analyses to evaluate whether preclinical symptoms might have influenced the results. Specific vegetables and fruits were evaluated as continuous

variables (g/day) and their multivariable RRs were expressed per increment of 25 g/day, corresponding to a consumption frequency of a vegetable approximately once per week. The independent contribution of each specific vegetable or fruit was assessed by an analysis in which total vegetable and fruit consumption was included in the model simultaneously. Tests for trend in the rate ratios were based on likelihood ratio tests. Rate ratios for quintiles of vegetable and fruits consumption were also computed separately in strata of smoking status and amount. For the same variables, subgroup analyses were performed after stratification by tumor morphology and invasiveness.

RESULTS

Table 1 shows the mean daily consumption of vegetables and fruits for bladder cancer cases and subcohort members for men and women separately. The consumption of total vegetables and cooked vegetables was higher for men than for women, whereas the consumption of raw vegetables and fruits was higher for women. Fruit consumption was higher in the subcohort than in the case group. Vegetable consumption was also higher in the subcohort, but only for women (Table 1).

Mean daily consumption of vegetables and fruits was calculated for categories of the most important potential confounders (Table 2). Age, coffee consumption, current cigarette smoking, cigarette smoking amount, cigarette smoking duration, high risk occupation and family history of bladder cancer were not associated with vegetable consumption, whereas alcohol, tea and total water consumption were positively associated. Concerning fruit consumption, we found no association with age, high risk occupation and family history of bladder cancer; negative associations were found with alcohol consumption, coffee consumption, current cigarette smoking, cigarette smoking amount and cigarette smoking duration; and positive associations were found with tea consumption and total water consumption. Vegetable and fruit consumption were positively associated with each other (Table 2). Despite these distributions of vegetable and fruit intake across potential confounders, only age, sex, cigarette smoking amount and cigarette smoking duration appeared to be the most influential confounders in multivariable analyses.

Incidence rate ratio estimates for quintiles or tertiles of consumption are presented in table 3 for total vegetable and fruit consumption and the consumption of groups of vegetables and fruits. The age- and sex-adjusted incidence rate ratio showed a negative association between the combined consumption of vegetables and fruits and bladder cancer risk, which disappeared after additional correction for cigarette smoking amount and cigarette smoking duration. Of all vegetable groups, only an increased consumption of brassicas was associated with a decreased bladder cancer risk (RR 0.75, CI 0.54-1.04, comparing extreme consumption quintiles), although the linear dose response trend was borderline statistically significant (p -trend=0.06). Both total fruit consumption and the consumption of citrus fruits were inversely associated with bladder cancer risk (RR 0.74, CI 0.53-1.04 (p -trend=0.02) and RR 0.85, CI 0.62-1.17 (p -trend=0.22)), respectively, comparing extreme consumption quintiles. Additional adjustment for cigarette smoking amount, cigarette smoking duration, total fruit consumption (for vegetable items) total vegetable consumption (for fruit items) (Table 3) or other potential confounders did not change the results essentially.

Table 1. Mean ($\pm$ sd) daily vegetable and fruits consumption in male and female bladder cancer cases and subcohort members, Netherlands Cohort Study (1986-1992)

Vegetable/fruit consumption (g/day)	Men		Women	
	Cases	Subcohort	Cases	Subcohort
Vegetables	<i>n</i> =463	<i>n</i> =1456	<i>n</i> =75	<i>n</i> =1497
Total vegetables	186.4 ($\pm$ 74.2)	187.1 ($\pm$ 76.3)	175.1 ($\pm$ 73.2)	191.0 ($\pm$ 74.5)
Cooked vegetables	150.8 ($\pm$ 62.5)	150.8 ($\pm$ 63.1)	133.4 ($\pm$ 62.1)	149.5 ($\pm$ 59.3)
Raw vegetables	35.6 ($\pm$ 27.5)	36.2 ($\pm$ 29.0)	41.7 ($\pm$ 25.9)	41.5 ($\pm$ 29.8)
Legumes*	36.0 ($\pm$ 24.3)	34.7 ($\pm$ 23.0)	29.4 ($\pm$ 19.7)	30.7 ($\pm$ 20.7)
String/French beans	21.4 ($\pm$ 16.7)	20.5 ($\pm$ 15.3)	17.6 ($\pm$ 12.8)	19.5 ($\pm$ 14.7)
Broad beans	5.5 ($\pm$ 7.8)	4.7 ($\pm$ 7.2)	4.9 ($\pm$ 7.5)	4.3 ($\pm$ 6.6)
Brassicas	32.6 ($\pm$ 21.3)	32.7 ($\pm$ 20.3)	25.4 ($\pm$ 17.5)	31.6 ($\pm$ 20.0)
Brussels sprouts	8.0 ($\pm$ 7.3)	7.7 ($\pm$ 6.7)	6.5 ($\pm$ 5.1)	7.7 ($\pm$ 7.4)
Cauliflower	14.0 ($\pm$ 10.1)	14.6 ($\pm$ 11.1)	11.1 ($\pm$ 9.1)	13.9 ($\pm$ 10.5)
Cabbage (white/green)	7.3 ($\pm$ 10.2)	7.2 ($\pm$ 8.2)	5.3 ($\pm$ 6.4)	6.9 ($\pm$ 8.0)
Kale	3.3 ($\pm$ 3.7)	3.3 ($\pm$ 3.4)	2.6 ($\pm$ 2.7)	3.2 ($\pm$ 3.4)
Leafy vegetables, cooked	21.9 ($\pm$ 17.0)	21.6 ($\pm$ 16.0)	18.8 ($\pm$ 16.0)	21.3 ($\pm$ 14.9)
Spinach	9.8 ($\pm$ 8.8)	9.6 ($\pm$ 8.9)	7.4 ($\pm$ 10.2)	9.4 ($\pm$ 8.3)
Endive	12.0 ($\pm$ 11.7)	12.0 ($\pm$ 10.8)	11.4 ($\pm$ 9.9)	11.8 ($\pm$ 10.2)
Leafy vegetables, raw	9.4 ($\pm$ 8.9)	9.9 ($\pm$ 9.2)	9.7 ($\pm$ 7.7)	10.1 ($\pm$ 8.4)
Endive	2.0 ($\pm$ 4.1)	2.4 ($\pm$ 4.9)	2.2 ($\pm$ 3.5)	2.5 ($\pm$ 4.3)
Lettuce	7.5 ($\pm$ 7.0)	7.6 ($\pm$ 6.7)	7.4 ($\pm$ 6.5)	7.6 ($\pm$ 6.6)
Alliums	27.8 ($\pm$ 23.5)	28.5 ($\pm$ 24.2)	27.9 ($\pm$ 24.2)	29.0 ($\pm$ 23.4)
Leek	8.6 ($\pm$ 11.8)	8.5 ($\pm$ 10.9)	6.5 ($\pm$ 7.2)	9.3 ($\pm$ 11.3)
Onions	19.2 ($\pm$ 17.8)	20.1 ($\pm$ 19.0)	21.4 ($\pm$ 21.2)	19.6 ($\pm$ 18.0)
Other vegetables				
Carrots, cooked	7.7 ($\pm$ 7.0)	9.0 ($\pm$ 8.9)	7.4 ($\pm$ 7.2)	8.9 ($\pm$ 8.6)
Carrots, raw	1.9 ($\pm$ 6.4)	2.1 ($\pm$ 7.8)	3.8 ($\pm$ 9.9)	3.5 ($\pm$ 9.4)
Sweet peppers	2.3 ($\pm$ 3.6)	2.5 ($\pm$ 4.0)	4.1 ($\pm$ 6.5)	3.3 ($\pm$ 4.9)
Tomatoes	19.5 ($\pm$ 19.4)	19.5 ($\pm$ 20.0)	22.6 ($\pm$ 15.5)	23.5 ($\pm$ 20.3)
Red beets	7.5 ($\pm$ 8.1)	7.7 ($\pm$ 8.7)	7.6 ($\pm$ 7.4)	8.1 ($\pm$ 7.7)
Sauerkraut*	5.7 ($\pm$ 5.3)	5.9 ($\pm$ 5.5)	5.5 ($\pm$ 5.2)	5.7 ($\pm$ 4.9)
Mushrooms	2.8 ($\pm$ 3.5)	3.2 ($\pm$ 3.9)	3.3 ($\pm$ 3.8)	3.7 ($\pm$ 4.4)
Gherkins	1.5 ($\pm$ 4.8)	1.9 ($\pm$ 8.1)	2.6 ($\pm$ 5.8)	1.8 ($\pm$ 6.6)
Rhubarb	2.1 ($\pm$ 5.7)	2.2 ($\pm$ 5.7)	1.1 ($\pm$ 4.1)	2.4 ($\pm$ 5.4)
Fruits	<i>n</i> =491	<i>n</i> =1,525	<i>n</i> =78	<i>n</i> =1,598
Total fruits	146.7 ($\pm$ 118.8)	154.4 ($\pm$ 111.8)	171.0 ($\pm$ 127.3)	196.1 ($\pm$ 118.9)
Citrus fruits	62.5 ($\pm$ 76.1)	64.8 ($\pm$ 69.8)	84.0 ($\pm$ 79.1)	88.2 ($\pm$ 73.2)
Oranges	41.6 ($\pm$ 63.8)	40.6 ($\pm$ 51.1)	46.7 ($\pm$ 58.9)	55.9 ($\pm$ 55.8)
Mandarins	3.0 ($\pm$ 5.7)	3.8 ($\pm$ 7.2)	4.7 ($\pm$ 9.1)	5.5 ($\pm$ 8.9)
Grapefruits	7.0 ($\pm$ 22.5)	6.6 ($\pm$ 21.7)	14.7 ($\pm$ 36.3)	11.4 ($\pm$ 26.3)
Orange/grapefruit juice	10.7 ($\pm$ 27.3)	13.8 ($\pm$ 38.2)	17.6 ($\pm$ 37.8)	15.3 ($\pm$ 35.4)
Other fruits				
Apples/pears	58.2 ($\pm$ 71.2)	67.4 ($\pm$ 74.6)	70.7 ($\pm$ 69.5)	84.2 ($\pm$ 82.0)
Bananas	14.6 ($\pm$ 29.6)	12.9 ($\pm$ 25.0)	12.4 ($\pm$ 28.1)	12.9 ($\pm$ 26.8)
Strawberries	6.7 ($\pm$ 8.4)	6.8 ($\pm$ 7.8)	7.0 ($\pm$ 6.4)	8.1 ($\pm$ 8.6)
Grapes	3.8 ($\pm$ 9.0)	3.9 ($\pm$ 8.5)	5.0 ($\pm$ 11.1)	5.0 ($\pm$ 9.7)
Raisins/other dried fruits	0.4 ($\pm$ 1.9)	0.7 ($\pm$ 3.2)	0.5 ($\pm$ 1.7)	0.9 ($\pm$ 2.8)

*mature beans also included

*not included in Brassica group because of lack of glucosinolates

Table 2. Mean ($\pm$ sd) consumption of total vegetables and fruits in the subcohort (men and women combined) for potential risk factors of bladder cancer, Netherlands Cohort Study (1986-1992)

Characteristics	Vegetable consumption		Fruit consumption	
	n=2953	g/day ($\pm$ sd)	n = 3123	g/day ($\pm$ sd)
Age (years)				
55-59	1132	188.7 ($\pm$ 76.6)	1198	170.8 ($\pm$ 119.8)
60-64	1025	191.3 ($\pm$ 74.4)	1082	176.1 ($\pm$ 114.8)
65-69	796	186.6 ($\pm$ 75.1)	843	182.3 ($\pm$ 116.8)
Alcohol consumption (g/day)				
no consumption	676	181.4 ($\pm$ 80.5)	719	185.8 ($\pm$ 120.2)
<15	1484	187.7 ($\pm$ 71.9)	1567	180.3 ($\pm$ 116.2)
≥ 15	710	200.4 ($\pm$ 76.9)	741	153.9 ($\pm$ 111.9)
Coffee consumption (cups/day)				
no consumption	89	196.4 ($\pm$ 94.0)	98	206.9 ($\pm$ 134.8)
<5	1709	187.4 ($\pm$ 70.7)	1800	181.9 ($\pm$ 115.4)
≥ 5	1155	190.9 ($\pm$ 80.4)	1225	164.14 ($\pm$ 117.6)
Tea consumption (cups/day)				
no consumption	334	184.2 ($\pm$ 87.6)	363	151.2 ($\pm$ 119.5)
<4	1476	187.7 ($\pm$ 73.8)	1564	168.5 ($\pm$ 109.9)
≥ 4	1143	192.2 ($\pm$ 73.6)	1196	192.6 ($\pm$ 123.7)
Total water consumption (ml/day)				
tertile 1 (<1847)	992	164.5 ($\pm$ 67.2)	1042	141.3 ($\pm$ 96.6)
tertile 2 (≥ 1847 -<2222)	1005	187.5 ($\pm$ 66.2)	1043	173.3 ($\pm$ 103.8)
tertile 3 (≥ 2222)	956	216.2 ($\pm$ 83.1)	1038	212.8 ($\pm$ 136.7)
Current cigarette smoking				
No	2130	191.0 ($\pm$ 74.7)	2248	187.4 ($\pm$ 118.9)
Yes	823	184.0 ($\pm$ 77.1)	875	145.9 ($\pm$ 107.7)
Cigarette smoking amount				
non smoker	1078	190.4 ($\pm$ 75.9)	1147	201.0 ($\pm$ 119.4)
<15 cig/day	875	187.3 ($\pm$ 71.5)	910	171.0 ($\pm$ 113.7)
≥ 15 cig/day	911	190.6 ($\pm$ 78.0)	950	154.5 ($\pm$ 114.8)
Cigarette smoking duration				
non smoker	1078	190.4 ($\pm$ 75.9)	1147	201.0 ($\pm$ 119.4)
<35 years	941	192.8 ($\pm$ 74.8)	992	175.3 ($\pm$ 115.5)
≥ 35 years	897	183.3 ($\pm$ 74.9)	944	146.7 ($\pm$ 109.6)
High risk occupation*				
No	2940	189.0 ($\pm$ 75.4)	3108	175.8 ($\pm$ 117.4)
Yes	13	203.2 ($\pm$ 83.7)	15	164.9 ($\pm$ 106.2)
Family history of bladder cancer				
No	2896	189.1 ($\pm$ 75.2)	3064	175.5 ($\pm$ 117.2)
Yes	57	186.4 ($\pm$ 88.6)	59	185.8 ($\pm$ 124.0)
Vegetable consumption (g/day)				
tertile 1 (<150)	N/A [†]	N/A [†]	982	151.4 ($\pm$ 110.1)
tertile 2 (≥ 150 -<212)	N/A [†]	N/A [†]	1056	170.0 ($\pm$ 105.3)
tertile 3 (≥ 212)	N/A [†]	N/A [†]	1085	203.4 ($\pm$ 128.6)
Fruit consumption (g/day)				
tertile 1 (<115)	970	173.9 ($\pm$ 72.2)	N/A [†]	N/A [†]
tertile 2 (≥ 115 -<205)	1022	188.5 ($\pm$ 72.2)	N/A [†]	N/A [†]
tertile 3 (≥ 205)	961	204.9 ($\pm$ 78.7)	N/A [†]	N/A [†]

*ever exposed to dye, rubber, leather, and vehicle fumes

[†]not applicable

Table 3a. Incidence rate ratios (RR) and 95% confidence intervals (CI) for bladder cancer according to the consumption of vegetables and fruits, Netherlands Cohort Study (1986-1992)

Vegetable/fruit consumption (g/day in quintiles/categories)	Cases in cohort	Person years in subcohort	RR (95% CI)*	RR (95% CI)†
Vegetables and fruits combined				
1 (<241)	149	3523	1.00 (reference)	1.00 (reference)
2 (≥241-<311)	111	3606	0.76 (0.57-1.02)	0.81 (0.59-1.11)
3 (≥311-<379)	101	3591	0.72 (0.54-0.97)	0.80 (0.58-1.12)
4 (≥379-<471)	90	3614	0.71 (0.53-0.96)	0.76 (0.51-1.12)
5 (≥471)	87	3638	0.75 (0.56-1.03)	0.98 (0.60-1.61)
p-value for linear trend			0.02	0.39
Vegetable consumption				
Total vegetables				
1 (<126)	110	3535	1.00 (reference)	1.00 (reference)
2 (≥126-<163)	122	3607	1.07 (0.80-1.44)	1.09 (0.80-1.50)
3 (≥163-<196)	107	3598	1.01 (0.74-1.37)	1.04 (0.75-1.44)
4 (≥196-<242)	99	3575	0.95 (0.70-1.30)	0.99 (0.71-1.38)
5 (≥242)	100	3659	0.90 (0.66-1.23)	0.91 (0.65-1.27)
p-value for linear trend			0.29	0.38
Cooked vegetables				
1 (<16)	105	3508	1.00 (reference)	1.00 (reference)
2 (≥16-<28)	130	3601	1.22 (0.91-1.64)	1.14 (0.84-1.56)
3 (≥28-<40)	90	3601	0.84 (0.61-1.16)	0.83 (0.59-1.16)
4 (≥40-<58)	99	3603	0.95 (0.69-1.30)	0.86 (0.62-1.20)
5 (≥58)	114	3659	1.01 (0.75-1.37)	0.98 (0.71-1.35)
p-value for linear trend			0.42	0.28
Raw vegetables				
1 (<99)	122	3551	1.00 (reference)	1.00 (reference)
2 (≥99-<128)	108	3586	0.87 (0.65-1.17)	0.94 (0.69-1.29)
3 (≥128-<155)	117	3561	1.12 (0.83-1.50)	1.24 (0.91-1.70)
4 (≥155-<193)	95	3610	0.91 (0.67-1.24)	1.02 (0.73-1.43)
5 (≥193)	96	3665	0.94 (0.69-1.27)	1.10 (0.78-1.53)
p-value for linear trend			0.81	0.38
Leafy vegetables, cooked				
1 (<9)	117	3592	1.00 (reference)	1.00 (reference)
2 (≥9-<16)	106	3549	0.85 (0.63-1.15)	0.84 (0.61-1.15)
3 (≥16-<22)	100	3268	0.92 (0.67-1.25)	0.84 (0.61-1.17)
4 (≥22-<33)	106	3936	0.85 (0.63-1.14)	0.84 (0.61-1.16)
5 (≥33)	109	3628	0.90 (0.66-1.21)	0.89 (0.65-1.23)
p-value for linear trend			0.45	0.46
Leafy vegetables, raw				
1 (<6)	199	6453	1.00 (reference)	1.00 (reference)
2 (≥6-<12)	199	5942	1.17 (0.93-1.47)	1.24 (0.97-1.58)
3 (≥12)	140	5578	0.86 (0.67-1.10)	0.94 (0.73-1.22)
p-value for linear trend			0.24	0.75
Pulses				
1 (<15)	82	3273	1.00 (reference)	1.00 (reference)
2 (≥15-<24)	96	3694	1.05 (0.75-1.46)	0.98 (0.69-1.39)
3 (≥24-<33)	113	3650	1.06 (0.77-1.47)	0.95 (0.68-1.33)
4 (≥33-<47)	114	3769	1.06 (0.77-1.46)	0.94 (0.67-1.33)
5 (≥47)	133	3587	1.24 (0.90-1.70)	1.03 (0.74-1.44)
p-value for linear trend			0.14	0.87
Brassicas				
1 (<16)	108	3363	1.00 (reference)	1.00 (reference)
2 (≥16-<24)	106	3463	1.00 (0.74-1.36)	0.99 (0.71-1.37)
3 (≥24-<33)	112	3677	0.92 (0.68-1.24)	0.89 (0.64-1.22)
4 (≥33-<46)	119	3822	0.96 (0.71-1.29)	0.95 (0.69-1.30)
5 (≥46)	93	3648	0.75 (0.55-1.03)	0.75 (0.54-1.04)
p-value for linear trend			0.05	0.06

Table 3b: Incidence rate ratios (RR) and 95% confidence intervals (CI) for bladder cancer according to the consumption of vegetables and fruits, Netherlands Cohort Study (1986-1992)

Vegetable/fruit consumption (g/day in quintiles/categories)	Cases in cohort	Person years in subcohort	RR (95% CI)*	RR (95% CI)†
Allium vegetables				
1 (<12)	160	5064	1.00 (reference)	1.00 (reference)
2 (≥12-<26)	144	4399	1.08 (0.83-1.40)	1.08 (0.82-1.42)
3 (≥26-<43)	120	4271	0.99 (0.76-1.30)	1.01 (0.76-1.35)
4 (≥43)	114	4239	0.90 (0.69-1.18)	0.89 (0.67-1.19)
p-value for linear trend			0.35	0.35
Fruit consumption				
Total fruits				
1 (<83)	188	3832	1.00 (reference)	1.00 (reference)
2 (≥83-<131)	101	3734	0.64 (0.49-0.85)	0.69 (0.51-0.93)
3 (≥131-<183)	104	3845	0.65 (0.49-0.86)	0.72 (0.53-0.97)
4 (≥183-<256)	95	3758	0.64 (0.48-0.85)	0.70 (0.51-0.96)
5 (≥256)	81	3827	0.66 (0.48-0.89)	0.74 (0.53-1.04)
p-value for linear trend			<0.01	0.02
Citrus fruits				
1 (<15)	194	3835	1.00 (reference)	1.00 (reference)
2 (≥15-<41)	96	3829	0.55 (0.42-0.73)	0.60 (0.45-0.82)
3 (≥41-<84)	109	4276	0.62 (0.47-0.81)	0.71 (0.53-0.96)
4 (≥84-<128)	70	3257	0.61 (0.45-0.84)	0.69 (0.49-0.98)
5 (≥128)	100	3799	0.75 (0.56-1.00)	0.85 (0.62-1.17)
p-value for linear trend			0.01	0.22

*adjusted for age (years) and sex

†adjusted for age (years), sex, number of cigarettes/day, years of cigarette smoking and total vegetable consumption (g/day, for fruits items) or total fruit consumption (g/day, for vegetable consumption)

For specific vegetables and fruits, RRs are presented for increments in mean daily consumption of 25 gram (Table 4). Statistically significant inverse associations were found for the consumption of cauliflower (RR 0.77 CI 0.61-0.98), cooked carrots (RR 0.66 CI 0.47-0.92) and mandarins (RR 0.63 CI 0.42-0.96). No statistically significant positive associations were observed. Table 4 also gives estimates for each vegetable or fruit after simultaneous adjustment for total vegetable and fruit consumption to assess their independent effects. The risk estimates remained practically stable (Table 4).

In table 5, the age-, sex- and smoking-adjusted RRs are presented for combinations of total vegetable and fruit consumption, both divided in tertiles of daily consumption. The protective effect of fruit consumption seemed largest among those in the lowest tertile of vegetable consumption. Subjects in the median tertiles of vegetable and fruit consumption had the lowest risk of bladder cancer (RR 0.66 CI 0.43-1.00, compared to those in the lowest tertiles of vegetable and fruit consumption) (Table 5).

Cigarette smoking is an important risk factor for bladder cancer. Therefore, the analyses were stratified on smoking status and smoking amount (Table 6). Total vegetable consumption was not associated with bladder cancer risk across different cigarette smoking categories. The protective effect of fruit consumption was most pronounced in ex-smokers who smoked more than 15 cigarettes per day and in current smokers, regardless smoking amount (Table 6).

It appeared that a protective effect of vegetable and fruit consumption was mainly confined to invasive transitional cell carcinomas (Table 7). The RRs for non-invasive tumors did not indicate a clear inverse association for vegetable or fruit consumption (Table 7).

Table 4. Incidence rate ratios (RR) and 95% confidence intervals (CI) for bladder cancer for continuous variables of vegetables and fruit consumption, Netherlands Cohort Study (1986-1992)

Vegetable/fruits (g/day)	RR* (95%CI) per 25 g	RR* (95%CI) per 25 g
Vegetable and fruits combined	0.99 (0.96-1.03)	N/A [‡]
Total vegetables	0.99 (0.96-1.03)	N/A [‡]
Total fruits	0.99 (0.96-1.01)	N/A [‡]
Individual vegetable items		
String/French beans	0.97 (0.81-1.15)	0.99 (0.80-1.21)
Broad beans	1.14 (0.81-1.60)	1.20 (0.84-1.70)
Brussels sprouts	1.03 (0.69-1.52)	1.09 (0.71-1.68)
Cauliflower	0.77 (0.61-0.98)	0.75 (0.57-0.98)
Cabbage (white/green)	0.99 (0.69-1.43)	1.04 (0.69-1.58)
Kale	1.02 (0.48-2.21)	1.10 (0.50-2.44)
Spinach	1.01 (0.75-1.37)	1.05 (0.77-1.44)
Endive, cooked	0.96 (0.75-1.23)	0.99 (0.75-1.30)
Endive, raw	0.70 (0.39-1.27)	0.71 (0.39-1.32)
Lettuce	1.05 (0.71-1.55)	1.09 (0.72-1.65)
Leek	1.00 (0.78-1.28)	1.03 (0.79-1.34)
Onions	0.94 (0.82-1.07)	0.94 (0.80-1.10)
Carrots, cooked	0.66 (0.47-0.92)	0.63 (0.43-0.92)
Carrots, raw	1.16 (0.89-1.50)	1.19 (0.92-1.55)
Sweet peppers	1.16 (0.62-2.16)	1.27 (0.66-2.44)
Tomatoes	1.02 (0.91-1.15)	1.05 (0.92-1.21)
Red beets	0.90 (0.66-1.22)	0.92 (0.66-1.27)
Sauerkraut	0.87 (0.54-1.42)	0.91 (0.54-1.54)
Mushrooms	0.68 (0.35-1.31)	0.69 (0.35-1.36)
Gherkins	0.91 (0.68-1.22)	0.92 (0.68-1.25)
Rhubarb	0.88 (0.52-1.50)	0.89 (0.52-1.51)
Individual fruit items		
Oranges	1.00 (0.94-1.06)	1.02 (0.95-1.10)
Mandarins	0.63 (0.42-0.96)	0.65 (0.43-1.00)
Grapefruits	1.04 (0.94-1.16)	1.07 (0.96-1.21)
Orange/grapefruits juice	1.00 (0.94-1.08)	1.00 (0.93-1.08)
Apples/pears	0.97 (0.93-1.01)	0.97 (0.91-1.03)
Bananas	1.05 (0.95-1.16)	1.08 (0.98-1.19)
Strawberries	0.83 (0.59-1.16)	0.86 (0.61-1.22)
Grapes	1.01 (0.71-1.44)	1.08 (0.75-1.54)
Raisins/other dried fruits	0.37 (0.10-1.43)	0.40 (0.11-1.50)

*adjusted for age (years), sex, number of cigarettes/day, years of cigarette smoking, total fruit consumption (g/day, for vegetable items), or total vegetable consumption (g/day, for fruits items)

[‡]adjusted for age (years), sex, number of cigarettes/day, years of cigarette smoking, total fruit consumption (g/day), and total vegetable consumption (g/day)

[‡]not applicable

Table 5. Incidence rate ratios (RR*) and 95% confidence intervals (CI) for bladder cancer for combinations of tertiles of vegetables and fruit consumption, Netherlands Cohort Study (1986-1992)

Fruit consumption	Vegetables consumption		
	≤ 150 g/day	150-212 g/day	≥ 212 g/day
≤ 115 g/day	1.00 (reference)	0.67 (0.45-1.01)	0.88 (0.56-1.39)
115-203 g/day	0.93 (0.64-1.34)	0.64 (0.43-0.95)	0.66 (0.43-1.00)
≥ 203 g/day	0.75 (0.51-1.12)	0.87 (0.59-1.28)	0.70 (0.47-1.05)

*adjusted for age (years) and sex, number of cigarettes/day, and years of cigarette smoking

Table 6. Incidence rate ratio (RR) of bladder cancer for vegetable and fruits consumption, according to cigarette smoking amount and cigarette smoking status categories, Netherlands Cohort Study (1986-1992)

Cigarette Smoking	No. of Cases	Vegetable and fruit consumption (g/day in quintiles)					p*
		1 (low)	2	3	4	5 (high)	
Vegetable consumption							
Never [‡]	59	1.00 (reference)	1.26 (0.56-2.86)	1.32 (0.57-3.07)	1.22 (0.52-2.87)	0.44 (0.15-1.28)	0.22
Ex							
<15 cig [‡]	83	1.00 (reference)	0.95 (0.43-2.11)	1.45 (0.70-3.03)	0.94 (0.42-2.10)	1.14 (0.50-2.58)	0.77
≥15 cig [‡]	151	1.00 (reference)	1.67 (0.89-3.14)	1.15 (0.58-2.29)	1.01 (0.52-1.99)	1.14 (0.60-2.15)	0.53
Current							
<15 cig [‡]	73	1.00 (reference)	0.84 (0.35-2.04)	0.47 (0.19-1.17)	1.15 (0.49-2.68)	0.83 (0.36-1.93)	0.02
≥15 cig [‡]	142	1.00 (reference)	0.84 (0.46-1.54)	1.06 (0.57-1.96)	0.86 (0.44-1.66)	0.78 (0.42-1.45)	0.42
Fruit consumption							
Never [‡]	62	1.00 (reference)	0.98 (0.40-2.41)	0.99 (0.41-2.36)	0.56 (0.22-1.42)	1.12 (0.49-2.59)	0.87
Ex							
<15 cig [‡]	85	1.00 (reference)	1.29 (0.57-2.91)	0.85 (0.36-2.03)	1.72 (0.78-3.78)	2.13 (0.96-4.72)	0.02
≥15 cig [‡]	160	1.00 (reference)	0.52 (0.29-0.91)	0.69 (0.40-1.19)	0.75 (0.42-1.33)	0.44 (0.23-0.85)	0.02
Current							
<15 cig [‡]	80	1.00 (reference)	0.81 (0.39-1.68)	0.71 (0.33-1.49)	0.56 (0.24-1.30)	0.55 (0.19-1.60)	0.05
>15 cig [‡]	150	1.00 (reference)	0.61 (0.35-1.07)	0.86 (0.46-1.59)	0.50 (0.27-0.94)	0.50 (0.23-1.10)	<0.01

*p-trend

[‡]adjusted for age (years) and sex

[‡]number of cigarettes smoked per day; adjusted for age (years), sex, and years of cigarette smoking

Table 7. Incidence rate ratios (RR) for male transitional cell carcinoma of the urinary bladder according to vegetable and fruit consumption, with respect to tumor invasiveness and morphology, Netherlands Cohort Study (1986-1992)

Vegetable and fruit consumption (g/day in quintiles)	Non-Invasive (Tis/Ta/Ti)		Invasive (T2-4)	
	Non-papillary* RR (95% CI) [‡]	Papillary* RR (95% CI) [‡]	Non-papillary* RR (95% CI) [‡]	Papillary* RR (95% CI) [‡]
Vegetable consumption				
1 (low)	1.00 (reference)	1.00 (reference)	1.00 (reference)	1.00 (reference)
2	4.15 (0.87-19.74)	1.08 (0.69-1.70)	0.87 (0.46-1.64)	0.92 (0.52-1.61)
3	0.55 (0.05-6.20)	1.24 (0.80-1.94)	0.75 (0.39-1.48)	0.98 (0.56-1.73)
4	2.95 (0.57-15.18)	0.91 (0.56-1.48)	0.86 (0.44-1.66)	1.08 (0.62-1.88)
5 (high)	1.51 (0.25-9.04)	1.07 (0.68-1.68)	0.89 (0.48-1.67)	0.65 (0.36-1.20)
p-value for linear trend	1.00	0.98	0.72	0.32
Fruit consumption				
1 (low)	1.00 (reference)	1.00 (reference)	1.00 (reference)	1.00 (reference)
2	0.81 (0.19-3.42)	1.12 (0.72-1.74)	0.67 (0.38-1.18)	0.71 (0.42-1.20)
3	1.85 (0.56-6.12)	1.26 (0.81-1.95)	0.53 (0.28-1.01)	0.88 (0.52-1.49)
4	1.27 (0.35-4.64)	0.93 (0.58-1.50)	0.80 (0.46-1.41)	0.77 (0.45-1.31)
5 (high)	1.33 (0.32-5.48)	1.06 (0.68-1.64)	0.60 (0.31-1.19)	0.75 (0.42-1.34)
p-value for linear trend	0.51	0.92	0.15	0.35

*ICD-O:M8120

*ICDO:M8130

[‡]adjusted for age (years), sex, number of cigarettes/day, and years of cigarette smoking

DISCUSSION

The results showed that total vegetable consumption was not associated with bladder cancer risk, whereas the consumption of brassicas (especially cauliflower) was inversely associated with bladder cancer risk. Citrus fruits (especially mandarins) turned out to be responsible for the inverse association found with fruit consumption.

Since the power of the present study is sufficiently large with 6.3 years of follow-up and more than 500 bladder cancer cases, it is unlikely that the overall results are caused by chance. However, chance might still be influential for individual food items. The prospective nature of this cohort study together with completeness of follow-up, as has been achieved in this study, reduced the potential for selection bias to a minimum. Information bias is also largely avoided because dietary habits were reported before bladder cancer was diagnosed. A change in dietary habits of subjects with latent bladder cancer at the time of completing the baseline questionnaire remains, however, a possibility, although this is much less likely compared to subjects with, e.g. gastrointestinal cancer. Repeating analyses after excluding cases diagnosed in the first one or two years hardly affected our results.

A potentially more realistic threat to the interpretation of the observed inverse associations is residual confounding by risk factors for bladder cancer that happen to be associated with vegetable or fruit consumption. The most important risk factor in this respect is cigarette smoking.²⁵ Unhealthy habits, such as smoking and eating less vegetables and fruits, tend to cluster in the same subjects in most populations.²⁹ In this way, it might happen that an observed inverse association between vegetable or fruit consumption and bladder cancer risk was due to residual confounding by smoking. Evidence of an association between smoking amount and fruit consumption was also present in the present study, although not very strong ($r=-0.17$). We modeled cigarette smoking habits such that they best explained bladder cancer risk.²⁵ This resulted in a model including the habitual number of cigarettes smoked per day (smoking amount) and the number of years smoked (smoking duration), both as continuous variables. When we added these smoking variables to an age and sex-adjusted model, the RR estimates changed slightly. We therefore believe that the associations observed between vegetable or fruit consumption and bladder cancer risk were not entirely due to residual confounding by smoking, although we cannot exclude some influence. We were not able to explain our results on the basis of confounding by other factors in addition to cigarette smoking, since our results were essentially unchanged after incorporating into the analyses many known or suspected risk factors for bladder cancer, including total consumption of alcohol, coffee, tea and water, high risk occupation and family history of bladder cancer.

Could the measurement of vegetable or fruit consumption itself have biased the results? Vegetables are generally considered as food items that are not very easy to assess in food-frequency questionnaires (as well as in other methods of dietary assessment), particularly if portion sizes have to be estimated. In the NLCS validation study, the correlation coefficient for total vegetable consumption was 0.38,²⁰ which is rather low, but comparable to the figure reported for many other prospective studies.³⁰ One of the reasons for this low correlation may be the relative lack of true contrast in the frequency of vegetable consumption in a population such as the Dutch, because people are accustomed to a diet including one hot meal per day, which usually includes vegetables. A consequence of a relatively large measurement error, resulting in an attenuation of the estimated RR, is underestimation of the inverse association between total vegetable consumption and bladder cancer risk. Due to individual preferences, contrast in consumption frequency of many specific vegetables is much higher. It was not possible to assess validity for specific vegetables in the NLCS validation study, since 9 days of dietary record are not sufficient to estimate consumption frequency of specific vegetables. To minimize the amount of uninformative data in addition to the general dietary exclusion criteria, we excluded subjects who appeared not to have understood how to fill in the questions on vegetable consumption; an extreme score on the vegetable error index defined those subjects.

To our knowledge, five case-control studies^{7,9,11,14,31} and five cohort studies^{5,10,12,13,32} have evaluated the relationship between total vegetable or fruit consumption and bladder cancer risk (Table 8). Most studies found inverse associations for total fruit consumption,^{7,9,12,13} total vegetable consumption⁷⁻¹¹ and total consumption of fruits and vegetables combined,^{8,13,31} although one case-control study¹⁴ and one cohort study¹³ did not find an association between total vegetable consumption and bladder cancer risk and one case-control study¹⁴ and one cohort study did not find an association between fruit consumption and bladder cancer risk⁸ (Table 8). Recently, a meta-analysis on epidemiological studies reported elevated bladder cancer risks among subjects with low fruit consumption (RR=1.40, CI: 1.08-1.83) and low vegetable consumption (RR=1.16, CI: 1.01-1.34)(28).

Table 8. Epidemiological studies of vegetable and fruit consumption in relation to bladder cancer incidence.

Author (Year)	Country	Design	Sex	Cases	Exposure measurement	RR (95%CI)* Total vegetables and fruits	RR (95%CI)* Total vegetables	RR (95%CI)* Total fruits
Claude, 1986 ³¹	Germany	case-control	men	340	interview	0.59 (0.37-0.95) [†]	N/A [‡]	N/A [‡]
Steineck, 1988 ³²	Sweden	cohort	women	91	questionnaire	0.90 (0.37-2.21) [†]	N/A [‡]	N/A [‡]
Mills, 1991 ¹⁰	US	cohort	both	80	questionnaire	1.0 (0.6-1.6)	N/A [‡]	N/A [‡]
Negri, 1991 ⁹	Italy	case-control	both	52	interview	N/A [‡]	0.54 (0.26-1.14)	N/A [‡]
Riboli, 1991 ¹⁴	Spain	case-control	both	365	interview	N/A [‡]	0.3 (0.2-0.4)	0.4 (0.3-0.6)
Shibata, 1992 ¹³	US	cohort	men	432	interview	N/A [‡]	1.04 (0.73-1.48)	0.95 (0.67-1.35)
Chyou, 1993 ¹²	US	cohort	men	71	questionnaire	0.85 (0.46-1.56)	1.10 (0.64-1.90)	0.56 (0.28-1.11)
D'Avanzo, 1995 ¹¹	Italy	case-control	both	96	questionnaire	N/A [‡]	N/A [‡]	0.63 (0.37-1.08)
Bruemmer, 1996 ⁷	US	case-control	both	431	interview	N/A [‡]	0.77 (0.53-1.11)	N/A [‡]
Michaud, 1999 ⁸	US	cohort	men	262	questionnaire	N/A [‡]	0.87 (0.53-1.45)	0.56 (0.32-0.96)
Zeegers, 2000	NL	cohort	men	252	questionnaire	0.75 (0.49-1.14)	0.72 (0.47-1.09)	1.12 (0.70-1.78)
			both	538	questionnaire	0.98 (0.60-1.61)	0.91 (0.65-1.27)	0.74 (0.53-1.04)

* comparing highest to lowest consumption categories

[†]yes vs. no

[‡]not applicable

One case-control study³³ and one cohort study⁸ examined the association between brassica consumption and bladder cancer risk. The results from both studies were in accordance with our results indicating an inverse association. As in our study, the cohort study also presented an inverse association between cauliflower consumption and bladder cancer risk.³⁴ Brassicas have been widely studied for their anticarcinogenic properties in experimental studies and have been associated with reduced cancer risk.^{8,15,35,36} They are unique in their high content of dithiolthiones and isothiocyanates. These organosulfur compounds have been shown to increase the activity of enzymes involved in the detoxification of carcinogens.^{8,15}

Two case-control studies^{33,37} have reported a significant inverse association between carrot consumption and bladder cancer risk, but three cohort studies^{8,13,38} have reported no association between orange-yellow vegetable consumption and bladder cancer risk. These vegetables (e.g., carrots) are sources of alpha and beta-carotene, which are antioxidants.¹⁵

To date, no study estimated the association between the consumption of citrus fruit (e.g., mandarins) and bladder cancer risk. Citrus fruits are known for their high content of vitamin C, which, as an antioxidant, may protect cell membranes and DNA from

oxidative damage. Vitamin C may further help prevent cancer via its ability to scavenge and reduce nitrite, thereby reducing substrate for the formation of nitrosamines. The anti-oxidant mechanism has received most attention. The lack of effect of vegetable and fruit consumption among non-smokers is consistent with this hypothesis since never smokers have not been exposed to smoking induced oxidative stress. Recently, Cohen³⁹ suggested that transitional cell carcinoma of the urinary bladder might represent two distinct diseases: papillary transitional cell carcinoma, which is usually non-invasive, and the non-papillary type, which tends to be invasive. In addition to different prognostic and pathogenic properties, these two diseases appear to involve different molecular events.³⁹ From an etiological point of view, some distinction might be important. An inverse association between both vegetables and fruits was most pronounced in invasive transitional cell carcinomas of the urinary bladder, irrespective of tumor morphology. Further research is needed to evaluate this matter. In conclusion, inverse associations have been found between consumption of brassicas, total fruit, citrus fruit, and bladder cancer incidence, which are unlikely to be due to residual confounding of smoking.

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REFERENCES

1. Parkin, D. M., Pisani, P., and Ferlay, J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer* 1999;80:827-41.
2. Silverman, D. T., Hartge, P., Morrison, A. S., and Devesa, S. S. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am* 1992;6:1-30.
3. Johansson, S. L., and Cohen, S. M. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol* 1997;13:291-8.
4. Ross, R. K., Jones, P. A., and Yu, M. C. Bladder cancer epidemiology and pathogenesis. *Semin Oncol* 1996;23:536-45.
5. World Cancer Research Fund, and American Institute for Cancer Research. *Bladder, Food, Nutrition and the Prevention of Cancer: a global perspective*, pp. 338-61. Menasha: Banta Book Group, 1997.
6. van der Meijden, A. P. M. Bladder cancer. *Br Med J* 1998;317:1366-1369.
7. Bruemmer, B., White, E., Vaughan, T. L., and Cheney, C. L. Nutrient intake in relation to bladder cancer among middle-aged men and women. *Am J Epidemiol* 1996;144:485-95.
8. Michaud, D. S., Spiegelman, D., Clinton, S. K., Rimm, E. B., Willett, W. C., and Giovannucci, E. L. Fruit and vegetable intake and incidence of bladder cancer in a male prospective cohort. *J Natl Cancer Inst* 1999;91:605-613.
9. Negri, E., La Vecchia, C., Franceschi, S., B, D. A., and Parazzini, F. Vegetable and fruit consumption and cancer risk. *Int J Cancer* 1991;48:350-4.
10. Mills, P. K., Beeson, W. L., Phillips, R. L., and Fraser, G. E. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol* 1991;133:230-9.
11. D'Avanzo, B., La Vecchia, C., Negri, E., Decarli, A., and Benichou, J. Attributable risks for bladder cancer in northern Italy. *Ann Epidemiol* 1995;5:427-31.
12. Chyou, P. H., Nomura, A. M., and Stemmermann, G. N. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol* 1993;3:211-6.
13. Shibata, A., Paganini Hill, A., Ross, R. K., and Henderson, B. E. Intake of vegetables, fruits, beta-carotene, vitamin C and vitamin supplements and cancer incidence among the elderly: a prospective study. *Br J Cancer* 1992;66:673-9.
14. Riboli, E., Gonzalez, C. A., Lopez Abente, G., Errezola, M., Izarzugaza, I., Escobar, A., Nebot, M., Hemon, B., and Agudo, A. Diet and bladder cancer in Spain: a multi-centre case-control study. *Int J Cancer* 1991;49:214-9.
15. Steinmetz, K. A., and Potter, J. D. Vegetables, fruit, and cancer prevention: a review. *J Am Diet Assoc* 1996;96:1027-39.
16. Prentice, R. L. A case-cohort design for epidemiologic cohort studies and disease prevention trials. *Biometrika*. 1986; 73: 1-11.

17. van den Brandt, P. A., Goldbohm, R. A., van het Veer, P., A., V., Hermus, R. J. J., and Sturmans, F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol* 1990;43:285-95.
18. van den Brandt, P. A., Schouten, L. J., Goldbohm, R. A., Dorant, E., and Hunein, P. H. M. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol* 1990;19:553-8.
19. Goldbohm, R. A., van den Brandt, P. A., and Dorant, E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond* 1994;72:80-4.
20. Goldbohm, R. A., van den Brandt, P. A., Brants, H. A., van het Veer, P., Al, M., Sturmans, F., and Hermus, R. J. S. O. Validation of a dietary questionnaire used in large-scale prospective cohort study on diet and cancer. *Eur J Clin Nutr* 1994;48:253-65.
21. Volovics, A., and van den Brandt, P. A. Methods for the analyses of case-cohort studies. *Biom J* 1997;2:195-214.
22. Stata Statistical Software: Release 6.0. College Station, TX: Stata Corporation.
23. Barlow, W. E. Robust variance estimation for the case-cohort design. *Biometrics* 1994;50:1064-72.
24. Lin, D. Y., and Ying, Z. Cox regression with incomplete covariate measurements. *JASA* 1993;88:1341-9.
25. Zeegers, M. P., Tan, F. E., Dorant, E., and van den Brandt, P. A. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer* 2000;89:630-9.
26. Zeegers, M. P. A., Tan, F. E. S., Goldbohm, R. A., and van den Brandt, P. A. Are coffee and tea consumption associated with urinary tract cancer risk: a systematic review and meta-analysis. *Int J Epidemiol* in press, 2001; in press.
27. Zeegers, M. P., Tan, F. E., Verhagen, A. P., Weijnenberg, M. P., and van den Brandt, P. A. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control* 1999;10:445-51.
28. Steinmaus, C. M., Nunez, S., and Smith, A. H. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol* 2000;151:693-702.
29. Morabia, A., and Wynder, E. L. Dietary habits of smokers, people who never smoked, and exsmokers. *Am J Clin Nutr* 1990;52:933-7.
30. Voorrips, L. E., Goldbohm, R. A., Verhoeven, D. T., van Poppel, G. A., Sturmans, F., Hermus, R. J., and van den Brandt, P. A. Vegetable and fruit consumption and lung cancer risk in the Netherlands Cohort Study on diet and cancer. *Cancer Causes Control* 2000;11:101-15.
31. Claude, J., Kunze, E., Frentzel Beyme, R., Paczkowski, K., Schneider, J., and Schubert, H. Life-style and occupational risk factors in cancer of the lower urinary tract. *Am J Epidemiol* 1986;124:578-89.
32. Steineck, G., Norell, S. E., and Feychting, M. Diet, tobacco and urothelial cancer: a 14-year follow-up of 16,477 subjects. *Acta Oncol* 1988;27:323-7.
33. Mettlin, C., and Graham, S. Dietary risk factors in human bladder cancer. *Am J Epidemiol* 1979;110:255-63.
34. Michaud, D. S., Spiegelman, D., Clinton, S. K., Rimm, E. B., Curhan, G. C., Willet, W. C., and Giovannucci, L. Fluid intake and the risk of bladder cancer in men. *N Engl J Med* 1999;340:1390-1397.
35. Verhoeven, D. T., Goldbohm, R. A., van Poppel, G., Verhagen, H., and van den Brandt, P. A. Epidemiological studies on brassica vegetables and cancer risk. *Cancer Epidemiol Biomarkers Prev* 1996;5:733-48.
36. Verhoeven, D. T., Verhagen, H., Goldbohm, R. A., van den Brandt, P. A., and van Poppel, G. A review of mechanisms underlying anticarcinogenicity by brassica vegetables. *Chem Biol Interact* 1997;103:79-129.
37. La Vecchia, C., Negri, E., Decarli, A., B., D. A., Liberati, C., and Franceschi, S. Dietary factors in the risk of bladder cancer. *Nutr Cancer* 1989;12:93-101.
38. Hirayama, T. A large scale cohort study on cancer risks by diet: with special reference to the risk reducing effects of green-yellow vegetable consumption. *Princess Takamatsu Symp* 1985;16:41-53.
39. Cohen, S. M. Urinary Bladder carcinogenesis. *Toxicol Pathol* 1998;26:121-127.

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Are Retinol, Vitamin C, Vitamin E, Folate and Carotenoids Intake associated with Bladder Cancer Risk? Results from the Netherlands Cohort Study

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ABSTRACT

Although dietary components may have anticarcinogenic properties, few studies have investigated prospectively the association between nutrient intake and bladder cancer.

In the Netherlands Cohort Study among 120,852 subjects aged 55-69 years at baseline (1986), the association between vitamins and carotenoids intake, vitamin supplement use, and bladder cancer incidence was examined. Exposure status was measured with a food-frequency questionnaire. After 6.3 years of follow-up, data from 569 cases and 3,123 subcohort members were available for case-cohort analyses.

The age-, sex-, and smoking-adjusted RRs for retinol, vitamin E, folate, α -carotene, β -carotene, lutein & zeaxanthin, and lycopene were 1.04, 0.98, 1.03, 0.99, 1.16, 1.11, and 1.08, respectively, comparing highest to lowest quintile of intake. Only vitamin C (RR:0.81, 95%CI:0.61-1.07, p -trend=0.08), and β -cryptoxanthin intake (RR:0.74, 95%CI:0.53-1.03, p -trend<0.01) were inversely associated with bladder cancer risk. The association with vitamin C disappeared after adjustment for β -cryptoxanthin but not vice versa. The RRs for supplemental use of vitamin A, C or E compared to no use were around unity.

We conclude that dietary or supplemental intake of vitamin A, vitamin C, vitamin E, and intake of folate, and most carotenoids are not associated with bladder cancer. In this study, only β -cryptoxanthin intake appeared to be inversely associated.

INTRODUCTION

Bladder cancer is the most common urological cancer and is the seventh most common cancer among men, accounting for approximately 200,000 new cases per year worldwide.¹ Over the last four decades, many epidemiologic studies and several reviews have been conducted to investigate determinants of bladder cancer.²⁻⁶ These studies suggested that bladder cancer is influenced by environmental factors, including cigarette smoking, fluid consumption, schistosomal infections, exposure to industrial chemicals (e.g., aromatic amines), and diet.

Among the dietary factors, it has been suggested that vegetable and fruit consumption may protect against cancer of at various anatomical sites. With respect to bladder cancer, most epidemiological studies support a possible protective effect of vegetable⁷⁻¹¹ and fruit consumption,^{7,9,12,13} although some studies have reported no association for vegetable^{13,14} or fruit consumption.^{8,14} There are many candidate agents in fruits and vegetables that influence bladder cancer risk, including carotenoids, vitamin C, vitamin E and folate.

The mechanisms of action by which these components may act include antioxidant effects (vitamins C, E, and carotenoids), inhibition of nitrosamine formation (vitamins C and E), or reduced methylation of DNA (folate). Some carotenoids (e.g., α -carotene, β -carotene, and β -cryptoxanthin) may also reduce cancer risk as a result of their conversion to retinol. Among the carotenoids, β -carotene has been studied predominantly, because most food tables contain data on β -carotene only.¹⁵

In the present prospective study, we were able to explore the influence of the intake of different vitamins (i.e., retinol, vitamin C, vitamin E, and folate) and the use of vitamin-containing supplements on bladder cancer risk with substantially more cases than in previous studies. Furthermore, with the use of a recently developed food composition table on levels of carotenoids in Dutch foods,¹⁶ analyses were performed with α -carotene, β -carotene, lutein plus zeaxanthin, β -cryptoxanthin, and lycopene.

MATERIALS AND METHODS

Cohort. This population based prospective cohort study on diet and cancer started in the Netherlands in September 1986. The cohort includes 58,279 men and 62,573 women aged 55 to 69 years at baseline. The study population originated from 204 municipal population registries throughout the country. The case cohort approach was used for data processing and analysis.¹⁷ Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up, during the follow-up period. The study design, including data collection strategies, has been described in detail previously.¹⁸

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.¹⁹ The completeness of cancer follow-up was estimated to be over 95 percent.²⁰ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 3,346 subcohort members (1,630 men and 1,716 women), and 619 incident cases (532 men and 87 women) with microscopically confirmed, incident carcinomas of the urinary bladder, ureters, renal pelvis or urethra were identified. Of these cases, 584 (94.3%) were diagnosed with bladder cancer of which 559 (95.7%) were transitional cell carcinomas. Because the overwhelming majority of tumors occurred in the urinary bladder and the ureters, renal pelvis, and urethra are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Exposure status. At baseline, the cohort members completed a mailed, self-administered questionnaire on risk factors for cancer. Usual consumption of food and beverages during the year preceding the start of the study was assessed with a 150-item semiquantitative food frequency questionnaire. The questionnaire was validated against a 9-day diet record. The energy and sex-adjusted Pearson correlation coefficients between the 9-day record and the questionnaire were 0.48 for total vitamin A and 0.55 for vitamin C.²¹ The questionnaire data were key entered twice and processed in a standardized manner blinded with respect to case/subcohort status in order to minimize observer bias in coding and interpretation of the data. In this study, variables of principle interest were retinol, vitamins C, and E, folate, several carotenoids, and the use of vitamin-containing supplements. The mean daily intake of retinol, vitamins C and E were calculated using the computerized Dutch Food Composition Table.¹⁶ Folate data were derived from a validated liquid chromatography trienzyme method with which the most important Dutch foods contributing to folate intake were analyzed.²² For calculating the intake of specific carotenoids an additional food composition table has been constructed recently.²³ Briefly, regularly eaten vegetables were comprehensively sampled and analyzed for their carotenoid content and the database was completed with data from the recent literature and information from food manufacturers.²³ With this carotenoid table, we were able to evaluate six of the most important types of carotenoids: α -carotene, β -carotene, lutein, zeaxanthin, β -cryptoxanthin, and lycopene. In the carotenoid food composition table, lutein and zeaxanthin were combined, because most literature sources had not distinguished these two carotenoids. Most vegetables, however, contain primarily lutein and only minor amounts of zeaxanthin. Information on dietary supplement use was collected using an open-ended question with space for adding a maximum of four different supplements. Participants were asked whether they used vitamin tablets, drops or other preparations during five years before baseline. Subjects were categorized in users or nonusers of supplements containing vitamin A, C or E.

Statistical analyses. Subjects with incomplete or inconsistent dietary data were excluded leaving 569 cases (491 men and 78 women) with bladder cancer and 3,123 subcohort members (1525 men and 1598 women) for the analyses.

Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models²⁴ with the Stata statistical software package.²⁵ Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow²⁶ and Lin and Ying.²⁷ We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against failure time.

Subjects were classified by quintile of intake of the vitamins and carotenoids or were classified by their use of vitamin-containing supplements (yes/no). For vitamin C, quintiles 2 and 3, and quintiles 4 and 5 were combined, because the validation study demonstrated that these quintiles could not be distinguished²¹. Tests for trends in risk for bladder cancer over multiple categories were assessed by fitting ordinal exposure variables as continuous terms and performing likelihood-ratio tests between regression models with and without these variables.

The following variables were subsequently considered as potential confounders based on earlier analyses:²⁸⁻³¹ consumption of alcohol (g/day), coffee (ml/day), tea (ml/day) water (ml/day), vegetables (g/day), and fruits (g/day), current cigarette smoking (yes/no), smoking amount (cigarettes/day), smoking duration (years of cigarette smoking), occupational exposure to dye, rubber, leather or vehicle fumes (ever/never), and first degree family history of bladder cancer (yes/no). Those variables that showed a more than ten percent influence on the risk of bladder cancer when considered in a multivariable model were included as covariates in the analyses. For β -cryptoxanthine intake, subgroup analyses were performed after stratification by tumor morphology and invasiveness.

RESULTS

Table 1 presents the distribution of the dietary and supplemental vitamin intakes and the distribution of potential confounding factors among bladder cancer cases and subcohort members. The dietary intakes of retinol, vitamin E, folate, α -carotene, β -carotene, lycopene, β -cryptoxanthin, and lutein/zeaxanthine were comparable between cases and subcohort members. The intake of vitamin C and the use of supplements containing vitamin A, C or E were higher among subcohort members than among cases. Of the potential confounders, the distribution of age, total water, alcohol, coffee, tea, total vegetable and total fruit consumption was similar for cases and subcohort members. More than 80 percent of the cases were male, whereas in the subcohort approximately 50 percent were male. Cigarette use was more often reported in cases than in subcohort members and smoking cases appeared to smoke more cigarettes per day for more years than smoking subcohort members. Less than 2 percent of the cases and subcohort members reported a family history of bladder cancer or had ever worked in a high-risk occupation (Table 1).

The association between nutrient intake and bladder cancer risk was similar in men and women for all nutrients (data not shown). Therefore, the results are presented for men and women combined. As is shown in table 2, except for vitamin C intake, none of the vitamins (retinol, vitamin E, and folate), appeared to be associated with bladder cancer risk. The age- and sex-adjusted RRs of bladder cancer were 0.92 (CI:0.68-1.26), 0.88 (CI:0.65-1.20) and 0.93 (CI:0.70-1.25) comparing highest to lowest quintiles of intake from retinol, vitamin E and folate respectively. The age- and sex-adjusted RRs of bladder cancer for quintiles 2 and 3 combined and quintiles 4 and 5 combined of vitamin C intake were 0.77 (CI:0.61-0.98) and 0.74 (CI:0.58-0.95) compared to the lowest quintile of intake, respectively (p -trend<0.01). Although cigarette smoking amount and cigarette smoking duration are important determinants of bladder cancer, adjustment for these covariates increased the risk estimates not essentially (Table 2). Also, adjustment for other potential confounders did not substantially change the risk estimates (data not shown).

The intake of the carotenoids α -carotene, β -carotene, luteine/zeaxanthine, and lycopene were not associated with bladder cancer risk (Table 3). The age- and sex-adjusted RRs for the variables were all around unity and statistically non-significant. The intake of β -cryptoxanthin appeared to be inversely associated with bladder cancer risk (p -trend<0.01) with corresponding age- and sex adjusted RRs per increasing quintiles of intake of 1.00 (reference), 0.63 (CI:0.48-0.83), 0.54 (CI:0.40-0.72), 0.59 (CI:0.44-0.80) and 0.71 (CI:0.53-0.95), respectively. Adjustment for cigarette smoking or other potential confounders did not change the results substantially (Table 3).

Since the intake of vitamin C and β -cryptoxanthin were correlated ($r=0.69$), and both vitamin C and β -cryptoxanthin were inversely associated with bladder cancer risk (table 2,3) we estimated the incidence rate ratios for both nutrients after adjustment for each other in addition to age, sex, cigarette smoking amount, and cigarette smoking duration (Table 4). After adjustment for β -cryptoxanthin intake, the inverse association for vitamin C intake disappeared. However, the RRs for β -cryptoxanthin intake did not change essentially after adjustment for vitamin C intake (Table 4).

Table 1. Description of mean daily intake of vitamins, carotenoids, and vitamin supplement use, and distribution of potential confounding factors, Netherlands Cohort Study (1986-1992)

	Cases (n=569)		Subcohort (n=3123)	
Exposure variables	<i>Mean</i>	<i>Sd</i>	<i>Mean</i>	<i>Sd</i>
Retinol (mg/day)	0.59	0.33	0.54	0.31
Vitamin C (mg/day)	97.0	44.1	103	43.0
Vitamin E (mg/day)	13.9	6.27	13.4	6.24
Folate (mg/day)	219	73.7	212	0.70
α -carotene (mg/day)	0.66	0.48	0.70	0.58
β -carotene (mg/day)	2.89	1.42	2.97	1.58
Lycopene (mg/day)	1.14	1.94	1.19	1.74
β -cryptoxanthin (mg/day)	0.15	0.20	0.18	0.17
Lutein/zeaxanthin (mg/day)	2.54	1.23	2.52	1.11
	<i>Number</i>	<i>Percentage</i>	<i>Number</i>	<i>Percentage</i>
Use of supplements containing vitamin A (% yes)	30	5.27	226	7.24
Use of supplements containing vitamin C (% yes)	48	8.44	325	10.41
Use of supplements containing vitamin E (% yes)	28	4.92	204	6.53
Potential confounding factors	<i>Mean</i>	<i>Sd</i>	<i>Mean</i>	<i>Sd</i>
Age (years)	62.7	4.07	61.4	4.22
total water consumption (l/day)	2.13	0.52	2.07	0.49
alcohol consumption (g/day)	18.2	18.2	13.3	14.9
coffee consumption (cups/day)	4.80	2.39	4.29	2.16
tea consumption (cups/day)	2.45	2.02	2.86	2.03
total vegetable consumption (g/day)	190	82.4	194	83.5
total fruit consumption (g/day)	151	200	177	117
cigarette smoking amount (cig/day)*	18.7	11.3	15.0	10.3
cigarette smoking duration (years)*	37.5	11.6	31.6	12.3
	<i>Number</i>	<i>Percentage</i>	<i>Number</i>	<i>Percentage</i>
Sex (% male)	491	86.3	1525	48.8
cigarette smoking status (% current)	255	44.8	875	28.0
family history of bladder cancer (% yes)	7	1.23	59	1.89
high risk occupation (% yes)†	4	0.70	15	0.48

*among smokers,
†ever exposed to dye, rubber, leather, and vehicle fumes

We investigate potential effect modification of the association between β -cryptoxanthin and bladder cancer risk by cigarette smoking amount and duration (Table 5). We found a statistical significant interaction effect with smoking amount ($p<0.01$) but not with smoking duration ($p=0.10$). The inverse association between β -cryptoxanthin and bladder cancer risk was not found among non-smokers and appeared to be limited to cigarette smokers who smoked more than 15 cigarettes per day or who smoked for more than 35 years (Table 5)

The protective effect of β -cryptoxanthin intake was observed in each stratum of tumor invasiveness or morphology of male transitional cell carcinomas of the bladder, although the association was less pronounced for non-invasive papillary tumors (Table 6).

The risks of bladder cancer for consumers of vitamin supplements containing vitamin A, vitamin C or Vitamin E were similar to the risk for non-consumers (Table 4). After adjustment for age, sex, cigarette smoking amount and cigarette smoking duration the RRs were 1.12 (CI:0.71-1.78), 1.01 (CI:0.69-1.48) and 1.05 (CI:0.63-1.73), respectively (Table 7).

Table 2. Incidence rate ratios and 95% confidence intervals for bladder cancer according to quintiles of intake of retinol, vitamins C, and E, and folate, Netherlands Cohort Study (1986-1992)

Vitamins	Cases	Quintiles of intake					p-trend
		1 (low)*	2	3	4	5 (high)	
Retinol							
median intake (mg/day)		0.26	0.38	0.48	0.61	0.87	
RR (95% CI) [†]	569	1.00	0.75 (0.54-1.06)	0.95 (0.69-1.30)	0.98 (0.72-1.33)	0.92 (0.68-1.26)	0.71
RR (95% CI) [‡]	475	1.00	0.83 (0.56-1.24)	1.04 (0.72-1.51)	1.02 (0.71-1.46)	1.04 (0.72-1.48)	0.67
Vitamin C[§]							
median intake (mg/day)	54.9		86.6		135		
RR (95% CI) [†]	569	1.00	0.77 (0.61-0.98)		0.74 (0.58-0.95)		<0.01
RR (95% CI) [‡]	475	1.00	0.87 (0.66-1.14)		0.81 (0.61-1.07)		0.08
Vitamin E							
median intake (mg/day)	6.59	9.27	12.2	15.8	21.7		
RR (95% CI) [†]	569	1.00	0.99 (0.72-1.36)	1.08 (0.79-1.48)	1.05 (0.77-1.43)	0.88 (0.65-1.20)	0.46
RR (95% CI) [‡]	475	1.00	1.10 (0.76-1.61)	1.28 (0.90-1.84)	1.11 (0.77-1.59)	0.98 (0.68-1.41)	0.10
Folate							
median intake (mg/day)	0.14	0.17	0.20	0.23	0.29		
RR (95% CI) [†]	569	1.00	0.80 (0.58-1.10)	0.80 (0.59-1.08)	0.72 (0.53-0.98)	0.93 (0.70-1.25)	0.55
RR (95% CI) [‡]	475	1.00	0.85 (0.59-1.23)	0.87 (0.62-1.24)	0.82 (0.58-1.17)	1.03 (0.73-1.44)	0.77

*reference category

[†]adjusted for age (years), and sex

[‡]adjusted for age (years), sex, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

[§]for vitamin C, quintiles 2 and 3, and quintiles 4 and 5 were combined

Table 3. Incidence rate ratios and 95% confidence intervals for bladder cancer according to quintiles of intake of carotenoids, Netherlands Cohort Study (1986-1992)

Carotenoids	Cases	Quintiles of intake					p-trend
		1 (low)*	2	3	4	5 (high)	
α-Carotene							
median intake (mg/day)		0.19	0.38	0.57	0.82	1.31	
RR (95% CI) [†]	569	1.00	0.85 (0.63-1.13)	0.74 (0.55-1.00)	0.90 (0.68-1.20)	0.83 (0.61-1.11)	0.27
RR (95% CI) [‡]	475	1.00	0.81 (0.58-1.13)	0.76 (0.54-1.06)	0.83 (0.60-1.16)	0.99 (0.71-1.39)	0.89
β-Carotene							
median intake (mg/day)		1.43	2.10	2.65	3.37	4.76	
RR (95% CI) [†]	569	1.00	1.04 (0.77-1.39)	0.87 (0.64-1.18)	0.89 (0.66-1.20)	0.98 (0.73-1.32)	0.51
RR (95% CI) [‡]	475	1.00	1.07 (0.77-1.49)	0.90 (0.63-1.27)	0.93 (0.66-1.32)	1.16 (0.82-1.63)	0.67
Lutein and zeaxanthin							
median intake (mg/day)		1.34	1.86	2.34	2.84	3.82	
RR (95% CI) [†]	569	1.00	0.97 (0.72-1.31)	0.86 (0.64-1.16)	0.84 (0.62-1.13)	1.02 (0.76-1.37)	0.73
RR (95% CI) [‡]	475	1.00	1.00 (0.71-1.41)	0.89 (0.63-1.26)	0.99 (0.70-1.40)	1.11 (0.79-1.57)	0.49
β-cryptoxanthin							
median intake (mg/day)		0.02	0.06	0.13	0.25	0.40	
RR (95% CI) [†]	569	1.00	0.63 (0.48-0.83)	0.54 (0.40-0.72)	0.59 (0.44-0.80)	0.71 (0.53-0.95)	<0.01
RR (95% CI) [‡]	475	1.00	0.70 (0.52-0.95)	0.56 (0.40-0.78)	0.62 (0.44-0.87)	0.74 (0.53-1.03)	<0.01
Lycopene							
median intake (mg/day)		0.15	0.48	0.81	1.21	2.21	
RR (95% CI) [†]	569	1.00	0.83 (0.62-1.11)	1.07 (0.81-1.42)	1.05 (0.78-1.41)	0.97 (0.72-1.31)	0.58
RR (95% CI) [‡]	475	1.00	0.85 (0.61-1.18)	1.07 (0.77-1.48)	1.18 (0.84-1.65)	1.08 (0.77-1.51)	0.16

*reference category

[†]adjusted for age (years), and sex

[‡]adjusted for age (years), sex, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

Table 4. Incidence rate ratios and 95% confidence intervals for bladder cancer according to quintiles of vitamins C₁ and β-cryptoxanthin adjusted for each other, Netherlands Cohort Study (1986-1992)

	Quintiles of intake					p-trend
	1 (low)*	2	3	4	5 (high)	
Vitamin C¹						
RR (95% CI) ²	1.00		1.08 (0.80-1.46)		1.07 (0.73-1.57)	0.67
β-cryptoxanthin						
RR (95% CI) ³	1.00	0.69 (0.50-0.95)	0.54 (0.38-0.78)	0.60 (0.40-0.89)	0.71 (0.46-1.11)	0.01

*reference category

¹for vitamin C, quintiles 2 and 3, and quintiles 4 and 5 were combined

²adjusted for age (years), sex, cigarette smoking amount (cig/day), cigarette smoking duration (years), and β-cryptoxanthin intake

³adjusted for age (years), sex, cigarette smoking amount (cig/day), cigarette smoking duration (years), and vitamin C intake

Table 5. Incidence rate ratios and 95% confidence intervals for bladder cancer according to β-cryptoxanthin intake, with respect to cigarette smoking amount and duration, Netherlands Cohort Study (1986-1992)

Cigarette smoking	Quintiles of β-cryptoxanthin intake					p-trend
	1 (low)*	2	3	4	5 (high)	
Non-smoker ¹	1.00	0.86 (0.35-2.13)	0.88 (0.37-2.09)	0.89 (0.38-2.08)	1.16 (0.51-2.64)	0.67
Amount (cig/day)²						
<15	1.00	0.83 (0.50-1.40)	0.59 (0.33-1.06)	0.98 (0.57-1.67)	1.34 (0.79-2.27)	0.25
15+	1.00	0.68 (0.47-1.00)	0.59 (0.39-0.88)	0.50 (0.32-0.78)	0.54 (0.35-0.83)	<0.01
Duration (years)³						
<35	1.00	0.83 (0.49-1.39)	0.52 (0.29-0.94)	0.71 (0.40-1.26)	0.90 (0.52-1.54)	0.47
35+	1.00	0.67 (0.46-0.99)	0.61 (0.41-0.91)	0.60 (0.39-0.91)	0.67 (0.43-1.03)	<0.01

*reference category

¹adjusted for age and sex

²adjusted for age (years), sex, and cigarette smoking duration (years)

³adjusted for age (years), sex, and cigarette smoking amount (cig/day)

Table 6. Incidence rate ratios and 95% confidence intervals for transitional cell carcinoma of the urinary bladder according to β-cryptoxanthin intake, with respect to tumor invasiveness and morphology, Netherlands Cohort Study (1986-1992)

β-cryptoxanthin (quintiles of intake)	Non-Invasive (T _{1s} /T _{1a} /T _{1i})		Invasive (T ₂₋₄)	
	Non-papillary (n=24)* RR (95% CI) ²	Papillary (n=232) ¹ RR (95% CI) ¹	Non-papillary (n=111)* RR (95% CI) ²	Papillary (n=146) ¹ RR (95% CI) ¹
1 (low)	1.00 (reference)	1.00 (reference)	1.00 (reference)	1.00 (reference)
2	0.38 (0.10-1.43)	1.03 (0.68-1.57)	0.66 (0.37-1.19)	0.51 (0.29-0.88)
3	0.30 (0.06-1.42)	0.75 (0.47-1.20)	0.54 (0.28-1.02)	0.44 (0.24-0.79)
4	0.51 (0.14-1.88)	0.82 (0.51-1.33)	0.55 (0.28-1.09)	0.49 (0.27-0.88)
5 (high)	0.18 (0.02-1.57)	0.84 (0.52-1.36)	0.72 (0.38-1.36)	0.71 (0.41-1.23)
p-value for linear trend	0.04	0.22	0.11	0.06

¹ICD-O:M8120

²ICDO:M8130

³adjusted for age (years), sex, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

Table 7. Incidence rate ratios and 95% confidence intervals for bladder cancer according to the use supplements, Netherlands Cohort Study (1986-1992)

Use of vitamin supplements	No. of Cases (users/nonusers)	Users vs. Nonusers
Vitamin A supplement		
RR (95% CI)*	30/539	0.88 (0.58-1.33)
RR (95% CI)†	27/448	1.12 (0.71-1.78)
Vitamin C supplement		
RR (95% CI)*	48/521	0.94 (0.67-1.32)
RR (95% CI)†	40/435	1.01 (0.69-1.48)
Vitamin E supplement		
RR (95% CI)*	28/541	0.91 (0.59-1.39)
RR (95% CI)†	23/452	1.05 (0.63-1.73)

*adjusted for age (years) and sex

†adjusted for age (years), sex, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

DISCUSSION

The results of the prospective Netherlands Cohort Study showed that in general the intake of retinol, vitamin C, vitamin E, folate, carotenoids, and the use of vitamin supplements are not associated with bladder cancer risk. The initially inverse association with vitamin C intake disappeared after controlling for β-cryptoxanthin intake. Only β-cryptoxanthin intake appeared to be inversely associated with bladder cancer risk, especially among heavy cigarette smokers

The NLCS was carried out in the general Dutch population of men and women aged 55-69 years at baseline. After 6.3 years of follow-up, 569 bladder cancer cases were detected which is substantially more than any other prospective study that investigated the association between these nutrients and the risk of bladder cancer. The prospective nature of a cohort study together with completeness of follow-up, as has been achieved in this study, reduced the potential for selection bias to a minimum. Nondifferential misclassification of exposure, may have resulted in underestimation of the strength of the association. However, the results of the validation study showed that the questionnaire was able to rank subjects adequately according to their intake of most nutrients. In a reproducibility study, it was further demonstrated that the single food frequency questionnaire measurement could characterize dietary habits for a period of at least 5 years.³² Information bias is also largely avoided because dietary habits were reported before bladder cancer was diagnosed. A change in dietary habits of subjects with latent bladder cancer at the time of completing the baseline questionnaire is much less likely than in subjects with, e.g. gastrointestinal cancer. Furthermore, repeating analyses after excluding cases diagnosed in the first one or two years hardly affected our results.

A potentially more realistic threat to the interpretation of the observed inverse associations is residual confounding by cigarette smoking. We modeled cigarette smoking habits such that they best explained bladder cancer.²⁸ This resulted in a model including number of years smoked and habitual number of cigarettes smoked per day, both as continuous variables. We therefore believe that the associations observed were not entirely due to residual confounding by smoking, although we cannot exclude some influence. When we added the smoking variables to an age- and sex-adjusted model, the RR estimates changed only slightly. We were not able to explain our results on the basis of confounding of other factors in addition to cigarette smoking, since our results were essentially unchanged after incorporating into the analyses many known or suspected risk factors for bladder cancer, including total consumption of alcohol, coffee, tea, and water, high risk occupation and family history of bladder cancer.

Vitamin A is important for cell differentiation and since lack of differentiation is a feature of cancer cells, a protective effect of vitamin A on cancer development may be expected.³³ Retinol is the physiologically active form of vitamin A and can also be derived from some carotenoids (e.g., α -carotene, β -carotene, and β -cryptoxanthin). Some carotenoids have antioxidant properties and may inhibit carcinogenesis by preventing DNA damage induced by free radicals. However, a recently published meta-analysis based on seven case control studies and three cohort studies did not find increased risks for diets low in retinol or β -carotene.³¹ A prospective study that was published more recently also found no association between supplemental and dietary vitamin A intake and bladder cancer risk.³³ In the present study, only the dietary intake of the carotenoid β -cryptoxanthin appeared to be inversely associated with bladder cancer risk. To our knowledge, except for its provitamin A activity no biological mechanism can explain this specific association. Citrus fruits are the main sources of β -cryptoxanthin. In a previous analysis we also presented an inverse association between citrus fruit consumption and bladder cancer risk. However, an other previously conducted cohort study did not find an association between β -cryptoxanthin and bladder cancer risk.⁸ Further research is needed to evaluate this matter.

Vitamin C is mostly known for its antioxidant capacity³⁴ and its ability to prevent formation of nitrosamine and other N-nitroso compounds.³⁷ In addition to β -cryptoxanthin, citrus fruits are also known for their high content of vitamin C. After adjustment for β -cryptoxanthin, the small inverse association between vitamin C intake and bladder cancer risk increased towards unity. Two previously conducted population based case-control studies found small increased risks of bladder cancer with increasing intake of dietary vitamin C for men and women combined³⁸ or for men only³⁹ after controlling for cigarette smoking. However, other population based case control studies presented decreased risks for men and women combined⁷ or for women only,³⁹ or found no association at all.¹⁴ One prospective study suggested an inverse association with dose of vitamin C supplement use, but not with dietary vitamin C intake,³³ which is in correspondence to the results of the present study.

Like vitamin C, vitamin E is an intracellular antioxidant and can inhibit nitrosation.^{35,36,40} Two population based case control studies and one prospective study reported inverse associations between dietary vitamin E intake and bladder cancer risk.^{7,14,33} We did not find statistically significant associations, although the point estimates were moderately negative.

Folate is essential for methylation reactions in the human body. Reduced methylation of DNA may contribute to loss of normal controls on proto-oncogene expression. Folate deficiency causes massive incorporation of uracil into human DNA and chromosome breaks. Such breaks could contribute to an increased risk of cancer.^{41,42} The epidemiologic evidence relating dietary folate and the risk of bladder cancer is limited. One previously conducted case-control study found a moderately inverse association between folate intake and bladder cancer risk,⁷ whereas a recently published prospective study did not find an association with bladder cancer risk.³³ In the present study, we found no association between folate intake and bladder cancer risk.

We conclude that dietary or supplemental intake of vitamin A, vitamin C, vitamin E, and dietary intake from folate and carotenoids are not associated with bladder cancer risk. Only β -cryptoxanthin appeared to be inversely associated. This deserves further scientific attention.

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REFERENCES

- 1 Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer*. 1999;80:827-41.
- 2 Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am*. 1992;6:1-30.
- 3 Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol*. 1997;13:291-8.
- 4 Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol*. 1996;23:536-45.
- 5 World Cancer Research Fund, American Institute for Cancer Research. In: Food, Nutrition and the Prevention of Cancer: a global perspective. Menasha: Banta Book Group, 1997.
- 6 van der Meijden APM. Bladder cancer. *Br Med J*. 1998;317:1366-9.
- 7 Bruemmer B, White E, Vaughan TL, Cheney CL. Nutrient intake in relation to bladder cancer among middle-aged men and women. *Am J Epidemiol*. 1996;144:485-95.
- 8 Michaud DS, Spiegelman D, Clinton SK, Rimm EB, Willett WC, Giovannucci EL. Fruit and vegetable intake and incidence of bladder cancer in a male prospective cohort. *J Natl Cancer Inst*. 1999;91:605-13.
- 9 Negri E, La Vecchia C, Franceschi S, B DA, Parazzini F. Vegetable and fruit consumption and cancer risk. *Int J Cancer*. 1991;48:350-4.
- 10 Mills PK, Beeson WL, Phillips RL, Fraser GE. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol*. 1991;133:230-9.
- 11 D'Avanzo B, La Vecchia C, Negri E, Decarli A, Benichou J. Attributable risks for bladder cancer in northern Italy. *Ann Epidemiol*. 1995;5:427-31.
- 12 Chyou PH, Nomura AM, Stemmermann GN. A prospective study of diet, smoking, and lower urinary tract cancer. *Ann Epidemiol*. 1993;3:211-6.
- 13 Shibata A, Paganini Hill A, Ross RK, Henderson BE. Intake of vegetables, fruits, beta-carotene, vitamin C and vitamin supplements and cancer incidence among the elderly: a prospective study. *Br J Cancer*. 1992;66:673-9.
- 14 Riboli E, Gonzalez CA, Lopez Abente G, Errezola M, Izarzugaza I, Escolar A, et al. Diet and bladder cancer in Spain: a multi-centre case-control study. *Int J Cancer*. 1991;49:214-9.
- 15 Verhoeven DT, Assen N, Goldbohm RA, Dorant E, van 't Veer P, Sturmans F, et al. Vitamins C and E, retinol, beta-carotene and dietary fibre in relation to breast cancer risk: a prospective cohort study. *Br J Cancer*. 1997;75:149-55.
- 16 NEVO-table, Dutch food composition table 1986-1987. The Hague, Netherlands: Voorlichtingsbureau voor de Voeding, 1986.
- 17 Prentice RL. A case-cohort design for epidemiologic cohort studies and disease prevention trials. *Biometrika*. 1986;73:1-11.
- 18 van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol*. 1990;43:285-95.
- 19 van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, Hunen PHM. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol*. 1990;19:553-8.
- 20 Goldbohm RA, van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond*. 1994;72:80-4.
- 21 Konings EJ. A validated liquid chromatographic method for determining folates in vegetables, milk powder, liver, and flour. *J AOAC Int*. 1999;82:119-27.
- 22 Goldbohm RA, Brants HA, Hulshof KF, van den Brandt PA. The contribution of various foods to intake of vitamin A and carotenoids in The Netherlands. *Int J Vitam Nutr Res*. 1998;68:378-83.
- 23 Goldbohm RA, van den Brandt PA, Brants HA, van het Veer P, Al M, Sturmans F, et al. Validation of a dietary questionnaire used in large-scale prospective cohort study on diet and cancer. *Eur J Clin Nutr*. 1994;48:253-65.
- 24 Volovics A, van den Brandt PA. Methods for the analyses of case-cohort studies. *Biom. J*. 1997;2:195-214.
- 25 StataCorp: Stata. Statistical Software: Release 6.0. College Station, TX: Stata Corporation, 1999.
- 26 Barlow WE. Robust variance estimation for the case-cohort design. *Biometrics*. 1994;50:1064-72.
- 27 Lin DY, Ying Z. Cox regression with incomplete covariate measurements. *JASA*. 1993;88:1341-9.
- 28 Zeegers MP, Tan FE, Dorant E, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer*. 2000;89:630-9.
- 29 Zeegers MPA, Tan FES, Goldbohm RA, van den Brandt PA. Arc coffee and tea consumption associated with urinary tract cancer risk: a systematic review and meta-analysis. *Int J Epidemiol*. 2001;in press.
- 30 Zeegers MP, Tan FE, Verhagen AP, Weijenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.
- 31 Steinmaus CM, Nunez S, Smith AH. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol*. 2000;151:693-702.
- 32 Goldbohm RA, van 't Veer P, van den Brandt PA, van 't Hof MA, Brants HA, Sturmans F, et al. Reproducibility of a food frequency questionnaire and stability of dietary habits determined from five annually repeated measurements. *Eur J Clin Nutr*. 1995;49:420-9.

- 33 Michaud DS, Spiegelman D, Clinton SK, Rimm EB, Willet WC, Giovannucci E. Prospective study of dietary supplements, macronutrients, micronutrients, and risk of bladder cancer in US men. *Am J Epidemiol.* 2000;152:1145-53.
- 34 Steinmetz KA, Potter JD. Vegetables, fruit, and cancer. II. Mechanisms. *Cancer Causes Control.* 1991;2:427-42.
- 35 Patterson RE, White E, Kristal AR, Neuhauser ML, Potter JD. Vitamin supplements and cancer risk: the epidemiologic evidence. *Cancer Causes Control.* 1997;8:786-802.
- 36 Kamat AM, Lamm DL. Chemoprevention of urological cancer. *J Urol.* 1999;161:1748-60.
- 37 Birt DF. Update on the effects of vitamins A, C, and E and selenium on carcinogenesis. *Proc Soc Exp Biol Med.* 1986;183:311-20.
- 38 Risch HA, Burch JD, Miller AB, Hill GB, Steele R, Howe GR. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol.* 1988;127:1179-91.
- 39 Nomura AMY, Kolonel LN, Hankin JH, Yoshizawa CN. Dietary factors in cancer of the urinary tract. *Int J Cancer.* 1991;48:199-205.
- 40 Knekt P, Aromaa A, Maatela J, Aaran RK, Nikkari T, Hakama M, et al. Serum vitamin E and risk of cancer among Finnish men during a 10-year follow-up. *Am J Epidemiol.* 1988;127:28-41.
- 41 Glynn SA, Albanes D. Folate and cancer: a review of the literature. *Nutr Cancer.* 1994;22:101-19.
- 42 Blount BC, Mack MM, Wehr CM, MacGregor JT, Hiatt RA, Wang G, et al. Folate deficiency causes uracil misincorporation into human DNA and chromosome breakage: implications for cancer and neuronal damage. *Proc Natl Acad Sci U S A.* 1997;94:3290-5.

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Prediagnostig Toenail Selenium and Risk of Bladder Cancer

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ABSTRACT

Context: The association between several cancers and selenium status has been investigated in epidemiological studies. However, few results concerning bladder cancer have been reported so far.

Objective: To estimate the association between toenail selenium status and subsequent bladder cancer incidence.

Design: Prospective cohort study conducted from September 1986 to December 1992.

Setting: The study population originated from 204 municipal population registries throughout the Netherlands.

Participants: The cohort consisted of 120,852 men and women aged 55-69 at baseline. The multivariable case-cohort analysis was based on 431 bladder cancer cases and 2,459 subcohort members.

Main Outcome: Measure Incident microscopically confirmed bladder cancer, established by record linkage to cancer registries.

Results: The age-, sex- and smoking-adjusted RRs (95% CI) for increasing quintiles of toenail selenium were 1.00 (reference), 1.09 (0.80-1.48), 0.55 (0.38-0.79), 0.63 (0.43-0.91) and 0.67 (0.46-0.97), respectively (p -trend<0.01). Analyses with selenium as continuous variable supported these findings. An inverse association between toenail selenium and bladder cancer risk was most pronounced among ex-smokers (p -trend<0.01), was similar for subjects with high *vs.* low intakes of β -carotene, vitamin C and vitamin E, and was mainly confined to invasive transitional cell carcinomas of the urinary bladder, irrespective of tumor morphology.

Conclusion: The authors found evidence for an inverse association between selenium and bladder cancer risk.

INTRODUCTION

Bladder cancer is the most common urological cancer and is the seventh most common cancer among men, accounting for approximately 200,000 new cases per year worldwide.¹ Over the last four decades, many epidemiological studies and several reviews have been conducted to investigate determinants of bladder cancer.²⁻⁶ These studies suggested that bladder cancer is influenced by environmental factors, including cigarette smoking, fluid consumption, schistosomal infections and exposure to industrial chemicals (e.g., aromatic amines) and diet.

It has been suggested that selenium may protect against cancer, particularly because of its antioxidant properties.⁷⁻⁹ Selenium is an essential trace element required for normal function, growth, and reproduction.^{10,11} Several animal, ecologic and epidemiological studies support the hypothesis that low intake of selenium is associated with an increased risk of cancer.⁸ However, only five epidemiological studies have concentrated on urinary bladder cancer incidence.^{7-9,12,13}

In the present study, we were able to explore the relationship of prediagnostic toenail selenium to human bladder cancer prospectively with substantially more cases than in previous studies.

METHODS

Cohort. The Netherlands cohort study is a population based prospective study on diet and cancer started in the Netherlands in September 1986. The cohort includes 58,279 men and 62,573 women aged 55 to 69 years at the start of the study. The study population originated from 204 municipal population registries throughout the country. The case cohort approach was used for data processing and analysis.¹⁴ Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after the baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up, during the follow-up period. The study design, including data collection strategies, has been described in detail previously.¹⁵

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.¹⁶ The completeness of cancer follow-up was estimated to be over 96 percent.¹⁷ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 3,346 subcohort members (1,630 men and 1,716 women) and 619 incident cases (532 men and 87 women) with microscopically confirmed, incident carcinomas of the urinary bladder, ureters, renal pelvis or urethra were identified. Of these cases, 584 (94.3%) were diagnosed with bladder cancer of which 559 (95.7%) were transitional cell carcinomas. Because the overwhelming majority of tumors occurred in the urinary bladder and the ureters, renal pelvis and urethra are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Exposure status. At baseline, all cohort members completed a self-administered questionnaire on risk factors for cancer and potential confounding variables. The questionnaire data were key entered twice and processed in a standardized manner blinded with respect to case/subcohort status in order to minimize observer bias in coding and interpretation of the data. Toenail clippings had been provided by 438 patients with bladder cancer (76.8%) and by 2,569 (70.8%) subcohort members. In 1992, the toenail selenium analyses for subcohort members were carried out by the Interfaculty Reactor Institute (IRI) at Delft University (Delft, The Netherlands), using Instrumental Neutron Activation Analysis (INAA). This method, utilizing the 'Snelle

Buizenpost'-facility (SBP), and evaluation of accuracy and precision have been described elsewhere.¹⁸

In 1996, toenail selenium levels of bladder cancer cases were determined with the Carbonfiber Autonomous Facility for Irradiation and Analysis (CAFIA) also carried out by the IRL.¹⁹ The selenium determination for the cases was also based on the measurement of the induced radioactivity of the short-lived ^{77m}Se radionuclide (half-life 17.5 s), but cyclic activation and measurement was applied to achieve the required sensitivity.²⁰ Each sample passed 6 successive cycles of 17 s irradiation at a thermal neutron flux of $3.7 \cdot 10^{16} \text{ m}^{-2}\text{s}^{-1}$, 3 s decay and 17 s counting at 1 cm from a 40 % Ge detector. The results of the 6 countings were added. The accuracy of the INAA was checked by analysis of a certified bovine liver standard (Standard Reference Material 1,577b of the U.S. National Institute of Standards and Technology (NIST)). For 30 determinations, a mean value ($\pm$ sd) of $0.74 \pm 0.04 \text{ }\mu\text{g/g}$ selenium ($n=49$) was observed against a certified value of $0.73 \pm 0.06 \text{ }\mu\text{g/g}$.

Because of problems with the detection of selenium in samples weighing less than 10 mg, and sometimes in samples with very high calcium contents, 7 and 110 specimens were excluded from the bladder cancer and subcohort groups, respectively. Together with the case group, the toenail selenium levels for 40 subcohort members were assessed with the CAFIA-facility in addition to original assessment with the SBP-facility to evaluate comparability. The mean selenium level ($\pm$ sd) using the SBP-facility ($0.551 \pm 0.04 \text{ }\mu\text{g/g}$) was comparable to mean selenium levels using the CAFIA-facility ($0.552 \pm 0.05 \text{ }\mu\text{g/g}$) for these subjects. The Pearson correlation coefficient between toenail selenium levels assessed by the CAFIA-facility and that estimated by the SBP-facility was 0.95 ($p<0.01$). Toenail selenium data were available for analysis from 431 bladder cancer cases (372 men and 59 women) and 2,459 subcohort members (1,211 men and 1,248 women).

Statistical analyses. To evaluate the potential influence of prediagnostic cancer on toenail selenium levels we categorized the cases according to the year of follow-up in which the diagnosis was made. Mean toenail selenium levels of bladder cancer cases were compared according to year of follow-up. Toenail selenium levels were categorized into quintiles according to the distribution in the subcohort. Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models²¹ with the Stata statistical software package.²² Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow²³ and Lin and Ying.²⁴ Because of skewedness to the right, continuous data on toenail selenium concentrations were normalized with a $\log_e$ transformation. The RR was calculated per standard unit (z-score) of toenail selenium levels. This is comparable to an increment of $0.202 \text{ }\mu\text{g/g}$ selenium. We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against failure time.

The following variables were considered as potential confounders: intake of alcohol (g/day), coffee (ml/day), tea (ml/day) water (ml/day), vegetables (g/day) and fruit (g/day), current cigarette smoking (yes/no), smoking amount (cigarettes/day), smoking duration (years of cigarette smoking), occupational exposure to dye, rubber, leather or vehicle fumes (ever/never), and first degree family history of bladder cancer (yes/no). Those variables that showed a more than ten percent influence on the risk of bladder cancer when considered in a multivariable model were included as covariates in multivariable analyses. If one or more dietary covariates were incorporated in the regression model, subjects with incomplete or inconsistent dietary data were excluded, according to criteria described previously.²⁵ Subgroup analyses conditional on cigarette smoking status (never/ex/current) were performed to evaluate interaction of selenium by smoking. Since earlier studies^{13,26,27} suggested a potential interaction between vitamin levels and selenium we stratified the results by high *vs.* low β -carotene, vitamin C and vitamin E intake status of the subjects. Low and high are defined as the two lowest quintiles and the two highest quintiles of intake, respectively. Furthermore, subgroup analyses were

conducted after stratification by morphology and invasiveness of bladder cancer. Since the RRs concerning toenail selenium level and bladder cancer risk were similar for men and women, the results are presented for men and women combined.

RESULTS

Table 1 displays the average toenail selenium levels among bladder cancer cases according to sex and year of follow-up. Those of the subcohort are presented in a footnote of the table. The mean toenail selenium level for the cases (men: 0.536 µg/g, women: 0.568 µg/g) was lower than for the subcohort members (men: 0.547 µg/g, women: 0.575 µg/g). Cases diagnosed in the first years of follow-up had lower toenail selenium levels than cases diagnosed later after adjustment for sex (Table 1).

Table 1. Toenail selenium levels (µg/g) in bladder cancer cases according to sex and year of follow-up, Netherlands Cohort Study (1986-1992)

Cases*		No of subjects	Toenail selenium level (µg/g)		
			Mean	Standard deviation	p-value†
Sex					
	Men	372	0.536	0.215	reference
	Women	59	0.568	0.122	0.02
Year of follow-up‡					
	1	47	0.518	0.091	0.04
	2	61	0.523	0.263	0.21
	3	74	0.546	0.128	0.50
	4	75	0.544	0.155	0.90
	5	70	0.568	0.154	0.63
	6	84	0.590	0.721	} reference
	7	20	0.595	0.123	

*Mean (± standard deviation (sd)) selenium levels in subcohort members were 0.547 (± 0.126) µg/g for men (n=1,211) and 0.575 (± 0.109) µg/g for women (n=1,248)

†(partial) t tests between strata and reference group, based on ln-transformed toenail selenium levels

‡presented results per year of follow-up were adjusted for sex through linear regression analysis

Toenail selenium was inversely associated with bladder cancer risk in both men and women (Table 2). The age- and sex-adjusted RR was 0.54 (CI 0.38-0.76) comparing highest to lowest quintiles of toenail selenium. After adjustment for age, sex, number of cigarettes smoked per day and years of cigarette smoking the RR was 0.67 (CI 0.46-0.97) comparing the same quintiles of toenail selenium (p -trend<0.01). This reduced bladder cancer incidence appears to be limited to the highest three quintiles of the toenail selenium distribution. The RR decreased by seven percent for each increment of one standard unit of toenail selenium (0.202 µg/g). After exclusion of those patients whose cancer was diagnosed during their first year of follow-up, the negative association between toenail selenium level and bladder cancer risk persisted (Table 2). Additional correction for current cigarette smoking, coffee, tea, water, vegetables, fruit, occupational exposure and family history of bladder cancer did not change the risk substantially (data not shown).

Table 2. Incidence rate ratio (RR) of bladder cancer according to toenail selenium levels, Netherlands Cohort Study (1986-1992)

Quintiles of toenail selenium (boundaries in µg/g)	All years of follow-up		First year excluded		
	Cases in cohort	Person years in subcohort	RR (95% CI)*	RR (95% CI)†	RR (95% CI)†
1 (≤0.483)	114	2985	1.00 (reference)	1.00 (reference)	1.00 (reference)
2 (0.483-≤0.530)	116	3022	1.05 (0.79-1.40)	1.09 (0.80-1.48)	1.19 (0.86-1.64)
3 (0.530-≤0.573)	78	3031	0.51 (0.36-0.73)	0.55 (0.38-0.79)	0.61 (0.42-0.89)
4 (0.573-≤0.630)	62	2993	0.53 (0.37-0.76)	0.63 (0.43-0.91)	0.63 (0.42-0.94)
5 (>0.630)	61	3001	0.54 (0.38-0.76)	0.67 (0.46-0.97)	0.78 (0.54-1.14)
p-value for linear trend			<0.01	<0.01	<0.01
increment in standard units‡			0.86 (0.77-0.96)	0.94 (0.83-1.05)	0.89 (0.79-1.00)

*adjusted for age (years) and sex

†adjusted for age, sex, number of cigarettes/day, and years of cigarette smoking

‡z-scores based on ln-transformed toenail selenium levels (increment of 0.202 µg/g)

We did not find statistically significant interactions regarding bladder cancer risk between toenail selenium and cigarette smoking status ($p=0.19$), amount ($p=0.76$) or duration ($p=0.61$) (Table 3). However, an inverse association between toenail selenium and bladder cancer risk was limited to ex-smokers (p -trend<0.01) and no inverse association was found among never and current smokers (Table 3).

Table 3. Incidence rate ratio (RR)* of bladder cancer according to toenail selenium levels and smoking status, Netherlands Cohort Study (1986-1992)

Smoking Status	No. of Cases	Quintile of toenail selenium (boundaries in µg/g)					p-trend
		1 (≤0.483)	2 (≤0.530)	3 (≤0.573)	4 (0.630)	5 (>0.630)	
Never*	51	1.00‡	2.69 (1.02-7.09)	1.37 (0.50-3.79)	1.09 (0.38-3.14)	1.36 (0.50-3.69)	0.62
Ex†	186	1.00‡	0.74 (0.46-1.19)	0.48 (0.28-0.81)	0.49 (0.29-0.83)	0.44 (0.26-0.75)	<0.01
Current†	170	1.00‡	1.36 (0.86-2.14)	0.45 (0.24-0.86)	0.76 (0.39-1.47)	1.13 (0.56-2.27)	0.25

*adjusted for age (years) and sex

†adjusted for age (years), sex, number of cigarettes/day, and years of cigarette smoking

‡reference category

To evaluate effect modification of the association between toenail selenium and bladder cancer risk by the intake level of antioxidant vitamins β -carotene, vitamin C and vitamin E, we also evaluated whether the association with bladder cancer depended on intake of these antioxidants (Table 4). The inverse association between toenail selenium and bladder cancer risk persisted for subjects with different intakes of β -carotene, vitamin C and vitamin E (Table 4). We found no statistically significant interaction effect between these antioxidants and toenail selenium ($p=0.58$, 0.16, and 0.96, respectively).

Table 4. Incidence rate ratio (RR)* of bladder cancer according to toenail selenium levels by category of vitamin intake, Netherlands Cohort Study (1986-1992)

Group	No. of Cases	Quintile of toenail selenium (boundaries in µg/g)					p-trend
		1 (<0.483)	2 (≤0.530)	3 (≤0.573)	4 [0.630)	5 (>0.630)	
β-carotene intake*							
Low	167	1.00 [‡]	0.98 (0.59-1.62)	0.50 (0.28-0.89)	0.71 (0.40-1.26)	0.65 (0.36-1.16)	0.02
High	155	1.00 [‡]	1.08 (0.65-1.80)	0.97 (0.55-1.71)	0.66 (0.34-1.26)	0.74 (0.41-1.34)	0.09
Vitamin C intake*							
Low	181	1.00 [‡]	0.95 (0.59-1.53)	0.45 (0.26-0.78)	0.72 (0.41-1.26)	0.77 (0.43-1.37)	0.05
High	140	1.00 [‡]	1.09 (0.64-1.85)	1.01 (0.57-1.80)	0.50 (0.25-1.00)	0.75 (0.41-1.37)	0.06
Vitamin E intake*							
Low	145	1.00 [‡]	1.37 (0.81-2.33)	0.58 (0.30-1.09)	0.76 (0.41-1.42)	0.82 (0.45-1.50)	0.07
High	170	1.00 [‡]	1.09 (0.68-1.75)	0.59 (0.34-1.03)	0.70 (0.38-1.28)	0.73 (0.41-1.29)	0.04

*Adjusted for age (years), sex, number of cigarettes/day and years of cigarette smoking

†Low and high are defined as the two lowest quintiles and the two highest quintiles of intake, respectively. For β-carotene, the cutoff values for low and high were: ≤2.36, >2.97 mg/day; for vitamin C the corresponding values were: ≤86.60, >107.71 mg/day and for vitamin E the corresponding values were: ≤10.72, <13.89

‡reference category

An inverse association between toenail selenium level and bladder cancer risk was most pronounced in invasive transitional cell carcinomas of the urinary bladder, irrespective of tumor morphology (*p*-trend<0.01 and <0.01 for invasive papillary and non-papillary tumors, respectively) (Table 5). The RRs for non-invasive tumors did not indicate a clear inverse association for toenail selenium (Table 5).

Table 5. Incidence rate ratios (RR) for male transitional cell carcinoma of the urinary bladder according to toenail selenium levels, with respect to tumor invasiveness and morphology, Netherlands Cohort Study (1986-1992)

Quintiles of toenail selenium (boundaries in µg/g)	Non-Invasive (T _{1s} /T ₁)		Invasive (T ₂₋₄)	
	Non-papillary* (n=16)	Papillary† (n=180)	Non-papillary* (n=81)	Papillary† (n=108)
	RR (95% CI)‡	RR (95% CI)‡	RR (95% CI)‡	RR (95% CI)‡
1 (<0.483)	1.00 (reference)	1.00 (reference)	1.00 (reference)	1.00 (reference)
2 (0.483-≤0.530)	1.24 (0.31-4.88)	1.23 (0.80-1.88)	1.11 (0.63-1.95)	0.96 (0.58-1.59)
3 (0.530-≤0.573)	1.07 (0.25-4.58)	0.62 (0.37-1.05)	0.45 (0.21-0.96)	0.44 (0.23-0.84)
4 (0.573-≤0.630)	1.28 (0.26-6.42)	0.78 (0.46-1.31)	0.17 (0.05-0.57)	0.64 (0.34-1.20)
5 (>0.6300)	0.83 (0.13-5.20)	0.91 (0.55-1.50)	0.62 (0.30-1.28)	0.36 (0.17-0.75)
p-value for linear trend	0.92	0.18	<0.01	<0.01
increment in standard units†	1.08 (0.74-1.58)	1.06 (0.87-1.29)	0.84 (0.69-1.02)	0.82 (0.72-0.94)

†ICD-O:M8120, †ICDO:M8130

‡adjusted for age, sex, number of cigarettes smoked per day, and years of cigarette smoking

DISCUSSION

These data showed an inverse association between base-line toenail selenium and subsequent bladder cancer risk. The association was most pronounced in ex-smokers and mainly confined to invasive carcinomas of the bladder. The association appeared to be independent of β -carotene, vitamin C and vitamin E intake.

Preclinical symptoms of bladder cancer might influence selenium status. However, one advantage of this and other prospective studies is that selenium status was assessed before diagnosis of bladder cancer. In the present study, cases diagnosed in the first years of follow-up had lower toenail selenium levels than cases diagnosed later. We found a strong inverse association between selenium status and first year bladder cancer incidence (data not shown). After exclusion of cases diagnosed in the first year of follow-up, the diluted results were similar to results including all cases. Selection bias is not likely, because the follow-up for cancer incidence and person years was almost complete.^{17,28} The large number of bladder cancer cases in this study was an other important advantage.

Diet-assessment methods are inadequate to assess selenium intake because selenium content of an individual food can vary widely due to geographic variation in soil selenium content. Therefore, previous epidemiological studies on the relation between selenium and bladder cancer have often relied on biological markers of selenium status, such as serum selenium^{7,8,13} or toenail selenium.¹² A selenium marker should preferably reflect long-term selenium intake. For this reason, we used levels of selenium toenails to assess selenium intake instead of serum selenium, a short-term marker of selenium intake.¹⁸ Available evidence suggests that selenium levels in toenails reflect intake integrated for the previous 12 months or longer.¹²

We were not able to explain our results on the basis of confounding, since our results were essentially unchanged after incorporating into the analyses many known or suspected risk factors for bladder cancer, including current cigarette smoking, total consumption of alcohol, water, vegetables and fruit, high risk occupation and positive family history of bladder cancer (data not shown). We made an attempt to model cigarette smoking habits such that they best explained bladder cancer. This resulted in a model including number of years smoked and habitual number of cigarettes smoked per day, both as continuous variables. When we added the smoking variables to an age and sex-adjusted model, the RR estimates changed only slightly. We therefore believe that associations observed were not entirely due to residual confounding by smoking, although we cannot exclude some influence. It should be considered that we couldn't completely eliminate confounding bias, since the inverse association found between low selenium levels and risk of bladder cancer may also indicate the protective effect of another nutrient in selenium-rich foods.²⁶

Recently, Cohen²⁹ suggested that transitional cell carcinoma of the urinary bladder might represent two distinct diseases: papillary transitional cell carcinoma, which is usually non-invasive, and the non-papillary type, which tends to be invasive. In addition to different prognostic and pathogenic properties, these two diseases appear to involve different molecular events.²⁹ From an etiological point of view some distinction might be important. An inverse association between toenail selenium level and bladder cancer risk was most pronounced in invasive transitional cell carcinomas of the urinary bladder, irrespective of tumor morphology. Further research is needed to evaluate this matter.

The association between total cancer incidence and selenium levels has been investigated in several prospective epidemiological studies. However, few results concerning urinary bladder cancer per se have been reported so far^{7,8,12,13} (Table 6). Although the number of bladder cancer cases in these studies was small and the mean selenium levels differed substantially between the studies, most studies, as in the present study, demonstrated an inverse association between selenium concentrations and the risk of bladder cancer. Nomura et al¹³ reported a decrease in relative risk of bladder cancer for subjects in the highest quintile compared to the lowest quintile of serum selenium (RR=0.32), although no statistically significant trend was found ($p=0.12$). Helzouer et al⁷ similarly reported a decreased bladder cancer risk with increasing serum levels ($p=0.03$). When examined by

tertiles, the relative risk associated with the highest tertile of selenium compared to the lowest tertile was 0.49. A Finnish study⁸ demonstrated a protective effect of selenium for men (RR=0.34) but not for women (RR=2.51) comparing the four highest quintiles against the lowest quintile of serum selenium, although the latter is based on only nine bladder cancer cases. Of interest in this connection is the fact that serum selenium levels in this Finnish study were approximately one-half of those found in the United States. Only one prospective study investigated the association between toenail selenium levels and female urinary tract cancer.¹² This study did not find an association between toenail selenium and bladder cancer risk ($p=0.58$)¹² (Table 6).

Recently, Clark et al⁹ conducted a randomized, placebo-controlled cancer prevention trial in low-selenium areas of the United States to investigate whether selenium supplementation would decrease cancer incidence. The investigators demonstrated that, compared with controls, subjects treated with selenium had a non-significant reduction in total cancer mortality (RR=0.50) and total cancer incidence (RR=0.63). For bladder cancer, the actual number of cases was too small to perform statistically meaningful analyses.⁹ Although promising, further human experimental research is needed to evaluate this matter.

The causal interpretation of an inverse association between selenium and cancer risk is supported by several animal experiments demonstrating anticarcinogenic effects of selenium, although bladder carcinogenesis has not often been investigated.³⁰ Several hypotheses have been proposed to explain the inhibition of tumorigenesis by selenium, including alterations in carcinogen metabolism, effects on the endocrine and immune systems, production of cytotoxic selenium metabolites, inhibition of protein synthesis, inhibition of specific enzymes, inhibition of tumor growth, stimulation of apoptosis and protection against oxidative stress involving the function of selenium as a constituent in the antioxidant enzyme glutathione peroxidase.^{7,9-12,26,31,32} The latter mechanism has received most attention. The lack of effect of selenium status among non-smokers is consistent with this hypothesis since never smokers have not been exposed to smoking induced oxidative stress,³³ although lack of effect among current smokers is not. However, these differences might be attributable the chance since no statistically significant effect modification was found.

The authors found evidence for an inverse association between toenail selenium and bladder cancer risk. The decreased cancer incidence seems largely limited to the highest three quintiles of the toenail selenium distribution. The association was most pronounced in ex-smokers and mainly confined to invasive carcinomas of the bladder. It appeared to be independent of β -carotene, vitamin C and vitamin E intake.

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REFERENCES

1. Parkin DM, Pisani P, Ferlay J. Estimates of the worldwide incidence of 25 major cancers in 1990. *Int J Cancer*. 1999;80:827-841.
2. Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am*. 1992;6:1-30.
3. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol*. 1997;13:291-298.
4. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol*. 1996;23:536-545.

5. World Cancer Research Fund, American Institute for Cancer Research. Bladder. Food, Nutrition and the Prevention of Cancer: a global perspective. Menasha: Banta Book Group; 1997:338-361.
6. van der Meijden APM. Bladder cancer. *Br Med J*. 1998;317:1366-1369.
7. Helzlsouer KJ, Comstock GW, Morris JS. Selenium, lycopene, alpha-tocopherol, beta-carotene, retinol, and subsequent bladder cancer. *Cancer Res*. 1989;49:6144-6148.
8. Knekt P, Aromaa A, Maatela J, et al. Serum selenium and subsequent risk of cancer among Finnish men and women. *J Natl Cancer Inst*. 1990;82:864-868.
9. Clark LC, Combs GF, Jr., Turnbull BW, et al. effects of selenium supplementation for cancer prevention in patients with carcinoma of the skin: a randomized controlled trial. *JAMA*. 1996;276:1957-1963.
10. Fan AM, Kizer KW. Selenium: nutritional, toxicologic and clinical aspects. *West J Med*. 1990;153:160-167.
11. Virtamo J, Valkeila E, Alfthan G, Punsar S, Huttunen JK, Karvonen MJ. Serum selenium and risk of cancer. A prospective follow-up of nine years. *Cancer*. 1987;60:145-148.
12. Garland M, Morris JS, Stampfer MJ, et al. Prospective study of toenail selenium and cancer among women. *J Natl Cancer Inst*. 1995;87:497-505.
13. Nomura A, Heilbrun LK, Morris JS, Stemmermann GN. Serum selenium and the risk of cancer, by specific sites: case-control analysis of prospective data. *J Natl Cancer Inst*. 1987;79:103-108.
14. Barlow WE, Ichikawa L, Rosner D, Izumi S. Analysis of case-cohort designs. *J Clin Epidemiol*. 1999;52:1165-1172.
15. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, Hermus RJJ, Sturmans F. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol*. 1990;43:285-295.
16. van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, Hunen PHM. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol*. 1990;19:553-558.
17. Goldbohm RA, van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond*. 1994;72:80-84.
18. van den Brandt PA, Goldbohm RA, van't Veer P, Bode P, Hermus RJ, Sturmans F. Predictors of toenail selenium levels in men and women. *Cancer Epidemiol Biomarkers Prev*. 1993;2:107-112.
19. Bode P, de Bruin M. An Automated system for activationanalysis with short half life radionuclides using a carbonfiber irradiation facility. *J Radioanal Chem*. 1987;132:365-375.
20. Spyrou NM. Activation Analysis. *J Radioanal Chem*. 1981;61:211-242.
21. Volovics A, van den Brandt PA. Methods for the analyses of case-cohort studies. *Biom J*. 1997;2:195-214.
22. StataCorp. Stata Statistical Software: Release 6.0. College Station, TX: Stata Corporation; 1999.
23. Barlow WE. Robust variance estimation for the case-cohort design. *Biometrics*. 1994;50:1064-1072.
24. Lin DY, Ying Z. Cox regression with incomplete covariate measurements. *JASA*. 1993;88:1341-1349.
25. Goldbohm RA, van den Brandt PA, Brants HA, et al. Validation of a dietary questionnaire used in large-scale prospective cohort study on diet and cancer. *Eur J Clin Nutr*. 1994;48:253-265.
26. Willett WC, Polk BF, Morris JS, et al. Prediagnostic serum selenium and risk of cancer. *Lancet*. 1983;2:130-134.
27. van den Brandt PA, Goldbohm RA, van't Veer P, et al. Toenail selenium levels and the risk of breast cancer. *Am J Epidemiol*. 1994;140:20-26.
28. van den Brandt PA, van het Veer P, Goldbohm RA. A prospective cohort study on dietary fat and the risk of postmenopausal breast cancer. *Cancer Res*. 1993;53:75-82.
29. Cohen SM. Urinary Bladder carcinogenesis. *Toxicol Pathol*. 1998;26:121-127.
30. Mohandas J, Marshall JJ, Duggin GG, Horvath JS, Tiller DJ. Low activities of glutathione-related enzymes as factors in the genesis of urinary bladder cancer. *Cancer Res*. 1984;44:5086-5091.
31. Patterson RE, White E, Kristal AR, Neuhaus ML, Potter JD. Vitamin supplements and cancer risk: the epidemiologic evidence. *Cancer Causes Control*. 1997;8:786-802.
32. Salonen JT, Alfthan G, Huttunen JK, Puska P. Association between serum selenium and the risk of cancer. *Am J Epidemiol*. 1984;120:342-349.
33. van den Brandt PA, Goldbohm RA, van't Veer P, et al. A prospective cohort study on selenium status and the risk of lung cancer. *Cancer Res*. 1993;53:4860-4865.

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Occupational Risk Factors for Male Bladder Cancer: Results from a Population Based Cas Cohort Study in the Netherlands

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ABSTRACT

Objectives: This study was conducted to estimate bladder cancer risk associated with occupational exposures to paint components, polycyclic aromatic hydrocarbons (PAH), diesel exhausts and aromatic amines among the general population in the Netherlands.

Methods: The authors conducted a prospective cohort study among 58,279 men. In September 1986, the cohort members (55-69 years) completed a self-administered questionnaire on cancer risk factors including job history. Follow-up for incident bladder cancer was established by linkage to cancer registries until December 1992. A case-cohort approach is used based on 532 cases and 1,630 subcohort members. A case-by-case expert assessment was carried out to assign the cases and subcohort members a cumulative probability of occupational exposure (CPE) for each carcinogenic exposure.

Results: Men in the highest tertiles of occupational exposure to paint components, PAH, aromatic amines and diesel exhaust had non-significantly higher age- and smoking-adjusted RRs of bladder cancer than men with no exposure: 1.29 (95%CI:0.71-2.33), 1.24 (95%CI:0.68-2.27), 1.32 (95%CI:0.41-4.23) and 1.21 (95%CI:0.78-1.88), respectively. The associations between paint components and PAH and bladder cancer risk were most pronounced for current smokers. Among former smokers it appeared that men who had smoked more than 15 cigarettes/day had RRs below unity for CPE to paint components and PAH compared with men who had smoked <15 cigarettes/day, whereas among current smokers the opposite was observed. Exposure to diesel exhaust was positively associated with bladder cancer risk among current and former smokers who had smoked more than 15 cigarettes per day.

Conclusions: This study provided only marginal evidence for an association between occupational exposure to paint components, PAH, aromatic amines and bladder cancer. Despite the small proportion of exposed subjects, the authors found interaction effect with cigarette smoking, specifically for paint components, suggesting that the carcinogenic effect on the bladder might decrease after smoking cessation.

INTRODUCTION

Bladder cancer is the most common urological cancer and is the seventh most common cancer among men, accounting for approximately 200,000 new cases worldwide annually.¹ Bladder cancer is considerably more common in men than in women (worldwide ratio is about 3.5:1), which by itself has been regarded as a possible indication for an occupational origin.² The incidence of this cancer varies worldwide; in general, the highest incidence is observed in North America and Europe.¹

Bladder cancer has frequently been reported to be associated with occupational exposures.³ As early as the late nineteenth century doctors have reported unusual incidences of bladder cancer in industry.⁴ In his classic study Case⁵ reported an exceptionally high incidence of bladder cancer in the British rubber industry. He was one of the first epidemiologists to apply a retrospective cohort design to investigate the effect of occupational exposure to a possible carcinogen.⁵ Later several other agents or occupations have been associated with an elevated risk of bladder cancer. Among these were exposure to polycyclic aromatic hydrocarbons in the aluminum industry, coal tar industry and coal gasification industry.³ In addition, a substantial number of studies have reported an excess of bladder cancer among painters,⁶⁻⁹ although others could not confirm this association.^{10,11} Most epidemiologic studies of the risk for bladder cancer among painters reported relative risks between 1.2 and 1.5.³ In some studies increased risks for bladder cancer were observed for increasing duration of exposure to paint components.^{12,13} Finally, a substantial number of epidemiologic studies have reported more bladder cancer among truck drivers, taxi drivers and bus drivers.¹⁴⁻¹⁶ Overall relative risks for bladder cancer in studies of these occupational groups varied from 1.3 to 2.2. Most studies observed positive trends with duration of exposure.³ The most likely causal agent is thought to be constituents of the diesel exhaust emissions. Again, these associations have not always been confirmed and are still a matter of debate.

Most epidemiologic studies that investigated associations between specific occupational exposures and bladder cancer applied retrospective designs; sometimes prospective cohorts have been used. The possible confounding effects of exposure to other carcinogenic factors, such as cigarette smoking and dietary factors that are suspect of influencing the occurrence of bladder cancer were only rarely taken into account. Epidemiologic studies that did take into account the possible confounding effect of non-occupational exposures used case-control designs, possibly suffering from a recall bias, since data collection took place after the diagnosis of the disease. We have used data from the Netherlands Cohort Study, a prospective cohort study in the general population on diet, lifestyle, job history and cancer to study the relations between various occupational exposures and the incidence of bladder cancer, adjusting for the effects of a number of other risk factors.

Because of the nature of the database, which is not focused on any particular occupational group, we restricted the study to occupational exposures that were expected to be quit common in the general population. Occupational exposures that were thought to be very rare were regarded to be beyond the scope of this study. We focused the study on specific occupational exposures from which it was either expected that these exposures would be relatively common or that the relative risk for bladder cancer could be so high that this risk could be detected in a general population study. Therefore, we selected exposure to paint components, polycyclic aromatic hydrocarbons (PAH), diesel exhaust and aromatic amines as potential risk factors for bladder cancer.

METHODS

Study population. The study design, including data collection strategies, has been described in detail previously.¹⁷ In short, the cohort includes 58,279 men aged 55 to 69 years at baseline. The study population originated from 204 municipal population registries throughout the Netherlands. The case cohort approach was used for data processing and analysis.¹⁸ Cases were enumerated from the entire cohort, while the

accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 1,688 men was randomly sampled from the cohort after baseline exposure measurement. The subcohort has been followed up for vital status information. No subcohort members were lost to follow-up.

Follow-up. Follow-up for incident cancer was established by record linkage to cancer registries and the Dutch national database of pathology reports.¹⁹ The completeness of cancer follow-up was estimated to be over 95 percent.²⁰ The presented analysis is restricted to cancer incidence in 6.3 years of follow-up, from September 1986 to December 1992. After excluding prevalent cases with cancer other than skin cancer a total of 1,630 male subcohort members and 532 male incident cases with microscopically confirmed carcinomas of the urinary bladder, ureters, renal pelvis or urethra were identified. Of these cases, 506 (95%) were diagnosed with bladder cancer of which 490 (97%) were transitional cell carcinomas. Because the overwhelming majority of tumors occurred in the urinary bladder and since the ureters, renal pelvis and urethra are covered by the same urothelium as the urinary bladder, the term bladder cancer is used as a synonym for these neoplasms.

Exposure assessment. At baseline, the cohort members completed a mailed, self-administered questionnaire on potential confounders and other risk factors for cancer. In this questionnaire, job history was addressed by questions on job-title, name of the company, type of the company, time period and information about what was being produced at the department. Experts in the fields of occupational epidemiology (G.M.H.S.) and occupational hygiene (I.J.K.) assessed the probability of four carcinogenic exposures: paint components, PAH, diesel exhaust, and aromatic amines without knowledge of the disease status of the subjects.²¹ First a list of all jobs in the Netherlands was independently reviewed. In this first examination all jobs with no potential carcinogenic exposures as mentioned above were excluded. Secondly, a more refined exposure assessment was conducted based on information about company name and type, time period, and product, since the probability of carcinogenic exposure may differ between types of industry and between time periods. Four exposure categories were defined: no exposure to the specific carcinogen, possible exposure (probability of exposure estimated to be lower than 30%), probable exposure (probability of exposure between 30% and 90%) and nearly certain exposure (probability of exposure over 90%). Typical examples of high exposure classifications were: furniture maker, engraver and maintenance painter for exposure to paint components; smith, coal miner, metal foundry worker, maintenance working in coking factory for exposure to PAH; marine engineer, car mechanic and truck driver for exposure to diesel exhausts; and vulcanizer in rubber factory and cable factory worker for exposure to aromatic amines. For a quantification of exposure a cumulative probability of exposure (CPE) was calculated, which combines information about the probability of exposure and the duration of possible exposure. Therefore a weight was assigned to each exposure category: no exposure – weight 0; possible exposure – weight 0.15; probable exposure – weight 0.6 and nearly certain exposure – weight 0.95. Each weight corresponds to the midpoint of probability in each exposure category. The CPE was calculated by multiplication of the weight given to each exposure category by the number of years exposed. Subsequently, for each person all weighted exposures were summed up for each of the four carcinogens separately and the exposed subjects were categorized in tertiles of exposure index.

Statistical analyses. Incidence rate ratios (RR) and corresponding 95% confidence intervals (CI) for bladder cancer were estimated using exponentially distributed failure time regression models²² with the Stata statistical software package.²³ Standard errors were estimated using the robust Hubert-White sandwich estimator to account for additional variance introduced by sampling from the cohort. This method is equivalent to the variance-covariance estimator presented by Barlow²⁴ and Lin and Ying.²⁵ We confirmed constancy of the baseline hazard visually by plotting the natural logarithm of the baseline survival function against hazard time.

The following variables were considered as potential confounders, based on earlier analyses:²⁶⁻²⁹ age (years), cigarette smoking amount (cigarettes/day), cigarette smoking duration (years of cigarette smoking), consumption of total fluid (ml/day), alcohol (g/day), coffee (ml/day), tea (ml/day), vegetables (g/day), fruits (g/day), and first degree family history of bladder cancer (yes/no). Those variables that showed a more than ten percent influence on the risk of bladder cancer when considered in a multivariable model were included as covariates in multivariable analyses.

RESULTS

Table 1 shows the distribution of the cumulative probability of exposure to each of the four carcinogens in the case and the subcohort group. The percentage of exposed persons to paint components, PAH and diesel exhaust were approximately equal between cases and subcohort members. However, there were relatively more cases in the highest exposure tertiles than subcohort members. Cases were also more often exposed to aromatic amines than subcohort members (Table 1).

Table 1. Distribution of categories of cumulative probability of exposure* to paint components, polycyclic aromatic hydrocarbons, diesel exhaust and aromatic amines in male bladder cancer cases and subcohort members, Netherlands Cohort Study (1986-1992)

Exposure	Cases (n=619)		Subcohort (n=1630)	
	N	%	N	%
Paint components				
0 No exposure	483	91.13	1494	92.00
1 Exposure tertile (low)	8	1.51	43	2.65
2 Exposure tertile	20	3.77	44	2.71
3 Exposure tertile (high)	19	3.58	43	2.65
PAH [†]				
0 No exposure	488	92.60	1477	90.89
1 Exposure tertile (low)	7	1.33	51	3.14
2 Exposure tertile	13	2.47	49	3.02
3 Exposure tertile (high)	19	3.61	48	2.95
Diesel Exhaust				
0 No exposure	428	81.37	1319	81.42
1 Exposure tertile (low)	35	6.65	105	6.48
2 Exposure tertile	31	5.89	96	5.93
3 Exposure tertile (high)	32	6.08	100	6.17
Aromatic Amines				
0 No exposure	522	98.31	1611	98.83
1 Low exposure	3	0.56	10	0.61
2 High exposure	6	1.13	9	0.55

*Product of probability (weights 0.15, 0.6, and 0.95) and duration (years) of exposure

[†]polycyclic aromatic hydrocarbons

The distribution of potential bladder cancer confounders according to occupational exposure to paint components, PAH, diesel exhaust and aromatic amines is presented in table 2. The confounders were equally distributed among never and ever exposed subcohort members for all investigated carcinogenic exposures. Exposure to paint components was correlated with exposure to PAH and diesel exhaust. Diesel exhaust was furthermore correlated to PAH and aromatic amines (Table 2).

Table 2. Association between possible confounders and occupational exposure in the subcohort, Netherlands Cohort Study (1986-1992)

Probability of exposure	Paint components		PAH*		Diesel Exhaust		Aromatic amines	
	Never	Ever	Never	Ever	Never	Ever	Never	Ever
Potential confounders:								
Age (mean years)	61.40	61.28	61.45	60.74	61.44	61.16	61.38	62.37
Smoking amount (mean cig/day)*	16.95	16.73	16.90	17.33	16.90	17.00	16.96	15.89
Smoking duration (mean years)*	33.62	33.58	33.64	32.90	33.75	33.07	33.58	33.83
Total fluid consumption (mean l/day)	2124	2151	2127	2115	2114	2178	2125	2153
Alcohol consumption (mean g/day)	14.42	14.84	14.39	15.01	14.60	13.53	14.43	16.65
Coffee consumption (mean cups/day)	45.53	47.76	45.52	47.39	45.02	48.64	45.71	46.32
Tea consumption (mean cups/day)	24.78	23.83	25.09	20.75	24.90	23.69	24.65	27.37
Vegetable consumption (g/day)	189.8	182.6	188.0	201.5	186.3	201.6	188.8	218.5
Fruit consumption (g/day)	152.6	138.2	151.3	154.7	149.2	161.8	151.3	165.5
Exposure to:								
Paint components (% ever)	N/A [†]	N/A [†]	5.13	40.76	5.19	21.57	8.19	7.14
PAH (% ever)	5.56	42.86	N/A [†]	N/A [†]	4.03	28.68	8.53	14.29
Diesel exhaust (% ever)	15.82	48.57	14.43	61.75	N/A [†]	N/A [†]	18.29	32.14
Aromatic Amines (% ever)	1.32	1.14	1.23	2.17	1.09	2.28	N/A [†]	N/A [†]

*polycyclic aromatic hydrocarbons
[†]only for current and former cigarette smokers
[‡]not applicable

Table 3 shows the associations of occupational exposure to bladder cancer risk. The age- and cigarette smoking-adjusted incidence rate ratio of bladder cancer was 1.29 (CI 0.71-2.33) comparing the highest tertile of CPE to paint components with no occupational exposure (p -trend=0.08). After additional adjustment for other occupational exposures, the RR changed to 1.31 (CI 0.72-2.40) (p -trend=0.09). Men with a higher cumulative probability of exposure to aromatic amines were also found to be at increased risk of bladder cancer, but the estimates were based on small numbers (age- and smoking-adjusted RR=1.32, CI 0.41-4.23). The corresponding age-, smoking- and occupation-adjusted RR was 1.36 (CI 0.42-4.35). Exposure to PAH or diesel exhaust did not substantially increase bladder cancer risk (p -trend=0.80 and 0.50, respectively). Men in the highest exposure categories of PAH had non-significantly higher risks of bladder cancer than men with no occupational exposure to PAH (age- and smoking-adjusted RR=1.24, CI 0.68-2.27) and diesel exhaust (age- and smoking-adjusted RR=1.21, CI 0.78-1.88). After adjustment for other occupational exposures and cigarette smoking the RRs changed to 1.18 (CI 0.62-2.24) and 1.17 (CI 0.74-1.84), respectively (Table 3). Additional correction for total fluid, alcohol coffee, tea, vegetables and fruit consumption did not essentially change the risk estimates for the carcinogenic exposures (data not shown).

The associations between CPE to paint components, PAH and bladder cancer risk were more explicit for current smokers compared with never or former smokers (Table 4). It was not possible to stratify CPE to aromatic amines on cigarette smoking characteristics, because of sparsity of data. Among the former smokers it appeared that men who smoked more than 15 cigarettes per day had lower incidence rate ratios for CPE to paint components and PAH compared with men who had smoked less than 15 cigarettes daily, whereas among current smokers the opposite was observed. The positive association between paint components, PAH and bladder cancer risk increased by increasing number of cigarettes smoked among current smokers. Exposure to diesel exhaust was positively associated with bladder cancer risk among current smokers and among men who smoked more than 15 cigarettes per day, irrespective of smoking status (Table 4).

Table 3. Incidence rate ratios for bladder cancer according to occupational exposures in age-adjusted and multivariable analysis, Netherlands Cohort Study (1986-1992)

Life time exposure index*	Cases	Subcohort (person years)	RR (95% CI) [†]	RR (95% CI) [‡]	RR (95% CI) [§]
Paint components					
0 No exposure	483	8967	1.00 (reference)	1.00 (reference)	1.00 (reference)
1 Exposure tertile (low)	8	258	0.67 (0.31-1.44)	0.74 (0.33-1.68)	0.75 (0.33-1.72)
2 Exposure tertile	20	262	1.37 (0.79-2.39)	1.76 (0.96-3.24)	1.78 (0.94-3.37)
3 Exposure tertile (high)	19	257	1.23 (0.71-2.14)	1.29 (0.71-2.33)	1.31 (0.72-2.40)
p-trend			0.25	0.08	0.09
PAH**					
0 No exposure	488	8864	1.00 (reference)	1.00 (reference)	1.00 (reference)
1 Exposure tertile (low)	7	303	0.47 (0.21-1.04)	0.47 (0.21-1.06)	0.51 (0.22-1.19)
2 Exposure tertile	13	298	0.85 (0.45-1.59)	0.85 (0.44-1.62)	0.97 (0.49-1.90)
3 Exposure tertile (high)	19	286	1.24 (0.72-2.13)	1.24 (0.68-2.27)	1.18 (0.62-2.24)
p-trend			0.98	0.80	0.85
Diesel Exhaust					
0 No exposure	428	7910	1.00 (reference)	1.00 (reference)	1.00 (reference)
1 Exposure tertile (low)	35	643	1.00 (0.68-1.51)	0.98 (0.64-1.51)	1.00 (0.65-1.54)
2 Exposure tertile	31	580	0.99 (0.65-1.52)	0.98 (0.62-1.55)	0.96 (0.60-1.53)
3 Exposure tertile (high)	32	586	1.07 (0.70-1.62)	1.21 (0.78-1.88)	1.17 (0.74-1.84)
p-trend			0.78	0.50	0.76
Aromatic Amines					
0 No exposure	522	9660	1.00 (reference)	1.00 (reference)	1.00 (reference)
1 Low exposure	3	63	0.77 (0.21-2.85)	0.63 (0.12-3.13)	0.64 (0.13-3.22)
2 High exposure	6	54	2.00 (0.68-5.88)	1.32 (0.41-4.23)	1.36 (0.42-4.35)
p-trend			0.24	0.78	0.72

*product of probability (weights 0.15, 0.6, and 0.95) x duration (years) of exposure

[†]adjusted for age

[‡]adjusted for age, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

[§]adjusted for age, other occupational exposures, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

**polycyclic aromatic hydrocarbons

Table 4. Incidence rate ratios for bladder cancer for occupational exposures (non-exposed *vs.* ever exposed) stratified upon cigarette smoking status and cigarette smoking amount, Netherlands Cohort Study (1986-1992)

Cigarette smoking	Paint components		PAH*		Diesel Exhaust	
	Cases (exp./non) [†]	RR (95% CI)	Cases (exp./non) [†]	RR (95% CI)	Cases (exp./non) [†]	RR (95% CI)
Never smoker [‡]	2/34	1.35 (0.26-7.12)	0/36	Too few cases	2/34	0.51 (0.11-2.35)
Smoking Status						
Former [§]	13/223	0.65 (0.34-1.26)	15/221	0.67 (0.36-1.27)	46/190	1.01 (0.68-1.49)
<15 [‡] cigarettes / day	5/64	1.13 (0.37-3.43)	4/64	0.59 (0.19-1.79)	7/61	0.38 (0.11-1.28)
≥15 [‡] cigarettes / day	8/151	0.42 (0.17-1.03)	11/149	0.25 (0.00-4.28)	38/122	1.37 (0.74-2.56)
Current**	32/223	2.22 (1.15-4.29)	24/228	1.16 (0.60-2.23)	50/201	1.24 (0.81-1.90)
<15 [‡] cigarettes / day	9/65	1.40 (0.50-3.93)	7/67	1.37 (0.25-7.44)	15/57	0.99 (0.41-2.43)
≥15 [‡] cigarettes / day	20/129	3.53 (1.52-8.22)	16/131	1.61 (0.62-4.17)	29/119	1.22 (0.63-2.39)

*polycyclic aromatic hydrocarbons

[†]number of exposed and non-exposed bladder cancer cases included in the analysis

[‡]adjusted for age and other occupational exposures

[§]adjusted for age, other occupational exposures, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

**adjusted for age, other occupational exposures, and cigarette smoking duration (years)

The positive association between CPE to paint components and transitional cell carcinoma of the bladder was similar for papillary and non-papillary (invasive) tumors. A positive association between CPE to PAH and bladder cancer risk was most pronounced in non-papillary invasive transitional cell carcinomas of the urinary bladder. The positive association between CPE to diesel exhaust and bladder cancer risk was most pronounced among men with non-papillary non-invasive tumors (Table 5). Subgroup analyses for CPE to aromatic amines with respect to tumor invasiveness and morphology were not possible because too few cases were exposed to aromatic amines.

Table 5. Incidence rate ratios for male transitional cell carcinoma of the urinary bladder for occupational exposures (non-exposed *vs.* ever exposed) with respect to tumor invasiveness and morphology, Netherlands Cohort Study (1986-1992)

Probability of exposure	Non-Invasive (T _{1s} / T _a / T ₁)		Invasive T ₂₋₄	
	Non-papillary (n=28)*	Papillary (n=248)*	Non-papillary (n=122)*	Papillary (n=162)*
	RR (95% CI)†	RR (95% CI)†	RR (95% CI)†	RR (95% CI)†
Paint components	Too few cases	1.45 (0.83-2.53)	1.34 (0.69-2.62)	1.18 (0.54-2.56)
PAH‡	Too few cases	0.69 (0.34-1.43)	1.59 (0.77-3.28)	0.51 (0.19-1.36)
Diesel exhaust	1.72 (0.51-5.82)	0.82 (0.49-1.36)	1.07 (0.54-2.12)	1.17 (0.65-2.12)

†ICD-O:M8120

‡ICDO:M8130

†adjusted for age, other occupational exposures, cigarette smoking amount (cig/day), and cigarette smoking duration (years)

‡polycyclic aromatic hydrocarbons

DISCUSSION

In this study we examined the association between four occupational exposures and the incidence of bladder cancer. The results suggest a possible positive association between cumulative exposure to paint components, PAH and aromatic amines and the incidence of bladder cancer, although statistical significance was not reached. Only very limited support, if any, was found for an association between diesel exhaust and bladder. If there is an association between diesel exhaust and bladder cancer at all, the results indicate that it may be limited to non papillary, non invasive bladder cancer. Statistical adjustment for the effect of other potential risk factors tended rather to increase the incidence rate ratios than to decrease the estimates.

Until now, many studies on occupational risk factors for bladder cancer have been conducted.³⁰ For the most part, epidemiologic evidence has been derived from either job titles (e.g., obtained through a Job Exposure Matrix (JEM)) or self-assessed exposures to certain agents. In only a few studies a case-by-case expert evaluation of exposure to occupationally related agents was performed.^{8,31-34} The quality of the exposure assessment strongly affects the outcome of the risk estimate³⁵ and ideally includes both intensity and duration of the occupational exposure for each specific study subject. However, in large population-based studies it is almost impossible to obtain this quantitative information. Moreover, in this type of study the range of jobs with potential exposures is large and within given jobs there is a large variation in exposures. This makes it even more difficult to identify a clear link between jobs and exposures as compared to industry based studies.^{36,37} Self-assessment of occupational exposures is considered inadequate since the chemical knowledge of the study participants in general is not sufficient enough for a valid recall of agent-specific, time-specific worksite exposures.²⁵ In our study the occupational history of the study subjects was obtained through questionnaires, which did not allow an estimation of the actual exposure

concentrations that were experienced in the past. The highest achievable was a retrospective exposure assessment in terms of probability of exposure.²¹ In general this can be obtained through a JEM or a case-by-case expert assessment. Both techniques have been used in studies on occupational risk factors for bladder cancer.³⁰ In this study a case-by-case expert assessment was used.²¹ The main advantage of this method – compared with the use of a JEM – is that all the available information (job title, type and name of the company, products, and time period) is used for the exposure assessment^{36,37} while the agent-specific and time specific information derived from a JEM is limited.^{30,36} Therefore, a JEM may produce more non-differential misclassification than exposure assessments by experts.^{36,37} However, with a case-by-case expert assessment in general there are no explicit definitions of the criteria, which are used for the determination of exposure,³⁷ and the exposure assessment is affected by the learning phenomenon of the expert(s). Therefore, a case-by-case expert assessment may be less reproducible than a JEM, which will produce the same exposure information for identical basic data. To improve reliability, we used a two-stage exposure assessment in which two experts independently assessed the exposure. Disagreements were solved through consensus meetings.

The database available for the analysis provided us with the opportunity to test the possible effect of other risk factors, in particular cigarette smoking, on the observed associations. We modeled cigarette smoking habits such that they best explained bladder cancer.²⁶ This resulted in a model including number of years smoked and habitual number of cigarettes smoked per day, both as continuous variables. We therefore believe that the associations observed were not entirely due to residual confounding by smoking, although we cannot exclude some effect. We were not able to explain our results on the basis of confounding of other factors in addition to cigarette smoking, since the results were essentially unchanged after incorporating into the analyses known or suspected risk factors for bladder cancer, including total consumption of fluid, alcohol, coffee, tea, vegetables and fruit.

After stratifying the study population into non-smokers, former smokers and current smokers a pattern emerged indicating that the effects of the occupational exposures were confined to the current smokers. The RR of CPE to paint components for bladder cancer in former smokers was 0.65, in contrast to a RR of 2.22 for current smokers, reaching statistical significance. A further breakdown of these groups into light smokers and heavy smokers revealed a pattern consistent with a stronger association in current smokers who smoked more than 15 cigarettes per day compared to those who smoked less frequently. These results show that current cigarette smoking might play a role in the bladder carcinogenesis due to occupational exposure and that the carcinogenic effect decreases after cessation of smoking. This effect was particularly clear for paint components exposure and to a lesser extent for PAH exposure and form a consideration to advise workers exposed to paint components, even only in the past, to refrain from smoking cigarettes. However it must be pointed out that despite the substantial size of this cohort study the number of cases limited to the possibilities for further breakdowns and stratifications that are involved in subgroup analyses.

Several other researchers have reported on possible interactions between smoking and the association between occupational exposures and bladder cancer risk. Schumacher et al³⁸ reported an interaction between exposure to aromatic amines and bladder cancer with smoking. The odds ratio for never smokers exposed to aromatic amines for 10 or more years was 1.65, for non-exposed current smokers 3.57 and for current smokers exposed to aromatic amines 6.74. From further modeling the researchers concluded that the study gave support for some deviation from the additive model. Several other studies investigated the risks for bladder cancer from occupational exposures and cigarette smoking, but no analyses were done to test interaction between smoking and occupational exposures.^{39,40}

In conclusion, our study provides only marginal evidence for an association between occupational exposure to paint components, PAH, aromatic amines and bladder cancer risk. It further shows, despite the small proportion of exposed subjects in the study, that there might be a potentially important interactive effect from cigarette smoking,

specifically for paint components, suggesting that the carcinogenic effect might decrease after smoking cessation.

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REFERENCES

1. Parkin DM, Vizcaino AP, Skinner ME, Ndhlovu A. Cancer patterns and risk factors in the African population of southwestern Zimbabwe, 1963-1977. *Cancer Epidemiol Biomarkers Prev.* 1994;3:537-47.
2. Lilienfeld AM, Lilienfeld DE. *Foundations of epidemiology.* New York Oxford: Oxford university press, 1980.
3. Schottenfeld D, Fraumeni JF. *Cancer epidemiology and prevention.* New York Oxford: Oxford university press, 1996.
4. Rehn L. Blasengeschwuelste bei Fuchsin-arbeitern. *Arch Klin Chir.* 1895;50:588-600.
5. Case RAM, Hosker ME. Tumour of the urinary bladder as an occupational disease in the rubber industry in England and Wales. *Br J Prev Soc Med.* 1954;8:39-50.
6. Matanoski GM, Stockwell HG, Diamond EL, Haring-Sweeney M, et al. A cohort mortality study of painters and allied tradesmen. *Scand J Work Environ Health.* 1986;12:16-21.
7. Bethwaite PB, Pearce N, Fraser J. Cancer risks in painters: study based on the New Zealand Cancer Registry. *Br J Ind Med.* 1990;47:742-6.
8. Siemiatycki J, Dewar R, Nadon L, Gerin M. Occupational risk factors for bladder cancer: results from a case-control study in Montreal, Quebec, Canada. *Am J Epidemiol.* 1994;140:1061-80.
9. Terstegge CW, Swaen GM, Slangen JJ, Van Vliet C. Mortality Patterns among Commercial Painters in The Netherlands. *Int J Occup Environ Health.* 1995;1:303-10.
10. Englund A. Cancer incidence among painters and some allied trades. *J Toxicol Environ Health.* 1980;6:1267-73.
11. Morrison AS, Ahlbom A, Verhoek WG, Aoki K, et al. Occupation and bladder cancer in Boston, USA, Manchester, UK, and Nagoya, Japan. *J Epidemiol Community Health.* 1985;39:294-300.
12. Jensen OM, Wahrendorf J, Knudsen JB, Sorensen BL. The Copenhagen case-referent study on bladder cancer. Risks among drivers, painters and certain other occupations. *Scand J Work Environ Health.* 1987;13:129-34.
13. Silverman DT, Levin LI, Hoover RN, Hartge P. Occupational risks of bladder cancer in the United States: I. White men. *J Natl Cancer Inst.* 1989;81:1472-80.
14. Baxter PJ, McDowall ME. Occupation and cancer in London: an investigation into nasal and bladder cancer using the Cancer Atlas. *Br J Ind Med.* 1986;43:44-9.
15. Claude JC, Frentzel Beyme RR, Kunze E. Occupation and risk of cancer of the lower urinary tract among men. A case-control study. *Int J Cancer.* 1988;41:371-9.
16. Steenland K, Burnett C, Osorio AM. A case-control study of bladder cancer using city directories as a source of occupational data. *Am J Epidemiol.* 1987;126:247-57.
17. van den Brandt PA, Goldbohm RA, van het Veer P, A. V, et al. A large-scale prospective cohort study on diet and cancer in the Netherlands. *J Clin Epidemiol.* 1990;43:285-95.
18. Prentice RL. A case-cohort design for epidemiologic cohort studies and disease prevention trials. *Biometrika.* 1986;73:1-11.
19. van den Brandt PA, Schouten LJ, Goldbohm RA, Dorant E, et al. Development of a record linkage protocol for use in the Dutch cancer registry for epidemiological research. *Int J Epidemiol.* 1990;19:553-8.
20. Goldbohm RA, van den Brandt PA, Dorant E. Estimation of the coverage of Dutch municipalities by cancer registries and PALGA based on hospital discharge data. *Tijdschr Soc Gezond.* 1994;72:80-4.
21. van Loon AJ, Kant IJ, Swaen GM, Goldbohm RA, et al. Occupational exposure to carcinogens and risk of lung cancer: results from The Netherlands cohort study. *Occup Environ Med.* 1997;54:817-24.
22. Volovics A, van den Brandt PA. Methods for the analyses of case-cohort studies. *Biometrical Journal.* 1997;2:195-214.
23. StataCorp. *Stata Statistical Software: Release 6.0.* College Station, TX: Stata Corporation, 1999.
24. Barlow WE. Robust variance estimation for the case-cohort design. *Biometrics.* 1994;50:1064-72.
25. Lin DY, Ying Z. Cox regression with incomplete covariate measurements. *JASA.* 1993;88:1341-9.
26. Zeegers MP, Tan FE, Dorant E, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer.* 2000;89:630-9.

27. Zeegers MPA, Tan FES, Goldbohm RA, van den Brandt PA. Are coffee and tea consumption associated with urinary tract cancer risk: a systematic review and meta-analysis. *Int J Epidemiol* 2001; in press.
28. Zeegers MP, Tan FE, Verhagen AP, Weijnenberg MP, et al. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.
29. Steinmaus CM, Nunez S, Smith AH. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol*. 2000;151:693-702.
30. Pesch B, Haerting J, Ranft U, Klimpel A, et al. Occupational risk factors for urothelial carcinoma: agent-specific results from a case-control study in Germany. MURC Study Group. Multicenter Urothelial and Renal Cancer. *Int J Epidemiol*. 2000;29:238-47.
31. Bonassi S, Merlo F, Pearce N, Puntoni R. Bladder cancer and occupational exposure to polycyclic aromatic hydrocarbons. *Int J Cancer*. 1989;44:648-51.
32. Cordier S, Clavel J, Limasset JC, Boccon Gibod L, et al. Occupational risks of bladder cancer in France: a multicentre case-control study. *Int J Epidemiol*. 1993;22:403-11.
33. Clavel J, Mandereau L, Limasset JC, Hemon D, et al. Occupational exposure to polycyclic aromatic hydrocarbons and the risk of bladder cancer: a French case-control study. *Int J Epidemiol*. 1994;23:1145-53.
34. Hours M, Danache B, Fevotte J, Bergeret A, et al. Bladder cancer and occupational exposures. *Scan J Work Environ Health*. 1994;20:322-30.
35. Dosemeci M, Cocco P, Gomez M, Stewart PA, et al. Effects of three features of a job-exposure matrix on risk estimates. *Epidemiology*. 1994;5:124-7.
36. Bouyer J, Hemon D. Retrospective evaluation of occupational exposures in population-based case-control studies: general overview with special attention to job exposure matrices. *Int J Epidemiol*. 1993;22:S57-64.
37. Bouyer J, Hemon D. Studying the performance of a job exposure matrix. *Int J Epidemiol*. 1993;22:S65-71.
38. Schumacher MC, Slattery ML, West DW. Occupation and bladder cancer in Utah. *Am J Ind Med*. 1989;16:89-102.
39. Burns PB, Swanson GM. Risk of urinary bladder cancer among blacks and whites: the role of cigarette use and occupation. *Cancer Causes Control*. 1991;2:371-9.
40. Porru S, Aulenti V, Donato F, Boffetta P, et al. Bladder cancer and occupation: a case-control study in northern Italy. *Occup Environ Med*. 1996;53:6-10.

13 **Epilogue**

Since the world population is increasing and given the ageing population in the world, the incidence of bladder cancer will probably increase in the next decades. Therefore prevention of bladder cancer, just as other forms of cancer, should continue to be of public interest. Avoidance of hazardous behaviour (e.g., by means of health education and promotion) is possibly one of the most effective strategies in bladder cancer prevention. In this dissertation we evaluated several behavioural factors that are potentially associated with bladder cancer risk. We investigated the effect of the following risk factors: cigarette, cigar, pipe, and environmental smoking (i.e., smoking); total fluid, alcohol, coffee, and tea consumption (i.e., beverage consumption); fruit, vegetable, vitamin, and selenium intake (i.e., diet); and occupational exposure to polycyclic aromatic hydrocarbons, paint components, aromatic amines, and diesel exhaust (i.e., occupation).

In this chapter, a rating system was used to summarize the strength of scientific evidence for these risk factors consisting of four levels: convincing, probable, possible, and no evidence (Table 1). The classification of levels of evidence is based on analyses from the Netherlands Cohort Study; previous epidemiologic studies (including ecological studies, case control studies, cohort studies, and controlled trials); and previous mechanistic evidence (including biological plausibility and animal experiments) (Table 1).

Table 1. Levels of evidence for etiological risk factors for disease

Number of epidemiologic studies showing consistent associations (N _{study})	Supportive mechanistic evidence*	No supportive mechanistic evidence*
N _{study} ≥ 15	Convincing	Probable
5 ≤ N _{study} < 15	Probable	Possible
1 ≤ N _{study} < 5	Possible	Possible
N _{study} = 0	No evidence	No evidence

*biologically plausible and supportive laboratory evidence
*biologically not plausible, or no supportive laboratory evidence

The strength of association between a potential risk factor and bladder cancer risk is described as substantially increased risk, moderately increased risk, slightly (or small) increased risk, no association, slightly (or small) decreased risk, moderately decreased risk, and substantially decreased risk. (Table 2) We constructed a matrix in which the levels of evidence were combined with the levels of association (Table 3).

Table 2. Levels of association for etiological risk factors for disease

Association	Relative risk for exposed vs. not exposed
Substantially increased risk	≥2.5
Moderately increased risk	1.5≤RR<2.5
Slightly (or small) increased risk	1.2≤RR<1.5
No association	0.8≤RR<1.2
Slightly (or small) decreased risk	0.7≤RR<0.8
Moderately decreased risk	0.4≤RR<0.7
Substantially decreased risk	RR<0.4

Etiologic fraction. For each risk factor with probable or convincing evidence for a moderately or substantially altered bladder cancer risk the etiologic fraction was calculated based on the level of association and the prevalence of exposure in the subcohort of the Netherlands Cohort Study. The subcohort is a representative sample (n=3500) of the Dutch adult population. The etiologic fraction is the proportion of the bladder cancer cases for whom a specific exposure contributed to the disease etiology.¹

Previous epidemiologic studies. During the last four decades, 43 epidemiologic studies² and 10 reviews³⁻¹² have consistently shown that cigarette smoking substantially increases the risk of bladder cancer, although until recently the association has never been quantified systematically in reviews. In the meta-analysis as presented in chapter 3, we estimated that current cigarette smokers have an approximately threefold higher risk of bladder cancer than non-smokers.² A positive dose-response relationship has been found with both the number of cigarettes smoked per day and the number of years smoked. Age at first exposure and cessation of cigarette smoking were inversely associated with bladder cancer risk.²

Few studies, however, investigated the association between other forms of tobacco smoking (i.e., cigar pipe and environmental smoking and bladder cancer risk. The evidence of an association between bladder cancer risk and cigar¹³⁻¹⁹ and pipe¹³⁻²⁰ smoking is not often reported ($n=7$ and 8 , respectively), inconsistent, and generally weaker than the association with cigarette smoking ($1.0 \leq RR < 2.5$ for cigar smokers and $1.0 \leq RR < 8.6$ for pipe smokers, comparing exposed with not exposed).^{4,6,10,12}

Only two case control studies investigated the association between environmental smoking and bladder cancer risk.^{14,21} These studies have reported small increased risks for those exposed to environmental smoking compared with those not exposed to environmental smoking ($0.6 \leq RR < 1.6$).

Netherlands Cohort Study. In the Netherlands Cohort study these detailed associations were further investigated (Chapter 6). In this study we concluded that the above-mentioned associations of cigarette smoking characteristics with bladder cancer risk were largely attributable to cigarette smoking duration and that these associations were independent of different tumour invasiveness or morphology. Furthermore, the tar and nicotine content of cigarettes, and filter-tip usage were only weakly associated with bladder cancer risk. Cigarette smoking cessation and age at first exposure were not associated with bladder cancer risk after taking into account the total smoking duration (Chapter 6). These results were comparable with the overall results from the meta-analysis (Chapter 3).² Therefore, inclusion of this study in the meta-analysis did not essentially change the estimates for the association between cigarette smoking and bladder cancer risk. The summary odds ratio for current cigarette smoking changed from 3.33 (confidence interval (CI) 2.63-4.21) to 3.31 (CI 2.67-4.10).

In this study cigar and pipe smoking and exposure to environmental smoking were not associated with bladder cancer risk (Chapter 6).

Mechanistic evidence. The precise mechanism by which cigarette smoking causes bladder cancer has yet to be determined. It seems most likely that the risk of bladder cancer is related to some of the large number of chemicals present in smoke. 2-Naphthylamine and 4-aminobiphenyl are the leading candidates as the specific etiologic agents.^{6,11,12,22,23} Several nitrosamines have been shown to produce bladder cancer in animal models.¹² Tars might induce bladder papillomas and carcinomas in mice.¹¹ Molecular studies have suggested that exposure to certain carcinogens in cigarette smoke may contribute to DNA damage, i.e., chromosome 9 defects²⁴ and p53 mutations.²⁵ Besides the effect that many compounds in cigarettes can cause carcinogenic events in the bladder, cigarette smoking has been found to increase proliferation, as evidenced by hyperplasia of the bladder epithelium.¹²

An association between cigar and pipe smoking and the risk of bladder cancer is biologically plausible since the smoke of both types of tobacco contains many of the same substances as present in cigarette tobacco. However, cigar and pipe smokers inhale less than cigarette smokers, which might explain an increased cancer incidence for cigar and pipe smokers in local areas of the human body (e.g., head/neck cancer) and no increased risk through a systemic effect (e.g., bladder cancer).

Data from experimental studies also support the hypothesis that non-smokers might be exposed to potential carcinogens through the cigarette smoke of others.²⁶ Mutagens have been detected in the blood and urine of passive smokers.^{26,27}

Conclusions. Judging the evidence, we conclude that there is convincing evidence from many epidemiologic and mechanistic studies that cigarette smoking substantially increases the risk of bladder cancer (Table 3).

Although an association with cigar and pipe smoking could be mechanistically explained, there is also biologically plausible evidence for no association with bladder cancer risk. The results from many epidemiologic studies concerning cigar and pipe smoking are not consistent. We therefore conclude that a small increased risk for cigar and pipe smoking is only possible.

An increased risk through exposure to environmental smoking is supported by mechanistic evidence, although more epidemiologic studies are needed to confirm this. A small association between exposure to environmental smoking and bladder cancer risk possibly exists (Table 3).

Etiologic Fraction. Since there is convincing evidence for a substantially increased risk of bladder cancer for cigarette smokers, the etiologic fraction of cigarette smoking was calculated based on the results of the Netherlands Cohort Study. Cigarette smoking accounts for at least 25% of all female bladder cancer, whereas in men more than 50 percent of the disease may be associated with cigarette smoking (Chapter 6).

TOTAL FLUID, ALCOHOL, COFFEE, AND TEA CONSUMPTION

Previous epidemiologic studies. Previous case control and cohort studies on total fluid consumption and bladder cancer risk showed inconsistent results. Seven case control studies reported positive associations ($1.4 \leq RR < 4.9$), comparing highest *vs.* lowest total fluid consumption),^{17,28-32} whereas two other case-control studies found no association.³³⁻³⁵ Only one case control study reported an inverse association between total fluid consumption and bladder cancer risk in women, but not in men.³⁶ Two cohort studies to date were conducted to investigate this association.^{37,38} One cohort study did not find an association between fluid consumption and bladder cancer risk,³⁷ whereas the other reported a preventive effect of total fluid consumption ($RR=0.5$).³⁸

Previous studies also have investigated associations of specific fluids (i.e., alcohol, coffee and tea consumption) with bladder cancer risk. We conclude, based on a meta-analysis of 30 epidemiologic studies (Chapter 4) that the available data suggest an increased cigarette smoking-adjusted risk of bladder cancer from alcohol consumption for men, which, however, is small ($RR=1.4$, $CI\ 0.9-2.0$, comparing any alcohol consumption with no alcohol consumption).³⁹ The risk related to alcohol consumption for women remains unclear.³⁹

Earlier narrative reviews on coffee consumption concluded that the epidemiologic evidence was consistent with a small positive association, possibly due to confounding by smoking.^{10,40-42} In our meta-analysis the available evidence based on 14 epidemiologic studies suggested a very small increased risk for coffee consumers remaining after adjustment for age, cigarette smoking and sex ($RR=1.2$, $CI\ 1.0-1.4$), although this was predominantly based on case-control data (Chapter 5).⁴³

We found in a systematic review based on 7 epidemiologic studies that current consumption of tea seems not to be related to an increased risk of bladder cancer. The summary odds ratio for tea drinking compared with no tea drinking was $1.0(CI\ 0.9-1.1)$ (Chapter 5).⁴³

Netherlands Cohort Study. In the Netherlands Cohort Study the association between fluid intake and bladder cancer risk was examined in detail (Chapter 7 and Chapter 8). In our study, neither total fluid consumption nor total water intake were associated with bladder cancer risk (Chapter 8).⁴⁴

The positive association between alcohol consumption and male bladder cancer found in the Netherlands Cohort Study was small and independent of different tumour invasiveness and morphology (Chapter 7).⁴⁵ We found no association between alcohol consumption and bladder cancer risk for women (Chapter 7).⁴⁵ Although the Netherlands Cohort Study included more detailed information about alcohol consumption, we repeated our meta-analysis to evaluate whether the summary odds ratios for male consumers compared with non-drinkers changed after including the Netherlands Cohort Study (Chapter 4).⁴³ The summary odds ratio remained stable at 1.4, although the precision increased from (CI 0.9-2.0) to (CI 1.0-1.9).

The results of the Netherlands Cohort Study supported a small positive association between coffee consumption and male bladder cancer, mainly confined to the higher consumption categories after detailed control for cigarette smoking (Chapter 8).⁴⁴ For women, an inverse association was found (Chapter 8).⁴⁴ After incorporating the results of the Netherlands Cohort Study in the meta-analysis on coffee consumption (Chapter 5) the summary odds ratios changed from 1.2 (CI 1.0-1.4) to 1.1 (CI 1.0-1.3), comparing any coffee consumption with no coffee consumption.

Tea consumption was inversely associated with bladder cancer for both men and women in the Netherlands Cohort Study, irrespective of consumption frequency (Chapter 8).⁴⁴ Inclusion of these results in the meta-analysis on tea consumption (Chapter 5) changed the summary estimates from 1.0 (CI 0.9-1.1) to 0.9 (CI 0.8-1.1), comparing any tea consumption with no tea consumption.

Mechanistic evidence. It has been suggested that drinking more fluids is associated with a decreased risk of bladder cancer.^{38,46} The urogenous-contact hypothesis associates the development of bladder cancer with prolonged exposure to carcinogens in urine.^{38,47} High consumption of fluids may reduce this exposure by diluting the urine and reducing contact time through increased frequency of urination. However, total fluid consumption has also been hypothesized to increase the risk of bladder cancer by increasing the workload of the bladder or by exposing the bladder to specific fluids that contain carcinogens.^{32,34}

Also, several possible mechanisms to explain carcinogenesis by alcohol intake have been postulated. Heavy alcohol consumption may, through its effect on the liver and hepatic enzyme systems, reduce the detoxification of carcinogenic substances⁵² Furthermore, ethanol slows down protein synthesis. One consequence of this is that cell repair mechanisms might be inhibited, which could lead to malignant changes.^{48,49} Ethanol might improve the permeability of membranes to carcinogens and might enhance carcinogenic activity.⁴⁹⁻⁵¹ Other explanations include the effect of ethanol on cell proliferation, possibly caused by the ethanol metabolite acetaldehyde.^{12,49,50,52}

Caffeine and other substances in coffee could have direct or indirect metabolic effects and modify concentrations of carcinogenic or anticarcinogenic substances in bladder epithelium.⁴² Since most of these substances or their metabolites are excreted by the bladder, they have to come into contact with the mucous membrane of the bladder first. However, there are no clear-cut conclusions from animal studies on the potential carcinogenic effects of coffee. Most animal studies indicate that coffee and caffeine do not have a carcinogenic effect in laboratory animals, nor can they induce tumours, potentiate or inhibit the carcinogenic effects of other substances.⁴²

Evidence from animal studies shows that certain compounds present in tea may have inhibitory effects on bladder tumour formation and growth. This inhibitory activity is believed to be mainly due to the antioxidative and possibly antiproliferative effects of polyphenol compounds (e.g., flavonoids).⁵³⁻⁵⁶ It has been pointed out that no other agent tested for possible chemoprevention effects in animal models has elicited such strong activity as tea and its components at concentrations usually consumed by humans.⁵⁵

Conclusions. We conclude that it is possible that total fluid is not associated with bladder cancer risk (Table 3). Although epidemiologic and mechanistic evidence are not consistent, the weight of evidence is not in favour of a protective effect of fluid intake.

There is convincing evidence for a small positive association between alcohol consumption and bladder cancer risk in men. The risk related to alcohol consumption for women remains unclear.

Coffee consumption appears to pose no particular threat in most people. Most epidemiologic and mechanistic studies were not supportive for an increased bladder cancer risk for coffee consumers. Probably there is no association between coffee consumption and bladder cancer. If an increased risk exists, it is probably weak and mainly confined to the higher consumption categories.

Although there is supportive mechanistic evidence for a negative association between tea consumption and bladder cancer risk, most epidemiologic studies could not confirm this. We conclude that tea consumption is probably not associated with the risk of bladder cancer (Table 3).

FRUIT, VEGETABLE, VITAMIN (A, C, E AND FOLATE), AND SELENIUM INTAKE

Previous epidemiologic studies. Almost all studies of vegetable and fruit consumption and the risk of bladder cancer have reported either a decreased risk or no relationship with higher consumption for vegetable or fruit categories.³ Recently, a meta-analysis on 7 epidemiologic studies concerning bladder cancer and fruit consumption reported small elevated smoking-adjusted bladder cancer risks among subjects with low fruit consumption *vs.* subjects with high fruit consumption (RR=1.5, CI 1.3-1.7).⁵⁷

The same meta-analysis, based on 7 epidemiologic studies also found a small positive smoking-adjusted association between vegetable consumption and bladder cancer risk comparing high *vs.* low consumption (RR=1.2, CI 1.0-1.4).⁵⁷

Furthermore, the meta-analysis based on seven case control studies and three cohort studies did not find increased risks of bladder cancer for diets low in retinol or β -carotene compared to diets high in retinol and β -carotene (RR=1.0, CI 0.8-1.2).⁵⁷ A prospective study that was published more recently also found no association between supplemental and dietary vitamin A intake.⁵⁸

The majority of epidemiologic studies on dietary vitamin C intake reported relative risks around unity for high *vs.* low intake.^{33,59-63}

Most epidemiologic studies reported inverse associations between supplemental intakes of vitamin E in relation to bladder cancer risk ($0.5 \leq \text{RR} < 0.7$, comparing high with low intake).^{58,61,64} Dietary vitamin E intake was also inversely associated with bladder cancer in one study.⁵⁹

The epidemiologic evidence relating dietary folate and the risk of bladder cancer is limited. One case control study found an inverse association with bladder cancer risk (RR=0.5, comparing highest with lowest quartiles of intake),⁶⁴ whereas one cohort study did not find an association between total folate intake and bladder cancer incidence.⁵⁸

Four prospective studies investigated the association between serum or toenail selenium and bladder cancer risk.⁶⁵⁻⁶⁸ Most studies found a moderate inverse association between selenium status and bladder cancer risk ($0.3 \leq \text{RR} < 0.5$, comparing high with low exposure), although one small study found an increased risk for women only (RR=2.51).⁶⁷

Netherlands Cohort Study. In the Netherlands Cohort Study, an inverse association was found between bladder cancer risk and fruit consumption (RR=0.7, CI 0.5-1.0, comparing highest *vs.* lowest quintile of consumption). This protective effect was mainly confined to invasive transitional cell carcinomas, irrespective of tumour morphology (Chapter 9)

The results showed that total vegetable consumption was not associated with bladder cancer risk (RR=0.9, CI 0.7-1.3, comparing highest *vs.* lowest quintile of consumption) (Chapter 9).

We concluded furthermore that dietary or supplemental intake of vitamin A, vitamin C, vitamin E, and dietary intake from folate and carotenoids were not associated with bladder cancer risk. Only β -cryptoxanthin appeared to be inversely associated with

bladder cancer risk, which appeared to be independent of different tumour invasiveness and morphology (RR=0.7, CI 0.5-1.0) (Chapter 10).

The results of the Netherlands Cohort Study showed a moderate inverse association between base-line toenail selenium and subsequent bladder cancer risk (RR=0.7, CI 0.5-0.9). The association was most pronounced in ex-smokers and mainly confined to invasive carcinomas of the bladder (Chapter 11).

Mechanistic evidence. There are many candidate agents in fruit and vegetables that potentially influence bladder cancer risk. Some possible anticarcinogenic mechanisms of substances in vegetables and fruits include antioxidant effects, effects on cell differentiation, increased activity of enzymes that detoxify carcinogens, blocked formation of nitrosamines, preserved integrity of intracellular matrices, effects on DNA methylation, maintenance of normal DNA repair, increased apoptosis of cancer cells and, decreased cell proliferation.⁶⁹

Retinol is the physiologically active form of vitamin A and can be derived from some carotenoids. Carotenoids have antioxidant properties and may inhibit carcinogenesis by preventing DNA damage induced by free radicals. Furthermore, vitamin A is important for cell differentiation and since lack of differentiation is a feature of cancer cells, a protective effect of vitamin A on cancer development may be expected.⁷¹⁻⁷³

Vitamin C is mostly known for its antioxidant capacity⁷¹ and its ability to prevent formation of nitrosamine and other *N*-nitroso compounds.⁷⁴

Like vitamin C, vitamin E is an intracellular antioxidant and can inhibit nitrosation.^{72,3,75} Furthermore, vitamin E has the capacity to keep selenium in reduced state.⁷¹

Folate is essential for methylation reactions in the human body. Reduced methylation of DNA may contribute to loss of normal controls on proto-oncogene expression. Folate deficiency causes massive incorporation of uracil into human DNA and chromosome breaks. Such breaks could contribute to an increased risk of cancer.^{76,77}

Several hypotheses have been proposed to explain the inhibition of tumourgenesis by selenium, including alterations in carcinogen metabolism, effects on the endocrine and immune systems, production of cytotoxic selenium metabolites, inhibition of protein synthesis, inhibition of specific enzymes, inhibition of tumour growth, stimulation of apoptosis and protection against oxidative stress involving the function of selenium as a constituent in the antioxidant enzyme glutathione peroxidase.^{66,68,72,78-82}

Conclusions. We conclude that high consumption of total fruit is probably associated with a small decreased bladder cancer risk. (Table 3)

There is probably no association between total vegetable intake and bladder cancer risk, although an influence of specific vegetables on bladder cancer risk cannot be excluded.

Although the mechanistic evidence is in favour of an inverse association between vitamin A, carotenoids, vitamin C intake, and subsequent bladder cancer risk, the epidemiologic evidence does not support this. We conclude tentatively that vitamin A, carotenoids, and vitamin C intake are probably not associated with bladder cancer risk.

Few epidemiologic studies and mechanistic evidence support a possibly moderate inverse association between vitamin E intake and bladder cancer risk.

The epidemiologic evidence for the intake of folate is still limited and conflicting. From mechanistic point of view a protecting effect from folate intake is however plausible. Until more epidemiologic studies are conducted, we conclude that there is possible evidence that folate is not associated with bladder cancer risk.

There is probably a moderate inverse association between selenium intake and bladder cancer risk (Table 3).

Etiologic Fraction. Since there is probable evidence for a moderately decreased risk of bladder cancer for subjects with high selenium status, the etiologic fraction was calculated based on the results of the Netherlands Cohort Study. If toenail selenium status of subjects in the two lowest quintiles of the Dutch population (≤ 0.530 $\mu\text{g/g}$) could be increased to $0.530\text{--}0.573$ $\mu\text{g/g}$ (e.g., through supplementation), approximately 20% of bladder cancer incidence could be prevented.

Previous epidemiologic studies. Occupational exposure to carcinogenic chemicals is an established risk factor of bladder cancer. Many previous epidemiologic studies have investigated the association between several occupations and bladder cancer risk.⁸³ Among these chemicals were exposure to polycyclic aromatic hydrocarbons (PAH) in the aluminium industry, coal tar industry and coal gasification industry.⁸⁴ In addition, a substantial number of studies have reported an excess of bladder cancer among painters,⁸⁵⁻⁸⁸ although others could not confirm this association.^{89,90} In a recent meta-analysis of 35 epidemiologic studies an increased occurrence of bladder cancer was found for truck drivers (RR=1.2, CI 1.1-1.3) and bus drivers (RR=1.3, CI 1.2-1.5).⁹¹ In this case the most likely causal agent is thought to be constituents of the diesel exhaust emissions. Ten studies, however, which considered diesel exhaust exposure specifically based on a job exposure matrix or a similar approach, did not show an association with bladder cancer risk (RR=1.1, CI 1.0-1.3).⁹¹ Although many epidemiologic studies were conducted to evaluate the associations between various occupations and bladder cancer risk, relatively few studies investigated the association with potential carcinogenic agents (except for aromatic amines).

Netherlands Cohort Study. In the Netherlands Cohort study we examined the associations between occupational exposures to PAH, paint components, aromatic amines, diesel exhaust and the incidence of male bladder cancer (Chapter 12). The results suggested a small positive association between cumulative probability of exposure to PAH and bladder cancer risk (RR=1.2, CI 0.7-2.3, comparing highest tertile of exposure with no exposure), which was most pronounced in non-papillary invasive transitional cell carcinomas of the bladder. Paint components and aromatic amines were also weakly associated with bladder cancer incidence, although the RRs were small (RR=1.3, CI 0.7-2.3 and 1.3, CI 0.4-4.2, respectively, comparing the same exposure categories). Limited support was found for a small positive association between diesel exhaust and bladder cancer risk (RR=1.2, CI 0.8-1.9). The results indicate this association is most pronounced to non-papillary, non-invasive bladder cancer (RR=1.7, CI 0.5-5.8) (Chapter 12).

Mechanistic evidence. β -Naphthylamine and benzidine, which are polycyclic aromatic amines are considered to be carcinogenic in animals.^{92,93} Also, many other PAHs show carcinogenic effects in animal experiments.⁹³ As intermediate metabolites, epoxides are formed, which are supposed to cause the carcinogenic activity as they react with macromolecules such as DNA. Especially the reaction with DNA is thought to affect normal cell proliferation.⁹³ Paint dust, which emerge at paint removal, contains pigments and other carcinogenic substances. PAHs and nitro-PAHs are also suggested to be of importance with respect to the carcinogenic properties of diesel exhaust.

Conclusions. High occupational exposure to aromatic amines is convincingly associated with a moderately increased risk of bladder cancer (Table 3). However, the majority of these aromatic amine carcinogens are now either no longer used industrially or exposure to workers has been greatly reduced as a result of improved technology. Therefore, no etiologic fraction is calculated for this risk factor.

A small positive association between bladder cancer risk and exposure to paint components, polycyclic aromatic hydrocarbons, and diesel exhaust is probable since these associations have been found in previous epidemiologic studies (on both occupations and occupational exposures), and mechanistic evidence support these associations. (Table 3).

Epidemiology has traditionally evaluated environmental risk factors for disease, including bladder cancer, which has provided an important contribution to our knowledge on bladder cancer aetiology. However, knowledge of environmental risk factors and possible pathways alone is probably not sufficient to explain completely the incidence of bladder cancer, and many other multifactorial diseases. Epidemiologists could extend their study designs to studies designed to evaluate the combined effect of environmental risk factors and genetic factors. This combination might explain more variation in bladder cancer incidence than using environmental risk factors alone.

Polymorphism-environment. Genetic polymorphisms could be seen as potential genetic risk factors for bladder cancer or as effect modifiers in the association between environmental risk factors and bladder cancer risk. For example, the NAT2 gene is involved in phase II detoxification of bladder carcinogens. Certain polymorphisms in the NAT2 gene result in a slow acetylation phenotype, which is thought to increase bladder cancer risk. A recently conducted meta-analysis showed that slow acetylators had an approximately 40% increase in risk compared with rapid acetylators. These results suggest that in most populations studied to date, NAT2 slow acetylation status is a risk factor for bladder cancer.⁹⁴ Future epidemiologic research could be aimed to identify genetic risk factors for bladder cancer.

Furthermore, genetic polymorphisms may interact with environmental risk factors in bladder carcinogenesis. For example, NAT2 or glutathione S-transferase M1 (GSTM1) polymorphisms, may modify metabolism of tobacco carcinogens. Slow acetylation or lack of GSTM1 activity (which is present in 50% of Caucasians) might result in a higher concentration of tobacco carcinogens in the bladder and consequently enhance the risk of bladder cancer among cigarette smokers.⁹⁵⁻⁹⁸ Through further epidemiologic research on polymorphisms-environment interaction it might become possible to identify individuals with a higher susceptibility of bladder cancer after exposure to potential carcinogens compared with the general population.

Mutation-environment. In parallel with these new genetic epidemiologic approaches to understanding bladder cancer aetiology, there have been important advances in understanding molecular genetics of bladder cancer, i.e. in identifying the series of genetic changes in bladder cells that in combination lead to the development of the malignant phenotype.¹¹ Genetic differences seem to exist between papillary and non-papillary carcinomas. The high-grade, non-papillary transitional cell carcinoma usually contains a defect in chromosome 17, which represents an abnormality in the p53 gene. By contrast, changes in chromosome 9 are associated with superficial papillary tumors.¹² We have found different associations with some of the investigated risk factors depending on tumour invasiveness and morphology. A positive association between occupational exposure to polycyclic hydrocarbons and subsequent bladder cancer risk was most pronounced in non-papillary invasive transitional cell carcinomas (Chapter 12). On the contrary, the positive association between exposure to diesel exhaust and bladder cancer risk was most pronounced among men with non-papillary non-invasive tumours (Chapter 12). Furthermore, it appeared that a protective effect of toenail selenium level and fruit consumption was mainly confined to invasive transitional cell carcinomas, irrespective of tumour morphology (Chapter 9 and 11). However, associations for cigarette smoking, alcohol intake, and exposure to paint components were independent of different tumour invasiveness and morphology (Chapter 7, 10, and 12). It might be interesting for epidemiologists to investigate associations between specific environmental risk factors and specific mutations that lead to different morphology and invasiveness of bladder cancer.

CONCLUSION

Based on the results of the Netherlands Cohort Study, previous epidemiologic studies, and mechanistic evidence we conclude that the most important risk factor for bladder cancer is cigarette smoking. Prevention of cigarette smoking would result in a decrease of more than 50% of male bladder cancer cases and 25% of female bladder cancer cases. Occupational exposure to aromatic amines is an established risk factor for bladder cancer as well. However, industrial exposure to aromatic amines is greatly reduced nowadays. Alcohol consumption is the only other convincing risk factor for bladder cancer, although previous research suggests that the risk elevation for alcohol consumers is small, which might not be of practical importance. We found probable evidence for a small increased risk for other forms of occupational exposure (i.e., PAH, diesel exhausts, and paint components). There is probably no association between coffee, tea, vitamin A and C intake and subsequent bladder cancer risk and a negative association with fruit consumption and selenium intake. It is possible that cigar, pipe and passive smoking increases bladder cancer risk slightly; that total fluid consumption and folate intake are not associated with bladder cancer risk; and that vitamin E intake decreases bladder cancer risk. Further research is needed to confirm these conclusions.

Table 3. Levels of evidence and levels of association for risk factors (exposed *vs.* not exposed) for urinary bladder cancer

Evidence	Convincing ¹	Probable ²	Possible ³
Substantially Increased risk ^a	Cigarette smoking		
Moderately Increased risk ^b	Aromatic amines exposure		
Small increased risk ^c	Alcohol consumption*	PAH exposure Paint components exposure Diesel exhaust exposure	Cigar smoking Pipe smoking Environmental smoking
No association ^d		Coffee consumption Tea consumption Vegetable consumption Vitamin A intake ^e Vitamin C intake	Total fluid consumption Folate intake
Small Decreased risk ^e		Fruit consumption	
Moderately Decreased risk ^f		Selenium intake	Vitamin E intake
Substantially Decreased risk ^g			

¹Convincing: ≥ 15 well designed epidemiologic studies showing consistent associations (N_{study}) and supportive mechanistic evidence, ²Probable: $5 \leq N_{study} < 15$ and supportive mechanistic evidence, or $N_{study} \geq 15$ and no supportive mechanistic evidence, ³Possible: $5 \leq N_{study} < 15$ and supportive mechanistic evidence, or $1 \leq N_{study} < 5$
^a($RR \geq 2.5$), ^b($1.5 \leq RR < 2.5$), ^c($1.2 \leq RR < 1.5$), ^d($0.8 \leq RR < 1.2$), ^e($0.7 \leq RR < 0.8$), ^f($0.4 \leq RR < 0.7$), ^g($RR < 0.4$), comparing exposure *vs.* no exposure.
*for men; the risk related to alcohol consumption for women remains unclear
^eand carotenoids

REFERENCES

1. Rothman K, Greenland S. *Modern Epidemiology*. 2 ed. Philadelphia: Lippincott-Rave Publishers, 1998.
2. Zeegers MP, Tan FE, Dorant E, van den Brandt PA. The impact of characteristics of cigarette smoking on urinary tract cancer risk: a meta-analysis of epidemiologic studies. *Cancer*. 2000;89:630-9.
3. World Cancer Research Fund, American Institute for Cancer Research. *Food, Nutrition and the Prevention of Cancer: a global perspective*. Menasha: Banta Book Group, 1997.
4. Matanoski GM, Elliott EA. Bladder cancer epidemiology. *Epidemiol Rev*. 1981;3:203-29.
5. Morrison AS. Advances in the etiology of urothelial cancer. *Urol Clin North Am*. 1984;11:557-66.
6. Dolin PJ. An epidemiological review of tobacco use and bladder cancer. *J Smoking Related Dis*. 1991;2:129-43.
7. Silverman DT, Hartge P, Morrison AS, Devesa SS. Epidemiology of bladder cancer. *Hematol Oncol Clin North Am*. 1992;6:1-30.
8. Shirai T. Etiology of bladder cancer. *Semin Urol*. 1993;11:113-26.
9. Shirai T, Fradet Y, Huland H, Bollack C, Droller M, Janknegt R, Jones P, Kagawa S. The etiology of bladder cancer: are there any new clues or predictors of behavior? *Int J Urol*. 1995;2:64-75.
10. Silverman DT, Morrison AS, Devesa SS. Bladder cancer. In: Schottenfeld D, Fraumeni JF, editors. *Cancer epidemiology and prevention*. New York and Oxford: Oxford University Press, 1996:1156-79.
11. Ross RK, Jones PA, Yu MC. Bladder cancer epidemiology and pathogenesis. *Semin Oncol*. 1996;23:536-45.
12. Johansson SL, Cohen SM. Epidemiology and etiology of bladder cancer. *Semin Surg Oncol*. 1997;13:291-8.
13. Howe GR, Burch JD, Miller AB, Cook GM, Esteve J, Morrison B, Gordon P, Chambers LW, Fodor G, Winsor GM. Tobacco use, occupation, coffee, various nutrients, and bladder cancer. *J Natl Cancer Inst*. 1980;64:701-13.
14. Burch JD, Rohan TE, Howe GR, Risch HA, Hill GB, Steele R, Miller AB. Risk of bladder cancer by source and type of tobacco exposure: a case-control study. *Int J Cancer*. 1989;44:622-8.
15. Slattery ML, Schumacher MC, West DW, Robison LM. Smoking and bladder cancer. The modifying effect of cigarettes on other factors. *Cancer*. 1988;61:402-8.
16. Jensen OM, Wahrendorf J, Blettner M, Knudsen JB, Sorensen BL. The Copenhagen case-control study of bladder cancer: role of smoking in invasive and non-invasive bladder tumours. *J Epidemiol Community Health*. 1987;41:30-6.
17. Claude J, Kunze E, Frentzel Beyme R, Paczkowski K, Schneider J, Schubert H. Life-style and occupational risk factors in cancer of the lower urinary tract. *Am J Epidemiol*. 1986;124:578-89.
18. Hartge P, Hoover R, Kantor A. Bladder cancer risk and pipes, cigars and smokeless tobacco. *Cancer*. 1985;15:901-6.
19. Steineck G, Norell SE, Feychting M. Diet, tobacco and urothelial cancer: a 14-year follow-up of 16,477 subjects. *Acta Oncol*. 1988;27:323-7.
20. Morrison AS, Buring JE, Verhoek WG, Aoki K, Leck I, Ohno Y, Obata K. An international study of smoking and bladder cancer. *J Urol*. 1984;131:650-4.
21. Sandler DP, Everson RB, Wilcox AJ. Passive smoking in adulthood and cancer risk. *Am J Epidemiol*. 1985;121:37-48.
22. Morrison AS, Cole P. Epidemiology of bladder cancer. *Urol Clin North Am*. 1976;3:13-29.
23. Vincis P. Epidemiological models of carcinogenesis: the example of bladder cancer. *Cancer Epidemiol Biomarkers Prev*. 1992;1:149-53.
24. Zhang ZF, Shu XM, Cordon Cardo C, Orlov I, Lu ML, Millon TV, Cao PQ, Connolly Jenks C, Dalbagni G, Lianes P, Lacombe L, Reuter VE, Scher H. Cigarette smoking and chromosome 9 alterations in bladder cancer. *Cancer Epidemiol Biomarkers Prev*. 1997;6:321-6.
25. Spruck CHD, Rideout WMD, Olumi AF, Ohnseit PF, Yang AS, Tsai YC, Nichols PW, Horn T, Hermann GG, Steven K, et al. Distinct pattern of p53 mutations in bladder cancer: relationship to tobacco usage. *Cancer Res*. 1993;53:1162-6.
26. Sandler DP, Wilcox AJ, Everson RB. Cumulative effects of lifetime passive smoking on cancer risk. *Lancet*. 1985;1:312-5.
27. Bos RP, Theuvs JL, Henderson PT. Excretion of mutagens in human urine after passive smoking. *Cancer Lett*. 1983;19:85-90.
28. Jensen OM, Wahrendorf J, Knudsen JB, Sorensen BL. The Copenhagen case-control study of bladder cancer. II. Effect of coffee and other beverages. *Int J Cancer*. 1986;37:651-7.
29. Kunze E, Chang Claude J, Frentzel Beyme R. Life style and occupational risk factors for bladder cancer in Germany. A case-control study. *Cancer*. 1992;69:1776-90.
30. Vena JE, Graham S, Freudenheim J, Marshall J, Zielezny M, Swanson M, Sufrin G. Drinking water, fluid intake and bladder cancer in western new york. *Arch Environ Health*. 1993;48:191-8.
31. Cantor KP, Hoover R, Hartge P, Mason TJ, Silverman DT, Altman R, Austin DF, Child MA, Key CR, Marrett LD, et al. Bladder cancer, drinking water source, and tap water consumption: a case-control study. *J Natl Cancer Inst*. 1987;79:1269-79.
32. Bianchi GD, Cerhan JR, Parker AS, Putnam SD, See WA, Lynch CF, Cantor KP. Tea consumption and risk of bladder and kidney cancers in a population- based case-control study. *Am J Epidemiol*. 2000;151:377-83.
33. Risch HA, Burch JD, Miller AB, Hill GB, Steele R, Howe GR. Dietary factors and the incidence of cancer of the urinary bladder. *Am J Epidemiol*. 1988;127:1179-91.

34. Slattery ML, West DW, Robison LM. Fluid intake and bladder cancer in Utah. *Int J Cancer*. 1988;42:17-22.
35. Dunham LJ, Rabson AS, Steward HS, Frank A, Young JL. Rates, interview, and pathology study of cancer of the urinary bladder in New Orleans, Louisiana. *J Natl Cancer Inst*. 1968;41:683-709.
36. Wilkens LR, Kadir MM, Kolonel LN, Nomura AM, Hankin JH. Risk factors for lower urinary tract cancer: the role of total fluid consumption, nitrites and nitrosamines, and selected foods. *Cancer Epidemiol Biomarkers Prev*. 1996;5:161-6.
37. Mills PK, Beeson WL, Phillips RL, Fraser GE. Bladder cancer in a low risk population: results from the Adventist Health Study. *Am J Epidemiol*. 1991;133:230-9.
38. Michaud DS, Spiegelman D, Clinton SK, Rimm EB, Curhan GC, Willett WC, Giovannucci L. Fluid intake and the risk of bladder cancer in men. *N Engl J Med*. 1999;340:1390-7.
39. Zeegers MP, Tan FE, Verhagen AP, Weijnenberg MP, van den Brandt PA. Elevated risk of cancer of the urinary tract for alcohol drinkers: a meta-analysis. *Cancer Causes Control*. 1999;10:445-51.
40. World Health Organization. IARC monographs on the evaluation of carcinogenic risks to humans: coffee, tea, mate, methylxanthines and methylglyoxal. Lyon: World Health Organization, 1991.
41. World Cancer Research Fund, American Institute for Cancer Research. Bladder. Food, Nutrition and the Prevention of Cancer: a global perspective. Menasha: Banta Book Group, 1997:338-61.
42. Nehlig A, Debry G. Coffee and cancer: a review of human and animal data. *World Rev Nutr Diet*. 1996;79:185-221.
43. Zeegers MPA, Tan FES, Goldbohm RA, van den Brandt PA. Are coffee and tea consumption associated with urinary tract cancer risk: a systematic review and meta-analysis. *Int J Epidemiol*. 2001:in press.
44. Zeegers M, Dorant E, Goldbohm R, van den Brandt P. Are coffee, tea and total fluid consumption associated with bladder cancer risk? Results from the Netherlands Cohort Study. *Cancer Causes and Control*. 2001:in press.
45. Zeegers MPA, Volovics A, Dorant E, Goldbohm RA, van den Brandt PA. Alcohol and cancer of the urinary tract: results from the Netherlands cohort study. *Am J Epidemiol*. 1998;153:38-41.
46. Charatan F. Fluid intake affects the risk of bladder cancer in men. *Br Med J*. 1999;318(7195):1372.
47. Jones PA, Ross RK. Prevention of Bladder Cancer. *N Engl J Med*. 1999;340:1424-6.
48. Berger A. Science commentary: Why wine might be less harmful than beer and spirits. *Br Med J*. 1998;317:848.
49. Seitz HK, Simanowski UA. Alcohol and carcinogenesis. *Annu Rev Nutr*. 1988;8:99-119.
50. Garro AJ, Lieber CS. Alcohol and cancer. *Annu Rev Pharmacol Toxicol*. 1990;30:219-49.
51. Freund G. Possible relationships of alcohol in membranes to cancer. *Cancer Res*. 1979;39:2899-901.
52. Blot WJ. Invited commentary: More evidence of increased risks of cancer among alcohol drinkers. *Am J Epidemiol*. 1999;150:1138-40.
53. Yang CS, Wang ZY. Tea and cancer. *J Natl Cancer Inst*. 1993;85:1038-49.
54. Goldbohm RA, Hertog MG, Brants HA, van Poppel G, van den Brandt PA. Consumption of black tea and cancer risk: a prospective cohort study. *J Natl Cancer Inst*. 1996;88:93-100.
55. Dreosti IE, Wargovich MJ, Yang CS. Inhibition of carcinogenesis by tea: the evidence from experimental studies. *Crit Rev Food Sci Nutr*. 1997;37:761-70.
56. Katiyar SK, Mukhtar H. Tea consumption and cancer. *World Rev Nutr Diet*. 1996;79:154-84.
57. Steinmaus CM, Nunez S, Smith AH. Diet and bladder cancer: a meta-analysis of six dietary variables. *Am J Epidemiol*. 2000;151:693-702.
58. Michaud DS, Spiegelman D, Clinton SK, Rimm EB, Willett WC, Giovannucci E. Prospective study of dietary supplements, macronutrients, micronutrients, and risk of bladder cancer in US men. [In Process Citation]. *Am J Epidemiol*. 2000;152:1145-53.
59. Riboli E, Gonzalez CA, Lopez Abente G, Errezola M, Izarzugaza I, Escolar A, Nebot M, Hemon B, Agudo A. Diet and bladder cancer in Spain: a multi-centre case-control study. *Int J Cancer*. 1991;49:214-9.
60. Vena JE, Graham S, Freudenheim J, Marshall J, Zielczynski M, Swanson M, Sufrin G. Diet in the epidemiology of bladder cancer in western new york. *Nutr Cancer*. 1992;18:255-64.
61. Shibata A, Paganini Hill A, Ross RK, Henderson BE. Intake of vegetables, fruits, beta-carotene, vitamin C and vitamin supplements and cancer incidence among the elderly: a prospective study. *Br J Cancer*. 1992;66:673-9.
62. Nomura AMY, Kolonel LN, Hankin JH, Yoshizawa CN. Dietary factors in cancer of the urinary tract. *Int J Cancer*. 1991;48:199-205.
63. Kolonel LN, Hinds MW, Nomura AM, Hankin JH, Lee J. Relationship of dietary vitamin A and ascorbic acid intake to the risk for cancers of the lung, bladder, and prostate in Hawaii. *Natl Cancer Inst Monogr*. 1985;69:137-42.
64. Brummer B, White E, Vaughan TL, Cheney CL. Nutrient intake in relation to bladder cancer among middle-aged men and women. *Am J Epidemiol*. 1996;144:485-95.
65. Nomura A, Heilbrun LK, Morris JS, Stemmermann GN. Serum selenium and the risk of cancer, by specific sites: case-control analysis of prospective data. *J Natl Cancer Inst*. 1987;79:103-8.
66. Helzlsouer KJ, Comstock GW, Morris JS. Selenium, lycopene, alpha-tocopherol, beta-carotene, retinol, and subsequent bladder cancer. *Cancer Res*. 1989;49:6144-8.
67. Knekt P, Aromaa A, Maatela J, Alfthan G, Aaran RK, Hakama M, Hakulinen T, Peto R, Teppo L. Serum selenium and subsequent risk of cancer among Finnish men and women. *J Natl Cancer Inst*. 1990;82:864-8.
68. Garland M, Morris JS, Stampfer MJ, Colditz GA, Spate VL, Baskett CK, Rosner B, Speizer FE, Willett WC, Hunter DJ. Prospective study of toenail selenium and cancer among women. *J Natl Cancer Inst*. 1995;87:497-505.

69. Steinmetz KA, Potter JD. Vegetables, fruit, and cancer prevention: a review. *J Am Diet Assoc.* 1996;96(10):1027-39.
70. Michaud DS, Spiegelman D, Clinton SK, Rimm EB, Willett WC, Giovannucci EL. Fruit and vegetable intake and incidence of bladder cancer in a male prospective cohort. *J Natl Cancer Inst.* 1999;91:605-13.
71. Steinmetz KA, Potter JD. Vegetables, fruit, and cancer. II. Mechanisms. *Cancer Causes Control.* 1991;2:427-42.
72. Patterson RE, White E, Kristal AR, Neuhauser ML, Potter JD. Vitamin supplements and cancer risk: the epidemiologic evidence. *Cancer Causes Control.* 1997;8:786-802.
73. Kamat AM, Lamm DL. Chemoprevention of urological cancer. *J Urol.* 1999;161(6):1748-60.
74. Birt DF. Update on the effects of vitamins A, C, and E and selenium on carcinogenesis. *Proc Soc Exp Biol Med.* 1986;183:311-20.
75. Knäkt P, Aromaa A, Maatela J, Aaran RK, Nikkari T, Hakama M, Hakulinen T, Peto R, Saxen E, Teppo L. Serum vitamin E and risk of cancer among Finnish men during a 10-year follow-up. *Am J Epidemiol.* 1988;127:28-41.
76. Glynn SA, Albanes D. Folate and cancer: a review of the literature. *Nutr Cancer.* 1994;22:101-19.
77. Blount BC, Mack MM, Wehr CM, MacGregor JT, Hiatt RA, Wang G, Wickramasinghe SN, Everson RB, Ames BN. Folate deficiency causes uracil misincorporation into human DNA and chromosome breakage: implications for cancer and neuronal damage. *Proc Natl Acad Sci U S A.* 1997;94:3290-5.
78. Clark LC, Combs GF, Jr., Turnbull BW, Slate EH, Chalker DK, Chow J, Davis LS, Glover RA, Graham GF, Gross EG, Krongrad A, Leshner JL, Jr., Park HK, Sanders BB, Jr., Smith CL, Taylor JR. effects of selenium supplementation for cancer prevention in patients with carcinoma of the skin: a randomized controlled trial. *JAMA.* 1996;276:1957-63.
79. Salonen JT, Alfthan G, Huttunen JK, Puska P. Association between serum selenium and the risk of cancer. *Am J Epidemiol.* 1984;120:342-9.
80. Willett WC, Polk BF, Morris JS, Stampfer MJ, Pressel S, Rosner B, Taylor JO, Schneider K, Hames CG. Prediagnostic serum selenium and risk of cancer. *Lancet.* 1983;2:130-4.
81. Fan AM, Kizer KW. Selenium: nutritional, toxicologic and clinical aspects. *West J Med.* 1990;153:160-7.
82. Virtamo J, Valkeila E, Alfthan G, Punsar S, Huttunen JK, Karvonen MJ. Serum selenium and risk of cancer. A prospective follow-up of nine years. *Cancer.* 1987;60:145-8.
83. t Manneje A, Kogevinas M, Chang Claude J, Cordier S, Gonzalez CA, Hours M, Jockel KH, Bolm Audorff U, Lynge E, Porru S, Donato F, Ranft U, Serra C, Tzonou A, Vincis P, Wahrendorf J, Boffetta P. Occupation and bladder cancer in European women. *Cancer Causes Control.* 1999;10:209-17.
84. Schottenfeld D, Fraumeni JF. cancer epidemiology and prevention. New York Oxford: Oxford university press, 1996.
85. Matanoski GM, Stockwell HG, Diamond EL, Haring-Sweeney M, Joffe RD, Mele LM, Johnson ML. A cohort mortality study of painters and allied tradesmen. *Scand J Work Environ Health.* 1986;12:16-21.
86. Bethwaite PB, Pearce N, Fraser J. Cancer risks in painters: study based on the New Zealand Cancer Registry. *Br J Ind Med.* 1990;47:742-6.
87. Siemiatycki J, Dewar R, Nadon L, Gerin M. Occupational risk factors for bladder cancer: results from a case-control study in Montreal, Quebec, Canada. *Am J Epidemiol.* 1994;140:1061-80.
88. Terstegge CW, Swaen GM, Slangen JJ, Van Vliet C. Mortality Patterns among Commercial Painters in The Netherlands. *Int J Occup Environ Health.* 1995;1:303-10.
89. Englund A. Cancer incidence among painters and some allied trades. *J Toxicol Environ Health.* 1980;6:1267-73.
90. Morrison AS, Ahlbom A, Verhoek WG, Aoki K, Leck I, Ohno Y, Obata K. Occupation and bladder cancer in Boston, USA, Manchester, UK, and Nagoya, Japan. *J Epidemiol Community Health.* 1985;39:294-300.
91. Boffetta P, Silverman DT. A meta-analysis of bladder cancer and diesel exhaust exposure. *Epidemiology.* 2001;12:125-30.
92. Vineis P. Epidemiology of cancer from exposure to arylamines. *Environ Health Perspect.* 1994;102 Suppl 6:7-10.
93. Parmeggiani, editor. Encyclopaedia of occupational health and safety. Geneva: International Labour Office, 1983.
94. Marcus PM, Vineis P, Rothman N. NAT2 slow acetylation and bladder cancer risk: a meta-analysis of 22 case-control studies conducted in the general population. *Pharmacogenetics.* 2000;10:115-22.
95. Brockmoller J, Cascorbi I, Kerb R, Roots I. Combined analysis of inherited polymorphisms in arylamine N-acetyltransferase 2, glutathione S-transferases M1 and T1, microsomal epoxide hydrolase, and cytochrome P450 enzymes as modulators of bladder cancer risk. *Cancer Res.* 1996;56:3915-25.
96. Brockmoller J, Kerb R, Drakoulis N, Staffeldt B, Roots I. Glutathione S-transferase M1 and its variants A and B as host factors of bladder cancer susceptibility: a case-control study. *Cancer Res.* 1994;54:4103-11.
97. Bartsch H, Malaveille C, Friesen M, Kadlubar FF, Vineis P. Black (air-cured) and blond (flue-cured) tobacco cancer risk. IV: molecular dosimetry studies implicate aromatic amines as bladder carcinogens. *Eur J Cancer.* 1993;29a:1199-207.
98. Marcus PM, Hayes RB, Vineis P, Garcia-Closas M, Caporaso NE, Autrup H, Branch RA, Brockmoller J, Ishizaki T, Karakaya AE, Ladero JM, Mommsen S, Okkels H, Romkes M, Roots I, Rothman N. Cigarette smoking, N-acetyltransferase 2 acetylation status, and bladder cancer risk: a case-series meta-analysis of a gene-environment interaction. *Cancer Epidemiol Biomarkers Prev.* 2000;9:461-7.

Summary

RISK FACTORS FOR URINARY BLADDER CANCER

Bladder cancer is the seventh most frequent tumour in men worldwide, accounting for approximately 200,000 men with newly diagnosed bladder cancer each year. Bladder cancer is considerably more common in men than in women, but still yearly approximately 60,000 women are diagnosed with the disease worldwide. Research is warranted to study bladder cancer aetiology, in which possible risk factors for bladder cancer are explicitly tested. Knowledge about these determinants may play an important role in bladder cancer prevention. In this dissertation, analyses regarding the associations between several environmental risk factors (i.e., tobacco smoking, fluid consumption, diet and occupation) and bladder cancer incidence are reported. These analyses were carried out through meta-analyses of epidemiologic studies and through specific analyses within the context of the Netherlands Cohort Study on diet and cancer.

META-ANALYSES

In observational research regarding aetiological questions, relations are studied between risk factors on the one hand and the risk of developing a certain disease (e.g., bladder cancer) on the other hand. The operationalisation of exposure, however, may vary per study. If studies have operationalised exposure differently, then the exposure categories have to be made comparable first in order to enable meta-analysis. In **chapter 2** a statistical method for doing so is discussed. Usually only two-way contingency tables together with exposure specific adjusted odds ratios and corresponding standard errors are presented in articles. The data needed to calculate the effect per unit of exposure, however, are not always available. Another method of making exposure categories of different studies comparable is by calculating a combined effect estimate for all exposure categories combined compared to a stable reference category. In chapter 2, a consistent estimate of exposed versus non-exposed adjusted odds ratio from summarised dose-response data is proposed. This estimate is based on the weighted sum of the exposure specific odds ratios, with the prevalences of the control group as weights. Large sample variance is derived accounting for the dependency between exposure specific adjusted odds ratios.

Chapters 3–5 describe three meta-analyses on risk factors for bladder cancer (i.e., cigarette smoking, alcohol consumption, and coffee and tea consumption). In these meta-analyses, the epidemiologic literature was identified through computerized database searches in Medline, Cancerlit and Current Contents for follow-up and case-control studies published from 1966 to 2000, and through reference checking. From each study both qualitative and quantitative data were abstracted. A criteria list was developed for the assessment of quality items in observational cancer research. In the meta-analyses, summary effects were estimated both adjusted and unadjusted for potential confounders with random effects meta regression analysis.

Although narrative reviews have concluded that there is strong support for an association between cigarette smoking and urinary tract cancer, the association has never been quantified systematically in reviews. The purpose of the systematic review, as presented in **chapter 3** was to summarize and quantify the impact of different smoking characteristics (status, amount, duration, cessation, and age at first exposure) both unadjusted and adjusted for age and gender. The results suggest a substantial increase in risk of cancer of the urinary tract for cigarette smokers. Based on the results of this study and previous literature, the authors conclude that current cigarette smokers have an approximately threefold higher risk of urinary tract cancer than non-smokers. In Europe, approximately half of urinary tract cancer cases among males and one-third of cases among females might be attributable to cigarette smoking.

Recent narrative reviews have concluded that there is no support for an association between alcohol consumption and urinary tract cancer. Many individual studies, however, have reported positive associations, although rarely statistically significant. The purpose of the meta-analysis as reported in **chapter 4** was to summarize and quantify the relationship between alcohol and bladder cancer with more statistical power and to perform a sensitivity analysis on the study characteristics. We conclude that the available data suggest a slightly increased risk of urinary tract cancer from alcohol consumption for men. The risk related to alcohol consumption for women and the influence of the amount and type of alcohol remains unclear.

Previous reviews have concluded that there is a small association between coffee consumption and an increased risk of urinary tract cancer, possibly due to confounding by smoking. No association for tea consumption has been indicated. A meta-analysis on coffee and tea consumption and bladder cancer risk is described in **chapter 5**. This systematic review attempts to summarize and quantify these associations both unadjusted and adjusted for age, smoking and sex. In accordance with earlier reviews, we found that coffee consumption increases the risk of urinary tract cancer by approximately 20%. The consumption of tea seems not to be related to an increased risk of urinary tract cancer.

NETHERLANDS COHORT STUDY

In **chapters 6-12**, analyses on risk factors for bladder cancer from the Netherlands Cohort Study (NLCS) on diet and cancer are presented. In 1986, this prospective cohort study, was initiated among 120,852 men and women aged 55-69 years. The subjects originated from 204 municipal registries throughout the country. Data regarding potential risk factors for cancer were collected by means of a self-administered questionnaire. Follow-up for incident bladder cancer was established by record linkage to all regional cancer registries in The Netherlands and with a national pathology register. The case cohort approach was used for data processing and analysis. Cases were enumerated from the entire cohort, while the accumulated person years in the cohort were estimated from a subcohort sample. Following this approach, a subcohort of 3,500 subjects (1,688 men, 1,812 women) was randomly sampled from the cohort after the baseline exposure measurement. 619 Incident bladder cancer cases were detected during a follow-up period of 6.3 years (September 1986 - December 1992).

In **chapter 6** the results from the NLCS regarding associations between cigarette, cigar, pipe, environmental tobacco smoking (ETS), and bladder cancer risk are presented. We conclude that current cigarette smokers have a threefold higher bladder cancer risk than non-smokers. Ex-smokers experience a twofold increased risk. Associations of cigarette smoking with bladder cancer were largely attributable to smoking duration and less attributable to cigarette smoking amount. In this study, about half of male bladder cancer and one fifth of female bladder cancer might be attributable to cigarette smoking. Other smoking types (cigar, pipe, or ETS) were not associated with increased risks.

Chapter 7 describes the results on alcohol consumption and bladder cancer risk. The incidence rate ratios (RR) for men show a moderately increased risk of bladder cancer compared with non-drinkers. This risk followed a dose response association. Alcohol consumed from specific beverages (i.e., beer, wine, and liquor) was also associated with a moderately elevated risk, although not statistically significant. The incidence rate ratios for women varied around unity. The results of this study do not suggest an important association between alcohol consumption and bladder cancer risk.

The results from the NLCS on coffee, tea and total fluid consumption, and consequent bladder cancer risk are presented in **chapter 8**. The data suggest a possible positive association between coffee consumption and bladder cancer risk in men and a probable inverse association in women. Tea consumption was inversely associated with bladder cancer risk. Total fluid consumption did not appear to be associated with bladder cancer in both men and women.

It has been suggested that vegetable and fruit consumption may protect against cancer at various anatomical sites. However, few detailed results concerning bladder cancer have been reported so far. The impact of vegetable and fruit consumption on bladder cancer risk is discussed in **chapter 9**. In this study, total vegetable consumption was not associated with bladder cancer risk, whereas the consumption of Brassicas, total fruit and citrus fruit might be inversely associated with bladder cancer risk.

Although dietary components may have anticarcinogenic properties, few studies have investigated prospectively the association between nutrient intake and bladder cancer. **Chapter 10** describes the intake of retinol, vitamin C, vitamin E, folate and carotenoids associated with subsequent bladder cancer risk. We conclude that dietary or supplemental intake of vitamin A, vitamin C, vitamin E, and intake of folate, and most carotenoids are not associated with bladder cancer. In this study, only β -cryptoxanthin intake appeared to be inversely associated.

The association between several cancers and selenium status has been investigated in epidemiologic studies. However, few results concerning bladder cancer have been reported so far. In **chapter 11** the results from the NLCS on toenail selenium status and risk of bladder cancer are presented. We found evidence for an inverse association between selenium status and bladder cancer risk (RR=0.67 for highest versus lowest quintile of selenium status)

Analyses to estimate bladder cancer risk associated with occupational exposures to paint components, polycyclic aromatic hydrocarbons (PAH), diesel exhausts and aromatic amines were presented in **chapter 12**. This study provided marginal evidence for an association between occupational exposure to paint components, PAH, aromatic amines and subsequent bladder cancer. Despite the small proportion of exposed subjects, we found an interaction effect between occupational exposures and cigarette smoking, specifically for exposure to paint components, suggesting that the carcinogenic effect on the bladder might decrease after smoking cessation.

The epilogue (**chapter 13**) discusses the results in perspective to previous epidemiological research and evidence from other scientific disciplines. In this chapter levels of evidence for risk factors for bladder cancer are formulated.

Samenvatting

Blaaskanker is wereldwijd de zevende meest voorkomende tumor bij mannen. Ieder jaar worden ongeveer 200.000 mannen gediagnosticeerd met blaaskanker. Blaaskanker komt aanzienlijk meer voor bij mannen dan bij vrouwen, maar toch krijgen wereldwijd 60.000 vrouwen per jaar deze ziekte. Wetenschappelijk onderzoek is nodig naar de oorzaken van blaaskanker, waarin mogelijke risicofactoren expliciet kunnen worden getoetst. Kennis over deze determinanten kan een belangrijke rol spelen in de preventie van blaaskanker. In dit proefschrift worden analyses gepresenteerd waarin het verband tussen verschillende omgevingsfactoren (zoals roken, drinken, voeding en beroep) en de kans op blaaskanker is onderzocht. Deze analyses zijn uitgevoerd door middel van meta-analyses van epidemiologische onderzoeken en door middel van specifieke analyses binnen de Nederlandse CohortStudie naar voeding en kanker.

META-ANALYSES

Gepoolde effectmaten uit een meta-analyse van gepubliceerde epidemiologische observationale onderzoeken moeten vaak gebaseerd worden op meerdere odds ratio's voor verschillende blootstellingniveaus per onderzoek. De operationalisatie van blootstelling in de afzonderlijke onderzoeken kan echter verschillen. Indien men in een meta-analyse onderzoeken wil opnemen die blootstelling op verschillende manieren geoperationaliseerd hebben, moeten de blootstellingscategorieën eerst vergelijkbaar gemaakt worden. In **hoofdstuk 2** wordt een statistische methode hiervoor gepresenteerd. In het algemeen worden naast de blootstellingspecifieke odds ratio's en corresponderende standaardfouten uitsluitend het aantal patiënten en controles gepresenteerd. De benodigde gegevens voor het berekenen van een dosis-respons effect per eenheid van blootstelling zijn echter niet altijd voorhanden. Door het combineren van blootstellingscategorieën, kan een samenvattende effectmaat worden berekend voor alle blootstellingscategorieën tezamen ten opzichte van een gelijke referentiecategorie. In hoofdstuk 2 wordt een methode voorgesteld om een dergelijke samenvattende odds ratio te berekenen. Deze schatting is gebaseerd op de gewogen som van de blootstellingspecifieke odds ratio's, waarbij de prevalentie van de controles als gewicht wordt gebruikt. De informatie over de blootstellingspecifieke standaardfouten kan worden gebruikt om een uitspraak te doen over de precisie van de voorgestelde samenvattende effectmaat.

Hoofdstuk 3-5 beschrijven drie meta-analyses naar risicofactoren van blaaskanker (roken, alcohol-, koffie- en theeconsumptie). De epidemiologische literatuur voor deze meta-analyses is verzameld via de literatuurbestanden van: Medline, Cancerlit en Current Contents waarin is gezocht naar cohort en patiënt-controle onderzoeken uit 1966-2000. Daarnaast zijn de referentielijsten van relevante publicaties doorzocht. Van ieder onderzoek zijn zowel kwalitatieve als kwantitatieve gegevens geabstraheerd. Er is een criterialijst ontwikkeld om de kwaliteitskenmerken van een onderzoekspublicatie te kunnen evalueren. In de meta-analyse zijn de gepoolde effectmaten geschat met behulp van 'random effects' meta-regressie analyse, waarin is gecorrigeerd voor potentiële confounders.

Hoewel verhalende reviews concluderen dat er een sterk verband bestaat tussen het roken van sigaretten en blaaskanker, is deze associatie nog nooit systematisch gekwantificeerd in een review. Het doel van de systematische review, zoals gepresenteerd in **hoofdstuk 3** is het samenvatten en kwantificeren van de invloed van verschillende rookkarakteristieken zoals rook status, frequentie, duur, stoppen met roken en startleeftijd gecorrigeerd voor leeftijd en geslacht. De resultaten suggereren een substantieel verhoogd risico op blaaskanker voor sigarettenrokers. Uit de resultaten van dit onderzoek en voorgaande literatuur blijkt dat sigarettenrokers drie keer meer kans lopen op het krijgen van blaaskanker dan mensen die niet roken. Vertaald naar de Europese Unie, waar 43 procent van de mannen en 28 procent van de vrouwen rookt,

kan op basis van deze analyse worden geconcludeerd dat naar schatting 50 procent van het vóórkomen van blaaskanker bij mannen en 35 procent bij vrouwen het gevolg is van roken

Recente verhalende reviews hebben geconcludeerd dat er geen verband is tussen het drinken van alcohol en de kans op blaaskanker. Veel afzonderlijke onderzoeken rapporteren weliswaar positieve relaties tussen alcohol consumptie en de kans op blaaskanker, maar de resultaten zijn vaak niet statistisch significant. Het doel van de meta-analyse uit **hoofdstuk 4** is het samenvatten en kwantificeren van de relatie tussen alcoholconsumptie en blaaskanker met meer statistische power dan in voorgaande onderzoeken en het uitvoeren van een sensitiviteitsanalyse op de onderzoekskenmerken. Wij concluderen uit de beschikbare gegevens dat er een 30 procent verhoogd risico op blaaskanker is voor mannen die alcohol drinken ten opzichte van geheelonthouders. Of ook vrouwen die alcohol drinken een verhoogd risico op blaaskanker hebben blijft vooralsnog onbekend.

Eerdere reviews hebben geconcludeerd dat er een zwak verband bestaat tussen het drinken van koffie en de kans op blaaskanker dat mogelijk veroorzaakt wordt door confounding door roken. Er is geen verband gevonden met het drinken van thee.

In **hoofdstuk 5** is de huidige literatuur naar het verband tussen koffie en theeconsumptie en de kans op blaaskanker systematisch samengevat en zijn gevonden associaties gekwantificeerd, waarbij is gecorrigeerd voor leeftijd, roken en geslacht. In overeenstemming met eerdere reviews, vonden wij dat door het drinken van koffie de kans op blaaskanker met 20% toeneemt. De consumptie van thee lijkt niet geassocieerd te zijn met een toename in het blaaskankerrisico.

NEDERLANDSE COHORTSTUDIE

In **hoofdstuk 6-12** worden analyses naar risicofactoren voor blaaskanker van de Nederlandse CohortStudie (NLCS) naar voeding en kanker gepresenteerd. In 1986 is dit prospectief cohortonderzoek van start gegaan onder 120.852 mannen en vrouwen in de leeftijd van 55-69 jaar. De onderzoeksdeelnemers zijn geïdentificeerd via gemeenteregisters van 204 Nederlandse gemeenten. De gegevens over potentiële risicofactoren van kanker zijn verzameld door middel van een vragenlijst. De follow-up voor incidentie blaaskanker is bewerkstelligd door een koppeling te maken met alle regionale kankerregistraties en met het nationaal pathologie register. Voor de analyse is gebruik gemaakt van een case-cohort benadering. Hierin ontstaan patiënten uit het hele cohort, terwijl de cumulatieve persoonsjaren worden geschat aan de hand van een random steekproef uit het cohort. Dit subcohort bestaat uit 3.500 personen (1.688 mannen en 1.812 vrouwen). Na een follow-up periode van 6,3 jaar (september 1986 t/m december 1992) zijn 619 patiënten met blaaskanker geïdentificeerd.

In **hoofdstuk 6** worden de resultaten van de NLCS omtrent de relatie tussen sigaretten, sigaar, pijp en passief roken en de kans op blaaskanker beschreven. We concluderen op basis van dit onderzoek dat het roken van sigaretten de kans op blaaskanker verdrievoudigt. Ex-rokers hebben nog steeds een twee keer zo hoog risico op blaaskanker dan niet-rokers. De associaties die met het roken van sigaretten zijn gevonden zijn bijna volledig terug te voeren op het aantal jaren dat iemand (heeft ge)rookt; de hoeveelheid sigaretten is van minder belang. Van ongeveer de helft van de mannelijke blaaskankerpatiënten en een vijfde van de vrouwelijke blaaskankerpatiënten in de NLCS is het ontstaan van blaaskanker mogelijk te wijten aan het roken van sigaretten. Andere vormen van roken, zoals sigaar, pijp en passief roken, lijken niet in verband te staan met een verhoogd blaaskankerrisico.

Hoofdstuk 7 beschrijft de resultaten over alcoholconsumptie en de kans op blaaskanker. De relatieve risico's (RR) voor mannen laten een matig verhoogd risico op blaaskanker zien voor alcohol drinkers in vergelijking met geheel onthouders. Dit risico neemt toe met het aantal gram alcohol per dag. Alcoholinname via bier, wijn en sterke drank zijn eveneens geassocieerd met matig verhoogde risico's, hoewel de meeste relatieve risico's niet statistisch significant zijn. De relatieve risico's voor vrouwen lieten geen verband zien tussen het drinken van alcohol en de kans op blaaskanker. De resultaten van dit onderzoek suggereren dat er geen belangrijk verband lijkt te bestaan tussen alcoholconsumptie en de kans op blaaskanker.

De resultaten van de NLCS over koffie, thee, totale vloeistofconsumptie en de kans op blaaskanker worden gepresenteerd in **hoofdstuk 8**. De gegevens suggereren een mogelijk positief verband tussen koffieconsumptie en de kans op blaaskanker bij mannen en een waarschijnlijk negatief verband bij vrouwen. Het drinken van thee was negatief geassocieerd met de kans op blaaskanker. De totale vloeistofconsumptie toonde geen verband met de kans op blaaskanker bij zowel mannen als vrouwen.

Groente en fruit kunnen mogelijk beschermend werken tegen verschillende vormen van kanker. Er zijn echter maar weinig onderzoeken uitgevoerd naar de relatie tussen groenten en fruit en de kans op blaaskanker. Deze invloed van groente en fruit is beschreven in **hoofdstuk 9**. Uit dit onderzoek blijkt dat de consumptie van groente in het algemeen niet geassocieerd is met de kans op blaaskanker, hoewel de consumptie van Brassicagroenten negatief geassocieerd is. Fruit in het algemeen en citrusfruit in het bijzonder laten eveneens een invers verband met het risico op blaaskanker zien.

Hoofdstuk 10 beschrijft de relatie tussen retinol, vitamine C, vitamine E, foliumzuur en de inname van carotenoiden en de kans op blaaskanker. We concluderen dat de inname van vitamine A, vitamine C, vitamine E via de voeding of via supplementen en de inname van foliumzuur en de meeste carotenoiden niet zijn geassocieerd met de kans op blaaskanker. In dit onderzoek lijkt alleen β -cryptoxanthine een negatief verband te vertonen.

Diverse onderzoeken hebben een beschermende werking van selenium tegen verschillende soorten kanker aangetoond. Er zijn echter nog weinig resultaten bekend omtrent de relatie tussen selenium en blaaskanker. In **hoofdstuk 11** wordt het seleniumgehalte in teennagels van deelnemers van de NLCS in verband gebracht met de kans op blaaskanker. We vonden een negatief verband tussen seleniumstatus en de kans op blaaskanker ($RR=0.67$ voor hoogste versus laagste quintiel van selenium status).

De relaties tussen enerzijds de kans op blaaskanker en anderzijds beroepsmatige blootstelling aan verfstoffen, polycyclische aromatische koolwaterstoffen (PAK), dieseluitlaatgassen en aromatische amines worden gepresenteerd in **hoofdstuk 12**. De resultaten bieden matig bewijs voor een positief verband tussen beroepsmatige blootstelling aan verfstoffen, PAKs, aromatische amines en de kans op blaaskanker. Ondanks de kleine aantallen blootgestelde personen in het onderzoek, vonden wij een interactie-effect tussen beroepsblootstellingen en het roken van sigaretten, met name voor blootstelling aan verfstoffen. Dit suggereert dat het carcinogene effect van verfstoffen op de blaas wellicht afneemt nadat men is gestopt met roken.

In de epiloog (**hoofdstuk 13**) worden de resultaten verder besproken in relatie tot voorgaande epidemiologische literatuur en wetenschappelijk bewijs uit andere disciplines. In dit hoofdstuk wordt een poging ondernomen om het beschikbare bewijs omtrent de risico factoren van blaaskanker samen te vatten.

Dankwoord en Curriculum Vitae

DANKWOORD

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ABOUT THE AUTHOR

Maurice Petrus Alphonsus was born on April 11, 1972 in Brunssum, the Netherlands. After completing the secondary school (MAVO/HAVO) in 1990 at the St. Janscollege in Hoensbroek, he studied Occupational Therapy at Hogeschool Heerlen. After obtaining his Bc. degree in 1994 Maurice worked as an occupational therapist in his own practice and in several health care institutes in Limburg until 1997. At the same time he studied Health Education and Promotion (M.H.S.) and Epidemiology (M.Sc.) at Maastricht University, Faculty of Health Science, Maastricht. After his graduation in September 1997 he started his Ph.D.-project on risk factors for bladder cancer using data from the Netherlands Cohort Study on diet and cancer. The research was carried out at the department of Epidemiology, Maastricht University and was embedded in the Nutrition and Toxicology Research Institute Maastricht. He was engaged in teaching epidemiology, statistics and meta-analysis to pre- en postgraduate students. Since February 2001 Maurice is appointed as an assistant professor of genetic epidemiology at this department.