Uric acid, blood pressure, and gout management

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Beneath the surface

Cover artwork: An Inescapable Pressure | oil on canvas, 140cm x 120cm, 2015. From the project Crystallisation of Thought | An artistic interpretation of the disease Gout by artist Jill Tegan Doherty. Image ©Jill Tegan Doherty 2017

"A collaboration between Prof. Michael Doherty and artist Jill Tegan Doherty, initiated to help raise awareness of the disease Gout. To bring to light that it is a disease which needs to be taken seriously. The project aims to break down the barriers to care and bring strength in knowledge to patients and potential sufferers."

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PROEFSCHRIFT

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CONTENTS

	Glossary of terms related to genetics *or* medication adherence	7
Chapter 1	General introduction	9
Part I	**Uric acid and blood pressure: the role of uric acid production**	**27**
Chapter 2	Xanthine oxidase gene variants and their association with blood pressure and incident hypertension: a population study	29
Chapter 3	Associations of plasma uric acid and purine metabolites with blood pressure in children: the KOALA birth cohort study	51
Chapter 4	Uric acid and blood pressure: exploring the role of uric acid production in The Maastricht Study	73
Part II	**Gout management by the patient and general practitioner**	**89**
Chapter 5	Medication adherence among gout patients initiated allopurinol: a retrospective cohort study in the Clinical Practice Research Datalink	91
Chapter 6	Medication adherence among patients with gout: a systematic review and meta-analysis	109
Chapter 7	Knowledge, illness perceptions and stated clinical practice behaviour in management of gout: a mixed methods study in general practice	137
Chapter 8	Summary and general discussion	153
Addendum		**175**
	Valorisation	177
	Samenvatting	183
	Acknowledgments	189
	Curriculum vitae	195
	List of publications	197

Glossary of terms related to genetics *or* medication adherence

Genetic terms	
Allele	A specific variation of a single-nucleotide polymorphism or gene.
Enzyme	Molecules that accelerate, or catalyse, chemical reactions.
Gene	Comprises a DNA sequence, including introns, exons, and regulatory regions, related to transcription of a given RNA.
Genotype	Part of a DNA sequence and determines a specific trait. If the alleles of the chromosomes are the same, the genotype is called homozygous, if different, heterozygous.
Linkage disequilibrium (LD)	Correlation between alleles at different loci within the population. The term LD describes a state which is opposite to the hypothetical situation in which all loci exhibit complete independence, called linkage equilibrium.
Mendelian randomization studies	In epidemiology, this is a method of using variation in a gene, as a proxy for an exposure, to examine the causal effect of the modifiable exposure on the disease.
Phenotype	Expression of characteristic of a trait, for example body height or blood pressure.
Pleiotropic effect	Situation in which a single gene / polymorphism influences a variety of traits (phenotypes)
Medication adherence terms	
Adherence to medication (*general term)*	The process by which patients take their medications as prescribed, including initiation, implementation and discontinuation.
Adherence	The extent to which a patient's actual medication intake corresponds to the prescribed dosing regimen during observation time.
Discontinuation	Marks the end of therapy, when the next dose to be taken is omitted and no more doses are taken thereafter *or* when the next dose is after the permissible gap length (e.g. at least 30 days after finishing the last prescription).
Implementation	The extent to which a patient's actual dosing corresponds to the prescribed dosing regimen, from initiation until discontinuation.
Initiation	The time from prescription until first dose is taken.
Non-persistence	*See Discontinuation*
Persistence	Is the length of time between initiation and the last dose, which immediately precedes discontinuation.
Time until discontinuation / non-persistence	*See Persistence*

1

General introduction

Adapted from

'Overproducers' and 'underexcretors': the key to understanding the complex relation between uric acid and cardiovascular diseases?

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To be submitted

GENERAL INTRODUCTION

Gout is the most prevalent form of inflammatory rheumatic disease, and is characterized by the deposition of monosodium urate crystals in the synovial fluid.[1] Unlike other forms of arthritis, the pathogenesis of gout is well understood and effective treatments to reduce serum uric acid concentration below the saturation point of monosodium urate are available.[2] If untreated, tophaceous gout can cause a severe inflammatory response leading to cartilage and bone destruction.[3,4] In the long-term, it can result in impaired function and a decline in health-related quality of life.[5] Although gout is considered a well treatable disease, it is often insufficiently controlled. Both patients and physicians factors can be barriers to optimal management.[6,7] To improve gout management, insights into patient medication adherence and gout management by the general practitioner are essential.

Patients with gout have an increased risk of cardiovascular disease, which is often attributed to the hyperuricemia.[8] With a prevalence of 20% asymptomatic hyperuricemia is far more prevalent than gout,[9] and has indeed been associated with an increased risk for hypertension and cardiovascular diseases.[10-12] However, there has been much debate on the causal nature of these associations. Some argue that elevated serum uric acid is just an innocent bystander in the pathogenesis of hypertension and cardiovascular diseases. From 2001 onwards, experimental studies have however established plausible mechanisms linking uric acid with the development of hypertension by using rat models.[13,14] One insight derived from these studies assigns an important role to the generation of free radicals during the production of uric acid. So far, epidemiological studies have focused on the association between blood pressure and concentrations of uric acid in serum or plasma and have ignored the distinction between uric acid *concentration* and its *production*.[12] Since the production of uric acid may contribute, independent of uric acid concentration, to the pathogenesis of hypertension, the production should be investigated as well.

This thesis comprises two main parts. In *Part I*, the association between uric acid and blood pressure, in particular the role of uric acid production is studied. In *Part II*, medication adherence in patients with gout requiring uric acid lowering therapy and the knowledge, illness perceptions and stated clinical practice behaviour of general practitioners when managing gout are addressed.

Uric acid metabolism

Uric acid is generated as part of the normal turnover of the purine nucleic acids adenine and guanine (Figure 1). The purine nucleotides are either derived from endogenous (DNA, RNA and ATP) or exogenous food-derived purines. During the last two steps of the purine metabolism, xanthine oxidoreductase (XOR) catalyses the breakdown of hypoxanthine to xanthine, and xanthine to uric acid. In most vertebrates uric acid is

further catabolized into the more soluble allantoin by the enzyme uricase, but humans and higher primates lack this enzyme.[15]

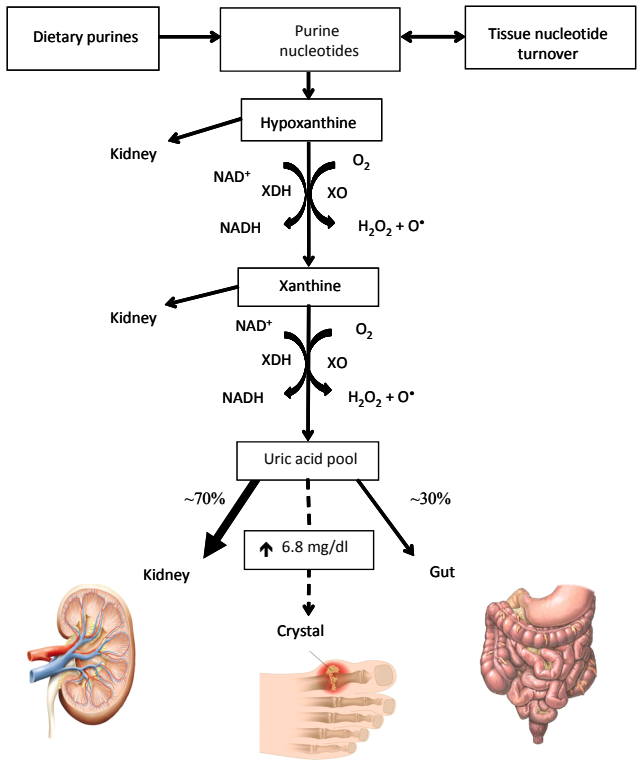

Figure 1. Purine metabolism

In healthy individuals, serum uric acid is maintained within the normal range by a complex system of secretion, reabsorption, and excretion via the kidneys or intestines. Under normal conditions, about 90% of the uric acid filtered is actively reabsorbed in the renal proximal tubules. The other 10% is for two-third excreted via the kidneys, and the remaining third via the 'extra-renal' pathway (gastrointestinal tract).[8] Several transporters involved in uric acid secretion and reabsorption are known. In recent years, genome-wide association studies found several novel loci that were associated with serum uric acid concentrations, including *SLC22A12 (*URAT1), *SLC2A* (GLUT9), *SLC17A3* (NPT4), and *ABCG2* (Q141K) etc.[16-18] These loci may indicate proteins involved in the regulation of uric acid excretion, but functional data are not yet available for all of these loci.

An unbalance, either in uric acid overproduction, underexcretion or both can cause hyperuricemia. Generally hyperuricemia is defined as a serum uric acid concentration

greater than the solubility of uric acid (408 µmol/L).[19] In approximately 90% of the cases hyperuricemia is caused by underexcretion of uric acid, as a result of reduced renal function or the use of diuretics. In the other cases, overproduction of uric acid might cause hyperuricemia. This can be due to conditions with a high cell turnover such as tumour lysis syndrome or a genetic disorder of enzymes involved in the purine metabolism. For example, a deficiency in hypoxanthine-guanine phosphoribosyl transferase (HGPRT), leads to accumulation of hypoxanthine and guanine, and causes elevated serum uric acid concentrations.[20] Next to this, consumption of diets containing large amount of purines (e.g. organ meat, seafood, beer), or intake of beverages sweetened by fructose whose digestion leads to ATP depletion and consequently the generation of adenosine monophosphate (AMP), may also increase serum uric acid concentrations.[21]

Uric acid and blood pressure

Hypertension, a lasting increase in blood pressure, is the most potent risk factor for cardiovascular disease. Globally, approximately 25–35% of the adult population and more than 60% of the individuals over the age of 65 years have high blood pressure.[22] Since the 1980s uric acid has been suspected to be a risk factor for hypertension.[23] It is proposed that uric acid induces hypertension via several mechanisms.

One of the underlying mechanisms is the activation of the renin-angiotensin system, leading to increased production of the vasoconstrictor angiotensin II.[24] Hereby, uric acid affects directly the blood vessel and causes a reversible increase in blood pressure. Prolonged elevation of uric acid leads to the second and irreversible phase of hypertension (Figure 2).[25] One pathway involves the uptake of uric acid by vascular smooth muscle cells (VSMCs), which causes proliferation and arteriolosclerosis that impairs pressure natriuresis, causing sodium-sensitive hypertension. Also, inflammation has been put forward as a possible underlying mechanism.[26-28] The crystallisation of monosodium urate in joints or other tissues triggers the inflammasome (e.g. cryopyrin/NLRP3) to generate the proinflammatory cytokine IL-1.[29,30]

Next to experimental studies, the association between uric acid and blood pressure has been extensively investigated in an epidemiological manner. Recent meta-analyses showed a significant association between serum uric acid and incident hypertension, independent of traditional risk factors.[12,31] However, the association weakened in more recent studies.[20] The later studies tended to adjust for more covariates, and some of these covariates might have been at the causal pathway of interest. Whether there is overadjustment or uric acid should indeed be considered as an innocent bystander instead of a true risk factor is still undecided.

More conclusive evidence can be provided by the use of a Mendelian randomization approach, which accounts for unmeasured confounding and for reverse causation by using genetic variants as proxies for the exposure.[32] Studies using a Mendelian randomization approach, by using genetic markers associated with serum uric acid

concentration (e.g. *SLC2A9,* a uric acid transporter), have not been able to convincingly demonstrate that the association between uric acid and blood pressure is causal.[16,33-39] The findings from these studies were generally interpreted as lack of a causal association between uric acid and blood pressure. However, polymorphisms in uric acid excretion genes only explain variation in serum uric acid concentration, but not in uric acid production, which we hypothesize to be a relevant factor for the development of hypertension.

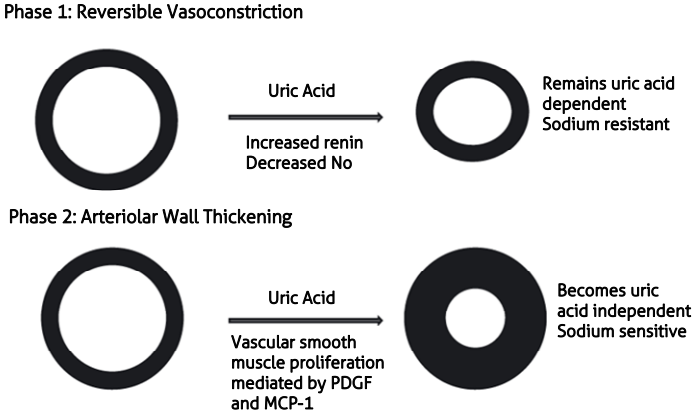

Figure 2. Two phases how uric acid may influence blood presure[25]

The role of uric acid production

In the two terminal steps of uric acid production, XOR is the sole enzyme responsible for the oxidation of hypoxanthine to xanthine, and the latter to uric acid. XOR exists in two forms, xanthine dehydrogenase (XDH) and xanthine oxidase (XO), which both occur in vivo. Initially, the XOR enzyme exists in XDH form, but when released into the circulation it is converted into XO and circulates to remote sites where it binds to the surface of endothelial cells.[15,40-45] Although XDH preferentially reduces nicotinamide adenine dinucleotide (NAD+), both forms of the enzyme can also reduce molecular oxygen to form the reactive oxygen species (ROS) superoxide and hydrogen peroxide. Thus, during the production of uric acid, ROS are formed which may cause oxidative stress. Furthermore, the generated superoxide increases the formation of peroxynitrite, leading to an increase in endothelial NO synthase (eNOS) uncoupling resulting in even more ROS formation. Next to this, angiotensin II substantially increases endothelial XO activation in cultured endothelial cells, subsequent to NAD(P)H oxidase activation, leading to even more ROS formation and inactivation of the vasodilator NO.[46] Through these mechanisms, XOR is believed to be involved in the pathogenesis of ischemia-reperfusion injury. Elevated concentrations of circulating XOR have been found in several conditions,

including atherosclerosis,[15] suggesting that indeed the increased production of uric acid and the concomitant formation of ROS may be involved in the aetiology of hypertension and cardiovascular diseases.

Proxies for uric acid production

Since the production of uric acid may contribute, independent of uric acid concentration, to the pathogenesis of hypertension, the production should be investigated as well. Facing the problem that it is not possible to directly measure uric acid production in a large population of individuals and the concentration of uric acid in serum is not an adequate marker for production, proxies for uric acid production need to be investigated. In this thesis we studied three proxies for uric acid production: (i) variants of the *XOR* gene, (ii) different ratios of the purine metabolites, and (iii) 24-h urinary uric acid excretion.

(i) Genetic variants of the xanthine oxidoreductase gene

The human *XOR* gene, encoding the XOR enzyme, has been mapped to chromosome 2p22.[47-49] The activity of the enzyme is the highest in liver[50-52] and intestine[53], whereas most other tissues show little activity.[54-56] Two in-vitro studies addressed the impact of variation in the *XOR* gene on XOR activity.[57,58] Molecular studies of the DNA of 96 Japanese individuals, revealed three nonsynonymous single nucleotide polymorphisms (SNPs). Functional characterization of 21 *XOR* variants revealed deficient enzyme activity in two, and reduced and enhanced activity in six and two, respectively.[57] Among 13 SNPs in the *XOR* 5'-flanking region, one was associated with decreased expression in transfected cells.[58] The association between XOR expression and variation in the gene offers the opportunity to explore the role of *XOR* as a hypertension susceptibility gene by using SNPs in the *XOR* gene as a proxy for uric acid production.

(ii) Ratios of purine metabolites

In the terminal steps of purine catabolism, the rate-limiting enzyme XOR catalyses the oxidation of hypoxanthine to xanthine and xanthine to uric acid. An increased XOR activity, resulting in increased uric acid production, may lead to a relative decrease in the more upstream metabolites, thus, lower hypoxanthine compared to xanthine and lower xanthine compared to uric acid concentration, resulting in higher ratios of xanthine/hypoxanthine, uric acid/xanthine, and uric acid/hypoxanthine. The latter is based on animal studies and studies examining patients with *XOR* deficiency, a genetic disorder called xanthinuria, who have decreased concentrations of the more upstream metabolites.[59-62]

(iii) Uric acid excretion in urine

Uric acid is produced during the metabolism of endogenous and exogenous purines. Once produced, uric acid cannot be further metabolized and needs to be eliminated by either renal or extra-renal excretion, so that serum uric acid remains stable and within the normal range of 200–408 μmol/L for men and 120-340 μmol/L for women. In the clinic gout patients with hyperuricemia are classified as overproducers or underexcretors based on the amount of uric acid excreted in the urine in 24 hours.[63] According to this criterion, approximately 10% of patients with hyperuricemia are overproducers.[8] Overproduction may however have implications beyond individuals with hyperuricemia and can also occur in patients with normal serum uric acid concentration. If their excretion capacity is adequate, they will not develop hyperuricemia, but may still suffer from the adverse consequences of increased uric acid production. Previous studies have shown that an increase in uric acid production, either by an increased endogenous or exogenous supply, increases urinary uric acid excretion.[64,65] Since uric acid is predominantly excreted via the kidneys, we studied 24-h urinary uric acid excretion as a proxy for uric acid production.

Blood pressure components

Several components can be useful in examining the association between uric acid and blood pressure, including diastolic and systolic blood pressure, mean arterial pressure and pulse pressure. Each component might reflect a different pathophysiological state and the relative importance of the component in predicting cardiovascular risk differs according to age (Table 1).

In younger individuals (<50 years of age), diastolic and to a lesser extent systolic blood pressure are the main predictors of cardiovascular risk. After the age of 50 pulse pressure supersedes these to become the main predictor of cardiovascular risk.[66-68] Franklin et al, showed that in these older individuals combining pulse pressure and mean arterial pressure is superior to any single blood pressure component for predicting cardiovascular risk.[69] Moreover, pulse pressure and mean arterial pressure may also provide greater insight into the different arterial stiffness and peripheral resistance, respectively.[69]

The steady component of blood pressure, captured by mean arterial pressure, reflects peripheral vascular resistance. As previously described, there are several plausible mechanisms how uric acid might increase vascular resistance, for example via accumulation of ROS.[24] Prolonged exposure to increased peripheral vascular resistance and high blood pressure leads to embedding of the vessel structure in a remodelled extracellular matrix.[70,71] Initially, the remodelling of arteries will be adaptive, but it eventually becomes maladaptive and increases stiffness, contributing to cardiovascular complications of hypertension.[70] This is reflected in the pulsatile blood components, captured by pulse pressure.

Table 1. Simplified overview of the characteristics of the different blood pressure components

Component	Causes for elevation	Reflecting	Calculation	Indicator for cardiovascular risk
Systolic blood pressure	Stiffening of the arteries, due to high blood pressure or fatty deposits damaging the walls (atherosclerosis)	Main determined of pulsatile component	Maximum pressure during one heartbeat	Young, <50 years of age
Diastolic blood pressure	Increased peripheral resistance, due to vasoconstriction	Main determined of steady component	Minimum pressure in between two heart beats	Young, <50 years of age
Pulse pressure	Stiffening of the arteries, due to high blood pressure or fatty deposits damaging the walls (atherosclerosis)	Pulsatile component	Systolic minus diastolic blood pressure	Old, >50 years of age
Mean arterial pressure	Increased peripheral resistance, due to vasoconstriction	Steady component	Diastolic blood pressure plus ⅓ of pulse pressure	Old, >50 years of age

Gout management

Gout is the most common form of inflammatory arthritis, caused by formation of monosodium urate crystals in and around peripheral joints as a consequence of persistent hyperuricemia. The prevalence varies from 1.4%[72] in Europe to 3.9%[9] in the United States and several studies showed an increase in incidence and prevalence. For example, the prevalence increased by 63.9% and the incidence increased by 29.6% between 1997 and 2012 in the UK.[73]

Gout is considered a well-treatable disease: the inflammation of acute attacks can be treated by nonsteroidal anti-inflammatory drugs (NSAIDS), colchicine or corticosteroids, and in case of recurrent attacks or tophaceous gout reduction of serum uric acid is recommended. The most used uric acid lowering agents are the XOR inhibitors allopurinol and febuxostat. Despite our understanding of the pathophysiology and availability of effective treatment, the management of gout is considered as suboptimal.[7,74] Given the rising burden of gout on the population[73] and its associated impact on patients, insights into the barriers of gout management are required. Common barriers for effective management can be distinguished between patients and physician barriers and, as it is likely that both play a significant role, both should be addressed.[6]

Patients medication adherence

Poor medication adherence to long-term uric acid lowering therapy may be an important contributor to suboptimal outcomes seen in gout. Patients not taking their medication risk having a gout flare and the chance of obtaining joint damage will become higher.[75] Moreover, the healthcare provider might unnecessarily increase the dose of uric acid lowering therapy or prematurely switch to other, more expensive therapies, like benzbromarone. As in other chronic conditions, medication adherence tends to be poor, because patients might not feel ill, do not experience the effect of the medication but

only the side-effects, or are more afraid for the side effects of the medication than for the disease. Across seven common chronic conditions including hypertension, osteoporosis, and diabetes, patients suffering from gout had the poorest medication adherence.[76] A systematic overview of available literature addressing the problem of poor medication adherence among gout patients is needed.

Definition and measurement of medication adherence

According to the World Health Organization adherence is defined as *'the extent to which a person's behaviour – taking medication, following a diet, and/or executing lifestyle changes, corresponds with agreed recommendations from a healthcare provider'*.[77] There is no "gold standard" for measuring adherence.[78] Medication adherence can be measured either directly, for example by measuring drug metabolites or drug levels in blood, urine or tissues, or indirectly, by self-report, electronic monitoring, pharmacy records or healthcare provider assessment . Independently of the measurement technique used, the threshold for defining "good" and "poor" medication adherence is arbitrary.[78]
To characterize medication adherence different concepts should be studied. According to the new taxonomy from 2012 it includes *initiation, implementation, non-persistence,* and overall *adherence* during observation time (Figure 3).[79] Poor medication adherence can thus occur in the following situations or combinations thereof: late or non-initiation of the prescribed treatment, sub-optimal implementation of the dosing regimen, or early discontinuation of the treatment.[79]

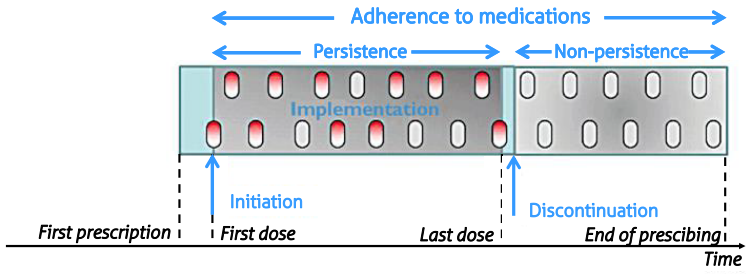

Figure 3. Illustration of the process of adherence to medication [79]

Thus far, the available literature addresses mostly the problem of *adherence* during the entire observation time, but little is known on the dynamics of medication use among gout patients. While non-persistence, the occurrence of a gap in therapy, may have little clinical effect in certain chronic diseases, an interruption in uric acid lowering therapy might actually trigger or prolong gouty attacks.[80] Furthermore, it is unknown whether those experiencing a gap return to therapy and if medication adherence improved after restarting therapy. Further research is therefore required to obtain insights into dynamic patterns of medication adherence and to identify patients at high risk of becoming non-adherent and non-persistent. Several factors such as older age, suffering from certain

comorbidities like hypertension and diabetes, and the use of anti-hypertensive medication were associated with higher adherence rates, but these factors have not or poorly (e.g. age) been studied in relation to persistence.

Gout management by the general practitioner

General practitioners are the most relevant healthcare professional when it comes to the diagnosis and treatment of gout. Several societies published recommendations for the management of gout, including the American College of Rheumatology (ACR) and the European League Against Rheumatism (EULAR).[81-83] Despite these recommendations, the management of gout is considered suboptimal.[84-86] A study conducted in the UK among primary care physicians showed that lifestyle advice was infrequently offered and allopurinol was only prescribed to a minority.[86] Appropriate initiation and dosing of uric acid lowering therapy are the main factors to achieve the therapeutic target level for serum uric acid.[87] Whenever this fails, patients are at increased risk for having more gout flares and for developing tophi and subsequent joint damage. Furthermore, a more severe and untreated disease will in the long run lead to increased gout-related healthcare costs.[88,89]

Clinical practice behaviour is influenced by several aspects, including gout knowledge and illness perceptions. Up to now only a few small qualitative studies have been performed to elucidate barriers from the general practitioner side. They showed a moderate knowledge of the causes and consequences of gout.[6,90] Some general practitioners revealed that they still had a negative stereotype view and perceived stigma that the disease is self-inflicted and only a consequence of an unhealthy life-style.[6] In some cases gout was still considered as an acute disease rather than a chronic disease, and the long-term management could therefore be inadequate.[6,91] Although it is recommended to prescribe uric acid lowering therapy in patients with recurrent flares or tophi, this is only done for a small percentage of patients. The same applies to measurement of serum uric acid concentrations on a regular basis.[73,91]

Taken together, knowledge, illness perceptions, and clinical behaviour of the general practitioner are important aspects to achieve optimal gout management. Because of the complex interactions between these aspects, studies addressing all these aspects at the same time are essential when aiming to improve gout management. However, this has not been done yet.

Main aims

The research presented in this thesis was conducted in several population-based cohort studies and made use of general practitioner interviews. The aim of this thesis was twofold:

(i) to investigate the role of uric acid production in the association between uric acid and blood pressure; and

(ii) to explore medication adherence among gout patients and gout management by the general practitioner.

OUTLINE OF THIS THESIS

This thesis is divided into two parts. In *Part I* we examined the association between uric acid and blood pressure, in particular the role of uric acid production. In *Part II* of this thesis we studied medication adherence among gout patients and gout management of general practitioners.

Part I Uric acid and blood pressure: the role of uric acid production

In *chapter 2,* we examined associations of genetic variation in the *XOR* gene, reflecting uric acid production, with secular trends in the steady and pulsatile blood pressure components and the risk of hypertension. The study was conducted in the Flemish Study on Environment, Genes and Health Outcomes (**FLEMENGHO**) and the European Project on Genes in Hypertension (**EPOGH**). The FLEMENGHO study is a prospective population-based cohort study.[92] From August 1985 to November 1990 participants were recruited in the geographically defined area in Northern Belgium. The EPOGH project recruited participants from 1999 to 2001 in four centres: Pilsen (Czech Republic), Mirano (Italy), Kraków (Poland), and Novosibirsk (Russian Federation).[93] In EPOGH, one follow-up examination took place from 2006 to 2008 and in FLEMENGHO follow-up lasted until 2013. In total, data from 4535 individuals who participated in FLEMENGHO or the EPOGH project were available. Data of 2769 participants with a blood pressure measurement *and* genotype at *XOR* was used.

In *chapter 3,* we examined associations of purine metabolite ratios, considered as proxies for an increased XOR activity, with blood pressure and extended existing evidence on the association of plasma uric acid with blood pressure to school-age children. The study was conducted in the Dutch **KOALA Birth Cohort Study**, an acronym (in Dutch) for: Child, parents and health: lifestyle and genetic constitution. From October 2000 until December 2002, a total of 2834 pregnant women were recruited at 34 weeks of gestation. A subgroup of 1204 parents was asked for consent for a home visit for anthropometric measurements, collection of venous blood, and a blood pressure measurement from their child at the age of 6-7 years. Data of 246 children with a blood pressure measurement *and* a venous blood collection was used.

In *chapter 4,* we examined associations of serum uric acid and uric acid production, captured by urinary uric acid excretion, with the steady and pulsatile blood pressure components and prevalent hypertension. This study was conducted in **The Maastricht Study**, an observational cohort study. All individuals aged between 40 and 75 years and living in the southern part of The Netherlands were eligible for participation. The study focuses on the aetiology, pathophysiology, complications, and comorbidities of type 2

diabetes mellitus and is characterized by an extensive phenotyping approach. For reasons of efficiency, the study population was enriched with type 2 diabetes mellitus patients. In total, 3451 participants completed the baseline survey between November 2010 and September 2013. Cross-sectional data was used from a subset of 2555 participants from whom a 24-h blood pressure measurement, 24-h urine collection *and* venous blood samples were obtained.

Part II Gout management by the general practitioner and patient

In *chapter 5*, we assessed medication adherence among gout patients treated with allopurinol. This study was conducted in the **Clinical Practice Research Datalink (CPRD)** from the UK, formerly known as the General Practitioner Database. The database comprises prospectively collected computerized medical records from general practitioners. Since 1987, data are prospectively recorded, and include patient demographics, prescription details, clinical events, laboratory test results, specialist referrals, and hospitalizations. Data of over 11.3 million patients from 674 practices from the UK is available.[94] For this study, newly diagnosed gout patients initiated with allopurinol in the period of valid data collection (1st January 1987 through 30th of June 2014), were included. Longitudinal data of 48,834 newly diagnosed gout patients initiated allopurinol were used to assess medication adherence, including non-adherence, non-persistence, and those who return to therapy after the occurrence of a gap in therapy and their subsequent adherence to medication. Additionally, the association of various factors with medication adherence was evaluated.

In c*hapter 6,* we performed a **systematic literature review and meta-analysis** on medication adherence among gout patients. Furthermore, we investigated which factors were associated with medication adherence.

In *chapter 7,* we explored knowledge, illness perceptions, and stated practice behaviour in relation to gout in primary care. For this study we collected data by **interviewing 32 general practitioners** from the southern part of The Netherlands. A mixed-methods approach, using questionnaires and structured interviews, was used.

In *chapter 8*, we discuss the main results and methodologic issues of the studies described in the earlier chapters, and implications for further research and clinical practice are given.

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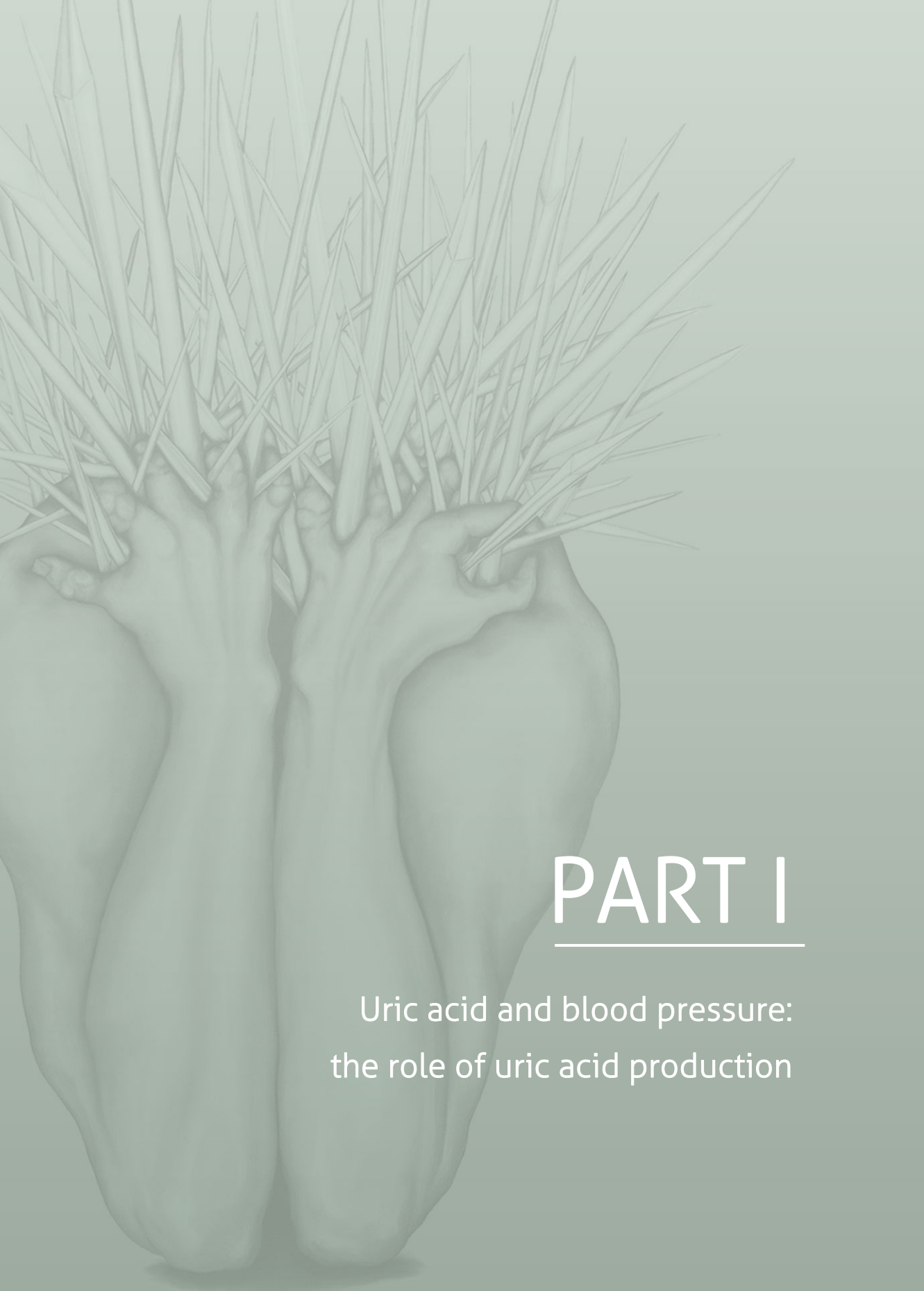

PART I

Uric acid and blood pressure: the role of uric acid production

2

Xanthine oxidase gene variants and their association with blood pressure and incident hypertension: a population study

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ABSTRACT

Objective

The enzyme xanthine oxidoreductase (XOR) generates uric acid in the terminal steps of the purine metabolism; meanwhile reactive oxygen species are formed. We hypothesized that uric acid production, as assessed indirectly from *XOR* variants, is associated with hypertension.

Methods

Among 2769 participants (48.3% men; mean age 40.7 years) randomly recruited from European populations, we genotyped 25 tagging *XOR* SNPs and measured blood pressure (BP) at baseline and follow-up (median 8.8 years). The relation between variants of the *XOR* gene with changes in pulse pressure and mean arterial pressure over time; and incidence of hypertension, were analyzed.

Results

Compared with nonminor allele carriers, pulse pressure increased approximately 2 mmHg more in minor allele carriers of rs11904439 (P=0.01), whereas mean arterial pressure and DBP increased approximately 1 mmHg less in minor allele carriers of rs2043013 (P=0.01). In 2050, participants normotensive at baseline, hazard ratios contrasting risk of hypertension in minor allele carriers vs. nonminor allele carriers were 1.31 (95% confidence interval 1.03–1.68; P=0.02) and 1.69 (95% confidence interval 1.11–2.57; P=0.01) for rs11904439 and rs148756340, respectively. With the false discovery rate set at 0.25, the aforementioned associations retained significance. The changes in SBP from baseline to follow-up and the serum levels of uric acid at baseline (n=1949) were not associated with *XOR*.

Conclusion

Pending confirmation, our findings suggest that variation in uric acid production, as captured by genetic variation in *XOR*, might be a predictor of changes in BP and in the risk of hypertension.

INTRODUCTION

Cardiovascular disease is the leading cause of death worldwide and is an important contributor to years lived with disability.[1,2] Hypertension is the most potent risk factor for cardiovascular disease. Globally, approximately 25–35% of the adult population and more than 60% of the individuals over the age of 65 years have high blood pressure (BP).[3]

BP and hypertension are associated with serum uric acid, the end product of purine catabolism, but whether this association is causal remains controversial.[4] Some experts propose uric acid as a mere risk marker, because its serum concentration is often elevated in conditions other than hypertension, such as obesity or the metabolic syndrome. On the other hand, several studies suggested plausible mechanism by which uric acid may be causally linked to elevated BP and the development of hypertension.[5]

So far, epidemiological studies focused on the concentrations of uric acid in the serum. However, serum uric acid concentration is mainly determined by uric acid 'excretion' and not by its 'production'.[6] During the last steps of uric acid production, xanthine oxidoreductase (XOR) catalyzes hypoxanthine to xanthine and xanthine to uric acid, thereby producing reactive oxygen species that inactivates the vasodilator nitric oxide (Figure 1).[5] In this study, we hypothesized that not hyperuricemia as such, but increased production of uric acid by XOR elevates BP and leads to hypertension, as a consequence of oxidative stress, reduced availability of nitric oxide, and endothelial dysfunction. As we cannot measure uric acid production directly in a large cohort of healthy humans, we used variation in the *XOR* gene as a proxy. Several studies showed association of XOR expression with variation in the gene, and therefore, support this research approach.[7,8]

This study aimed to explore the role of *XOR* as a hypertension susceptibility gene by using 25 tagging single nucleotide polymorphisms (SNPs) in the *XOR* gene and by relating them to changes in BP over time and to the incidence of hypertension in a European population study.

PATIENTS AND METHODS

Study population

Recruitment for the Flemish Study on Environment, Genes, and Health Outcomes (FLEMENGHO) started in 1985.[9]

From August 1985 to November 1990, a random sample of the households living in a geographically defined area of northern Belgium was investigated with the goal to recruit an equal number of participants in each of six subgroups by sex and age (20-39, 40-59, and ≥60 years). From June 1996 until January 2004, recruitment of families continued using the former participants (1985-1990) as index persons and also, including teenagers.[9] Each participant underwent up to seven follow-up examinations. In

all study phases, we used the same standardized methods to measure clinical and biochemical variables, administer questionnaires, and to determine incidence of fatal and nonfatal outcomes.[10] The European Project on Genes in Hypertension (EPOGH) recruited participants from 1999 to 2001 in four centers: Pilsen (Czech Republic), Mirano (Italy), Kraków (Poland), and Novosibirsk (Russian Federation).[11] The EPOGH investigators received training at the Studies Coordinating Centre in Leuven, Belgium, and applied the same protocol, questionnaires, and follow-up procedures, as used in FLEMENGHO.[11] In EPOGH, one follow-up examination took place from 2006 to 2008 and in FLEMENGHO, follow-up lasted until 2013. Both studies were conducted according to the principles outlined in the Helsinki declaration for investigation of human participants.[12] Each local Institutional Review Board approved the study protocol. Participants provided written informed consent.

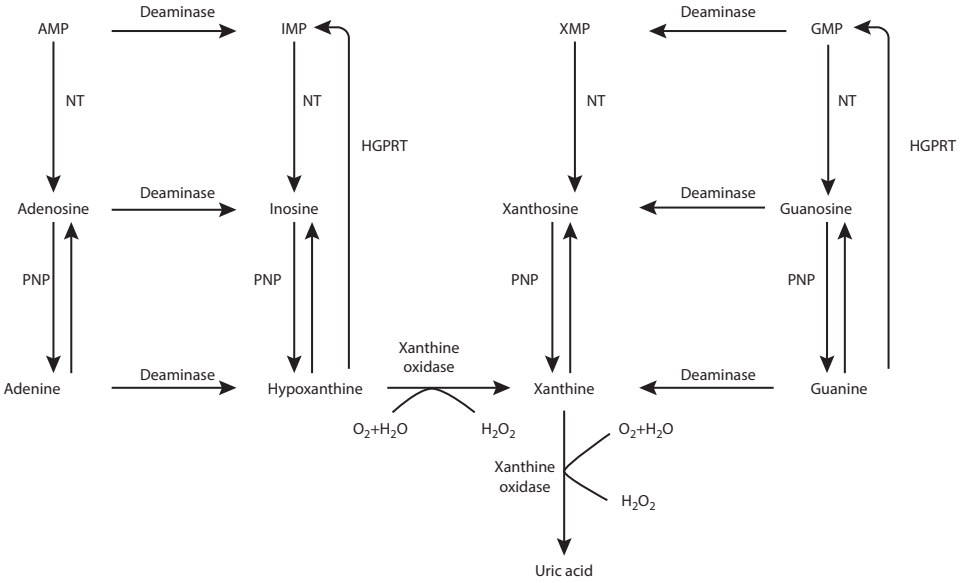

Figure 1. Purine metabolism. During the final stage of purine metabolism, xanthine oxidoreductase catalyzes hypoxanthine to xanthine, and xanthine to uric acid whereas reactive oxygen species is formed. These reactive oxygen species inactivate the vasodilator nitric oxide. HGPRT, hypoxanthine-guanine phosphoribosyltransferase; IMP, inosine monophosphate; NT, nucleotidase; PNP, purine nucleoside phosphorylase; XMP, Xanthosine monophosphate

Definition of cohorts

As shown in the flow chart (Figure 2), 3343 participants took place in FLEMENGHO and 1192 in EPOGH. Participants were excluded from analysis, if their BP measurements were missing at baseline (*n*=61) or because *XOR* had not been genotyped (*n*=1307)

because of exhaustion of the biobank or DNA degradation. Of the remaining 3167 participants, 356 were not followed up and 42 had died. The 'blood pressure cohort' included 2769 participants to study the association between change in BP and genotype. The 'hypertension cohort' consisted of 2050 participants, who at baseline were normotensive and not on antihypertensive drug treatment.

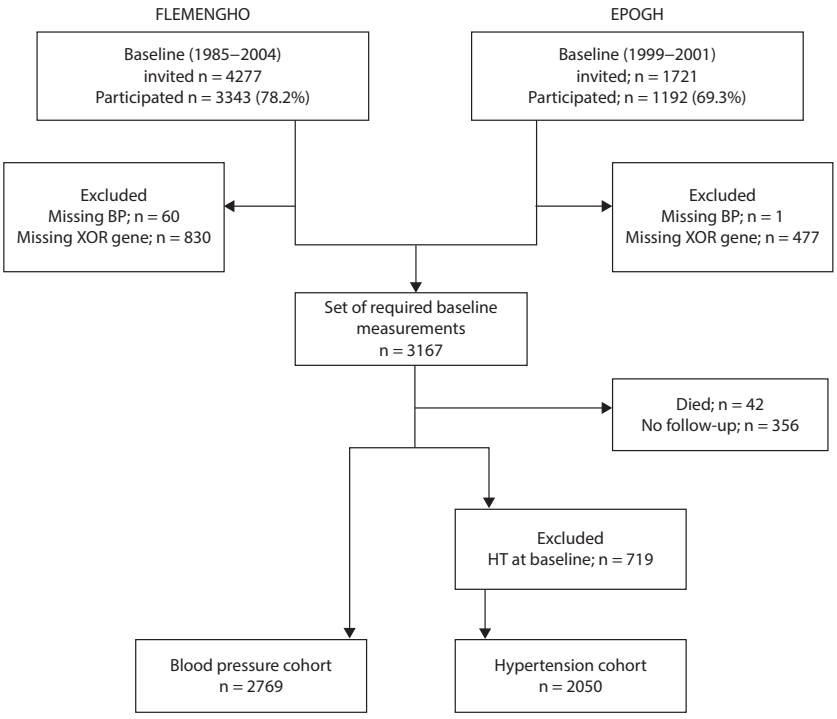

Figure 2. Flowchart of participants. The blood pressure cohort and hypertension cohort refer to participants used to study the association with change in BP and the incidence of HT over follow-up. BP, blood pressure; EPOGH, European Project on Genes in Hypertension; FLEMENGHO, Flemish Study on Environment, Genes, and Health Outcomes; HT, hypertension; XOR, xanthine oxidoreductase

Measurements

Experienced observers measured each participant's BP at baseline and follow-up by auscultation of the Korotkoff sounds. After the participant had rested for 5 min in the sitting position, they obtained five consecutive BP readings (phase V DBP) to the nearest 2 mmHg, using mercury sphygmomanometers. Standard cuffs had a 12x24 cm inflatable portion, but if upper arm girth exceeded 31 cm, larger cuffs with 15x35 bladders were used. Previous publications describe the quality control of the BP measurements.[11,13]

For analysis, the five BP readings were averaged. Hypertension was a BP equal to or exceeding 140 mmHg SBP or 90 mmHg DBP or use of antihypertensive drugs. We analyzed the pulsatile and steady component of BP as captured by pulse pressure (PP) and mean arterial pressure (MAP), respectively. PP is the difference between SBP and DBP. MAP is DBP and one-third of PP. Combining PP and MAP is superior in addressing the relative contributions of both arterial stiffness and peripheral arterial resistance to cardiovascular risk.[14] BP changes over time were calculated as the last follow-up minus the baseline measurement. Trained nurses measured anthropometric characteristics and administered the same questionnaire at baseline and follow-up to collect each participant's medical history, smoking status, drinking habits, and use of medications. BMI was weight in kg/m^2. Participants fasted for at least 6 h prior to venous blood sampling.

Genotyping, imputation, and single-nucleotide polymorphisms selection

The FLEMENGHO and EPOGH participants were genotyped at the University of Milan as part of the HYPERGENES project[15] (European Network for Genetic-Epidemiological Studies; www.hypergenes.eu), using the 1M Duo chip, the Metabochip and/or a 15K custom chip (Illumina Inc., San Diego, California, USA).[15] Imputation was performed using Minimac[16] and 1000 Genomes haplotypes as reference (release March 2012). With the 1M Duo chip, 87 *XOR* SNPs for 507 individuals were genotyped, six *XOR* SNPs for 994 individuals were genotyped with the Metabochip, and 20 *XOR* SNPs for 2742 individuals were genotyped with the 15K custom chip. For each sample, 1299 SNPs were imputed, but only SNPs with an imputation quality of r^2 at least 80% were used in the association analysis. This resulted in 300 imputed SNPs for the 1M Duo array, none for the Metabochip, and 76 SNPs for the 15K.

XOR (80 423 base pairs) maps to chromosome 2 (2p23.1,2 :31,557,187–31,637,611) (Figure Supplement 1 (S1)). To select the *XOR* SNPs to be included in the analyses, we first reviewed all SNPs in this gene, including the flanking regions (±20 kb from the gene). We excluded SNPs with a minor allele frequency of less than 1% or a genotyping call rate less than 0.99. We selected 28 tagging SNPs (rs1054889, rs207440, rs9308919, rs1896846, rs12621192, rs732436, rs2281548, rs17011368, rs2043013, rs10181969, rs11900892, rs45622435, rs11894627, rs45449798, rs45520334, rs3769618, rs4407290, rs2236168, rs148756340, rs561525, rs11904439, rs206857, rs10175754, rs206800, rs206803, rs3769616, rs206804, and rs206811) that were in high linkage disequilibrium (r^2≥0.80) with 77 neighboring SNPs, but were not in high linkage disequilibrium (r^2≥0.80) with each other (Supplemental Table S1). The 28 selected SNPs covered the entire gene with extension into the 3' and 5'-flanking regions. For analyses, the minor alleles were determined by using the European haplotype map population as reference (www.ncbi.nlm.nih.gov/snp).

Statistical analysis

For statistical analysis, we used SAS software version 9.3 (SAS Institute Inc., Cary, North Carolina, USA). We tested Hardy–Weinberg equilibrium in 1222 unrelated individuals, by using the exact statistics available in the PROC ALLELE procedure of the SAS package. We applied mixed models to investigate the association between BP changes over time and genotype, while accounting for family clusters and study center (FLEMENGHO or EPOGH) as random effects and adjusting for sex, age, BMI, BP at baseline, smoking and drinking alcohol, use of female sex hormones (all at baseline), duration of follow-up, and three indicator variables coding for starting or stopping antihypertensive drug treatment from baseline to follow-up or remaining on treatment. In 1949, participants whose serum uric acid concentration at baseline was known, we also examined its association with the genetic variants.

Next, we modelled the risk of hypertension in relation to the genetic variants by Cox proportional hazard regression analysis, as implemented in the PROC SURVIVAL procedure of the SUDAAN software (Research Triangle Institute, Research Triangle Park, North Carolina, USA) version 10.01. We censored study participants from further analysis after occurrence of the first diagnosis of hypertension. We checked the proportional hazards assumptions by the Kolmogorov-Smirnov supremum test. Hazard ratios were calculated for participants of the 'hypertension cohort,' while accounting for family clusters and study center as random effects. We additionally adjusted for sex, age, BMI, SBP and DBP, smoking and drinking alcohol, and use of female sex hormones (all at baseline).

In phenotype–genotype analyses, we contrasted minor allele carriers with nonminor allele carriers and we adjusted significance levels for multiple testing by Benjamini–Hochberg's approach with the false discovery rate set at 0.25.[17]

RESULTS

The baseline characteristics of the study population by study center are shown in Table 1. The 'blood pressure cohort' included 2769 participants, of whom 1337 (48.3%) were men. At baseline, participants were on average (±SD) 40.7±15.2 years old, and 719 had hypertension (26.0%), of whom 322 (44.8%) were on antihypertensive drug treatment. The 'hypertension cohort' included 2050 participants, of whom 969 (47.3%) were men. Their mean age at baseline was 37.3±14.3 years. All participants were white Europeans.

Table 1. Baseline characteristics of participants by cohort

	Blood pressure cohort	Hypertension cohort
Number of participants (*n*)	2769	2050
Median follow-up years (IQR)	8.8 (6.7– 12.8)	9.1 (6.7 – 14.0)
Number (%) with characteristics		
FLEMENGHO cohort	2069 (75.7)	1631 (79.6)
Men	1337 (48.3)	969 (47.3)
Hypertension	719 (26.0)	0 (0.0)
Antihypertensive drug use	322 (11.6)	0 (0.0)
Use of female sex hormones	261 (9.4)	230 (11.2)
Current smoking	780 (28.2)	610 (29.8)
Alcohol consumption	930 (33.6)	659 (32.2)
Uric acid lowering therapy	17 (0.6)	5 (0.2)
Mean (SD) of characteristic		
Age (years)	40.7 (15.2)	37.3 (14.3)
BMI ($kg/m^{2)}$	25.2 (4.6)	24.3 (4.1)
SBP (mmHg)	125.0 (17.0)	118.1 (10.5)
DBP (mmHg)	77.0 (11.1)	73.1 (8.3)
Serum uric acid (mmol/L)[a]	293 (84)	286 (81)

Blood pressure cohort and hypertension cohort refer to the participants used to study the changes in blood pressure and the incidence of hypertension over follow-up. Blood pressure was the average of five consecutive readings at a single visit. Hypertension is defined as untreated blood pressure of at least 140 mmHg SBP, or 90 mmHg DBP, or use of antihypertensive drugs. IQR, interquartile range; FLEMENGHO, Flemish Study on Environment, Genes, and Health Outcomes. [a]No data available on serum uric acid concentrations for 820 (29.6%) participants of the blood pressure cohort and 634 (30.9%) participants of the hypertension cohort.

Genotypes

Of the 28 selected SNPs, 25 complied with Hardy–Weinberg equilibrium (*P*-value >0.10) and were included in the analyses (Supplemental Table S2). The frequencies of the minor alleles ranged from 1.7% for rs148756340 and rs455203344 to 48.6% for rs1054889.

Longitudinal changes in mean arterial pressure and pulse pressure

Follow-up of BP was available at one, two, or at least three occasions for 2769, 1450, and 1035 participants, respectively. PP changes (follow-up minus baseline) over a median of 8.8 years (interquartile range, 6.7-12.8) in minor allele carriers ranged from 1.7 mmHg for rs4407290 to 5.0 mmHg for rs11904439 (Supplemental Table S3) In nonminor allele carriers, this range encompassed 1.9 mmHg for rs11904439 to 2.3 mmHg for rs732436. In multivariable-adjusted analyses, a significant difference in the PP change over time (nonminor allele carriers minus minor allele carriers) was associated with rs11904439 [2.09 mmHg; 95% confidence interval (CI), 0.43-3.75 mmHg] (Supplemental Table S3).

Furthermore, MAP changes in minor allele carriers ranged from 2.8 mmHg for rs148756340 to 4.9 mmHg for rs206803 (Supplemental Table S3). In nonminor allele carriers, the corresponding range encompassed 3.9 mmHg for rs2236168 to 4.8 mmHg for rs2043013. In multivariable-adjusted analyses, a significant difference in MAP changes (nonminor allele carriers minus minor allele carriers) was observed for

rs12621192 (-0.78 mmHg; CI, 1.52 to -0.04 mmHg; *P*=0.04) and rs2043013 ('0.99 mmHg; CI, -1.77 to -0.20 mmHg; *P*=0.01). The MAP results for rs2043013 depended on a lesser increase in DBP in minor allele carriers compared with nonminor allele carriers (-1.06 mmHg; CI, -1.82 to -0.30 mmHg; *P*-value <0.01) (Supplemental Table S3).
With Benjamini–Hochberg's correction for multiple testing applied, the aforementioned associations of PP with rs11904439 and of MAP and DBP with rs2043013 retained significance. Even without Benjamini–Hochberg's correction, none of the associations of changes in SBP or the serum levels of uric acid at baseline (*n*=1949) with *XOR* variants was significant (*P*-value ≥0.05).

Incidence of hypertension

Follow-up for incident hypertension was available at one, two, or at least three occasions in 2050, 1119, and 805 participants, respectively. The median number of follow-up years was 9.1 years (interquartile range, 6.7-14.0). In the entire 'hypertension cohort', 753 (36.4%) participants who were normotensive at baseline developed hypertension. Of these, 230 (30.8%) were on antihypertensive drug treatment. In the remaining 517 patients (69.2%), the diagnosis of hypertension relied on thresholds being exceeded for SBP (*n*=188; 25.0%), DBP (*n*=186; 24.7%), or both (*n*=149; 19.8%).
The hazard ratios contrasting the risk of hypertension in minor allele carriers vs. nonminor allele carriers ranged from 0.90 (CI, 0.77-1.06; *P*=0.21) for rs9308919 to 1.31 (CI, 1.03-1.68; *P*=0.02) and to 1.69 (CI, 1.11-2.57; *P*=0.01) for rs11904439 and rs148756340, respectively (Figure 3). With Benjamini–Hochberg's correction for multiple testing applied, the hazard ratios remain significant for rs11904439 and rs148756340 (Supplemental Table S4).

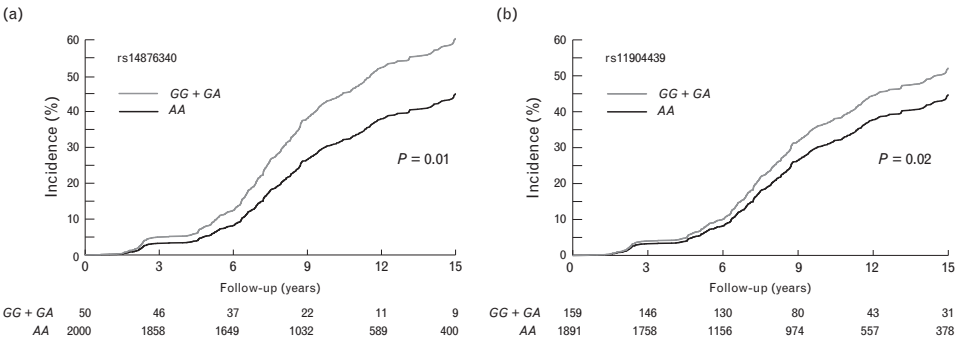

Figure 3. Cumulative incidence plots. Sex and age-standardized cumulative incidence of hypertension in relation to rs148756340 (a) and rs11904439 (b) XOR genotypes. XOR, xanthine oxidoreductase.

DISCUSSION

In the present study, using genetic variation in *XOR* as a proxy, we tested the hypothesis that variation in the uric acid production might be associated with secular trends in the steady and pulsatile BP components and the risk of hypertension. Accepting a false discovery rate of 0.25, the increase in PP and MAP and the risk of hypertension over 8-9 years of follow-up (median) were associated with three of 25 SNPs in *XOR*. Compared with nonminor allele carriers, PP increased by approximately 2 mmHg more in minor allele carriers of rs11904439, whereas MAP and DBP increased approximately 1 mmHg less in minor allele carriers of rs2043013. The risk of hypertension was 30-70% higher in minor allele carriers of rs11904439 and rs148756340, respectively. The XOR enzyme is responsible for the two final steps of purine degradation, at which hypoxanthine is converted to xanthine and xanthine to uric acid (Figure 1). The present study therefore lends some support for the hypothesis underlying its rational.

It is generally accepted that an association between uric acid and BP exists, and Johnson and coworkers[18] have been key investigators in studying this association. They showed in animal models[5] and observational studies[19] that elevated uric acid concentrations are associated with elevated BP. The meta-analysis performed by Grayson et al.[20] in 2011, which included data from 57,607 participants of 18 prospective cohort studies, has shown that hyperuricemia is associated with an increased risk for incident hypertension, independent of traditional hypertension risk factors (adjusted relative risk ratio 1.41; CI, 1.23–1.58). Furthermore, for a 1 mg/dL increase in uric acid concentrations, the pooled adjusted relative risk for incident hypertension was 1.13 (CI, 1.06–1.20).[20]

Despite this body of evidence, Mendelian randomization studies aiming to establish causality have not been able to convincingly demonstrate that the association between uric acid and BP is causal. Only three[21–23] out of eight studies[21–28] showed a significant association between polymorphisms of the *SLC2A9* gene, a robust proxy for uric acid concentrations, and BP. Furthermore, a meta-analysis of randomized controlled trials to assess the efficacy of the XOR inhibitor allopurinol on BP showed that allopurinol treatment decreases SBP by 3.3 mmHg (CI, 1.4–5.3 mmHg) and DBP by 1.3 mmHg (CI, 0.1-2.5 mmHg).[29] Allopurinol furthermore improved endothelial-dependent vasodilation, whereas the uricosuric drug probenecid did not.[30] These findings further suggest that next to uric acid concentrations, the role of uric acid production by XOR should be considered as a determinant of BP.

XOR is expressed in many tissues but its activity is strongest in the liver and the gut.[31,32] Individual differences in the activity of human liver XOR exist. XOR activity depends on sex, age, and ethnicity. Men show stronger hepatic XOR activity than women do and the enzymatic activity increases with advancing age.[33–35]

So far, genetic variation in the *XOR* gene has not been studied extensively.[7,8,36–39] Most of the available studies focused on genetic variants that cause xanthine oxidase deficiency, also known as the rare disease xanthinuria type I.[36–39] Two in-vitro studies from Kudo et al.[7,8] addressed the impact of variation in the *XOR* gene on xanthine

oxidase activity. Molecular studies of the DNA of 96 Japanese, revealed three nonsynonymous SNPs. Functional characterization of 21 *XOR* variants revealed deficient enzyme activity in two, and reduced and enhanced activity in six and two, respectively.[7] Among 13 SNPs in the *XOR* 5'-flanking region, one was associated with decreased expression in transfected cells.[8] Both studies examined polymorphisms of the *XOR* gene in a Japanese population and as its activity might be influenced by ethnicity, caution is required when applying these results to a European study population.[7,8] The SNPs investigated in the Japanese studies were not determined and/or in linkage disequilibrium with the SNPs from the current study.

So far, only two candidate gene studies investigated the association between BP and *XOR* variants.[40,41] The cross-sectional study from Yang et al.[40] investigated the genetic variation in the promotor and all exon regions of the *XOR* gene in 48 randomly chosen Japanese patients with hypertension. They identified three missense mutations (G172R, A932T, and N1109T) in a heterozygous state in addition to 34 variations, including 15 common SNPs in 48 hypertensive patients. Additionally, 953 hypertensive patients and 1818 patients from the general Japanese population were genotyped for three missense mutations and eight common SNPs. Four hypertensive patients with rare missense mutations (G172R or N1109T) in homozygous form had resistant hypertension, despite antihypertensive drug therapy. Multivariable adjusted logistic regression showed a significant association of three SNPs with hypertension in men: 47686C>T [rs229475; odds ratio (OR), 1.52; *P*=0.047] and 69901A>C (intron 31; OR, 3.14; *P*=0.039) in the recessive model and 67873A>C (N1109T; rs45547640; OR, 1.84; *P*=0.018) in the dominants model. SNP rs2295475 was also present in our dataset and in high linkage disequilibrium with SNP rs12621192. The latter was retained for analysis but was not associated with any of the outcomes considered. As our study included white Europeans and was longitudinal instead of cross-sectional, caution is required by comparing our result with the previous studies.[40]

The genome-wide association studies (GWAS) published so far on SBP, DBP, and hypertension,[42-45] and on PP and MAP[46-48] identified numerous novel loci, but none of these were located near the *XOR* gene. Cross-sectional GWAS studies offer the opportunity for searching for associations between BP and densely distributed SNPs across the whole genome in large numbers of study participants. Such studies require significance levels of 10^{-6} to 10^{-9}. In contrast, our study was prospective and population based, involving only 25 common tagging SNPs in *XOR* gene. We did, therefore, not rely on such extreme *P* values, but applied the Benjamin–Hochberg's approach for multiple testing. Admittedly, our sample size was smaller than in GWAS studies. As the functionality of the SNPs is unknown, caution is required by interpreting the results and the present findings can only be considered as hypothesis generating. Future population-based research projects might address this tissue.

The prospective study design, genotyping of 25 common tagging SNPs in *XOR* gene, the extensive and reliable phenotyping of BP, and adjustment for potential confounders are major strengths of our study. Nevertheless, our current study must be interpreted within

the context of its limitations. First, the functionality of the tagging SNPs analyzed in our current study remains unknown. Second, we accepted a false discovery rate of 0.25, so that the chance of rejecting a true-positive SNP is low, however, 25% of the significant results might be wrong. Therefore, further investigation is required to confirm our findings. Third, our sample size was relatively small (*n*=2796) and might have been insufficient to identify SNPs associated with BP in a reliable manner. However, if a larger sample would be required, it follows that the effects on BP must be minute. Finally, our current findings are probably representative for white Europeans, but should not be extrapolated to other ethnic groups or people with a different lifestyle with possible epigenetic impact on gene expression or functionality.

In conclusion, we demonstrated that BP rise over time and/or the risk of hypertension are associated with three SNP in the *XOR* gene: rs2043013, rs11904439, and rs148756340. Given the burden of chronic hypertension and the fact that only 37% of the patients obtain a well managed BP,[49] a better understanding of these associations should be subject to further experimental, clinical, and epidemiologic research. Further experimental research is required to examine whether the identified SNPs from the present study alter enzyme activity. Previous studies have shown that genetic variation in other genes, even if this is in the intron, can influence gene regulation. For example, by altering transcription activity[50] or the role of RNAs derived from introns.[51,52] Moreover, our current findings need replication in other longitudinal studies in patients and populations. In view of the role of circulating uric acid as a marker of the risk of death and cardiovascular complications,[53] further studies should also address the association between incidence cardiovascular disease and variation in the *XOR* gene. Pending confirmation, the rs2043013, rs11904439, and rs148756340 polymorphisms might then be used to identify overproducers of uric acid; and combined with other genetic markers might contribute to the stratification of cardiovascular risk.

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SUPPLEMENTAL MATERIAL

Table S1. 25 Common Tagger SNPs in *XOR* Gene that are in High Linkage Disequilibrium ($r^2 \geq 0.80$) with 77 Other SNPs

SNP ID	Position (Base-Pair)	Location	MAF	Tagged SNP
rs1054889	31557308	3utr	0.48	rs1042039, rs2268800, rs10490361, rs932559, rs736492, rs207429, rs12614330, rs169596
rs207440	31562412	coding	0.05	rs207445
rs9308919	31568726	intronic	0.20	rs4952083, rs7598968, rs7572658, rs7599175, rs1366811, rs1366813, rs4952084, rs4952085, rs1884725, rs13415401, rs7557095
rs12621192	31588664	intronic	0.33	rs2295475, rs761926
rs732436	31590156	intronic	0.24	rs2365842, rs1429376
rs17011368	31590917	coding	0.03	rs45565237, rs17395161, rs17323225, rs45443498, rs115074795, rs17395224, rs45449798, rs114281245, rs45550432, rs45457594, rs116648715
rs2043013	31592323	intronic	0.43	rs1366817, rs2070293
rs10181969	31593125	intronic	0.03	-
rs11900892	31594524	intronic	0.03	-
rs13398137	31594789	intronic	0.06	rs45596941, rs45445692, rs7597755
rs11894627	31597410	intronic	0.44	rs4952088, rs4952087, rs4952086, rs4952089, rs6720163, rs1864280
rs45449798	31598810	intronic	0.02	rs17395224, rs114281245, rs115074795, rs45443498, rs17323225, rs45550432, rs45457594, rs116648715, rs17395161, rs17011368, rs45565237
rs45520334	31602147	intronic	0.02	-
rs3769618	31605447	intronic	0.35	rs4952089, rs2281547
rs4407290	31606670	coding	0.03	-
rs2236168	31607128	intronic	0.46	-
rs148756340	31607275	intronic	0.01	-
rs561525	31608855	intronic	0.02	rs473935, rs528551, rs206856
rs11904439	31614900	intronic	0.04	-
rs206857	31619101	intronic	0.16	rs13418515
rs10175754	31621598	intronic	0.13	rs13431382, rs10199855, rs11895133, rs13418515
rs206800	31627947	intronic	0.20	-
rs206803	31629030	intronic	0.14	rs206798, rs494852, rs1366814, rs6759319, rs114978564
rs3769616	31630382	intronic	0.03	rs111749408, rs76333723
rs206804	31630846	intronic	0.16	rs206805, rs206798, rs494852

SNP, single-nucleotide polymorphism; MAF: minor allele frequency. SNP ID is a GenBank ID number (NCBI). Position and location type were taken from the most recent human genome sequence assemblies (NCBI Build 37.3). MAFs were calculated in the present population.

Table S2. Allele and genotype frequencies in 1222 unrelated individuals to test the Hardy–Weinberg equilibrium

SNP	Minor allele, n (%)		Major allele, n (%)		Genotype, n (%)						*P*
rs1054889	A	1187 (48.6)	G	1257 (51.4)	AA	280 (22.9)	AG	627 (51.3)	GG	315 (25.8)	0.34
rs207440	A	123 (5.0)	G	2321 (95.0)	AA	3 (0.3)	AG	117 (9.6)	GG	1102 (90.2)	0.95
rs9308919	T	492 (20.1)	C	1952 (79.9)	TT	57 (4.7)	CT	378 (30.9)	CC	787 (64.4)	0.18
rs1896846	C	604 (24.7)	G	1840 (75.3)	CC	87 (7.1)	CG	430 (35.2)	GG	705 (57.7)	0.06*
rs12621192	T	821 (33.6)	T	1623 (66.4)	TT	149 (12.2)	CT	523 (42.8)	CC	550 (45.0)	0.15
rs732436	A	654 (26.8)	G	1790 (73.2)	AA	84 (6.9)	AG	486 (39.8)	GG	652 (53.4)	0.61
rs2281548	G	785 (32.1)	A	1659 (67.9)	GG	149 (12.2)	AG	487 (39.9)	AA	586 (48.0)	<0.001*
rs17011368	C	86 (3.5)	T	2358 (96.5)	CC	1 (0.1)	CT	84 (6.9)	TT	1137 (93.0)	0.67
rs2043013	C	1067 (43.7)	A	1377 (56.3)	CC	246 (20.1)	AC	575 (47.1)	AA	401 (32.8)	0.13
rs10181969	C	84 (3.4)	T	2360 (96.6)	CC	1 (0.1)	CT	82 (6.7)	TT	1139 (93.2)	0.70
rs11900892	A	172 (7.0)	G	2272 (93.0)	AA	4 (0.3)	AG	164 (13.4)	GG	1054 (86.3)	0.37
rs13398137	C	184 (7.5)	A	2260 (92.5)	CC	4 (0.3)	AC	176 (14.4)	AA	1042 (85.3)	0.23
rs11894627	G	1100 (45.0)	A	1344 (55.0)	GG	236 (19.3)	AG	628 (51.4)	AA	358 (29.3)	0.18
rs45449798	C	56 (2.3)	T	2388 (97.7)	CT	56 (4.6)	TT	1166 (95.4)			0.41
rs45520334	A	42 (1.7)	G	2402 (98.3)	AG	42 (3.4)	GG	1180 (96.6)			0.54
rs3769618	T	868 (35.6)	C	1572 (64.4)	TT	162 (13.3)	CT	544 (44.6)	CC	514 (42.1)	0.34
rs4407290	A	84 (3.4)	G	2360 (96.6)	AA	1 (0.1)	AG	82 (6.7)	GG	1139 (93.2)	0.70
rs2236168	C	1096 (44.9)	T	1344 (55.1)	CC	247 (20.3)	CT	602 (49.3)	TT	371 (30.4)	0.92
rs148756340	G	42 (1.7)	A	2402 (98.3)	AG	42 (3.4)	AA	1180 (96.6)			0.54
rs561525	C	43 (1.8)	T	2401 (98.2)	CC	1 (0.1)	CT	41 (3.4)	TT	1180 (96.6)	0.30
rs11904439	G	109 (4.5)	A	2333 (95.5)	AG	109 (8.9)	AA	1112 (91.1)			0.10
rs206857	A	480 (19.7)	G	1962 (80.3)	AA	43 (3.5)	AG	394 (32.3)	GG	784 (64.2)	0.44
rs10175754	C	422 (17.3)	T	2018 (82.7)	CC	35 (2.9)	CT	352 (28.9)	TT	833 (68.3)	0.77
rs206800	G	502 (20.5)	A	1942 (79.5)	GG	55 (4.5)	AG	392 (32.1)	AA	775 (63.4)	0.55
rs206803	C	391 (16.0)	A	2053 (84.0)	CC	29 (2.4)	AC	333 (27.3)	AA	860 (70.4)	0.63
rs3769616	T	117 (4.8)	C	2327 (95.2)	TT	2 (0.2)	CT	113 (9.3)	CC	1107 (90.6)	0.62
rs206804	T	544 (22.3)	C	1900 (77.7)	TT	62 (5.1)	CT	420 (34.4)	CC	740 (60.6)	0.81
rs206811	A	890 (36.4)	G	1554 (63.6)	AA	180 (14.7)	AG	530 (43.4)	GG	512 (41.9)	0.03*

Values indicate number of alleles and genotypes (%). *P*-values test departure from the Hardy-Weinberg equilibrium.*Excluded for analyses, because Hardy–Weinberg criteria were unmet (*P*<0.10).

Table S3. Longitudinal changes in pulse pressure and mean arterial pressure according to variation in the XOR gene (Blood Pressure Cohort, n=2769)

SNP	Minor allele	Non-minor allele	Mean Change in Pulse Pressure				Mean Change in Mean Arterial Pressure			
			Minor allele carriers	Non-minor allele carriers	Adjusted difference mm Hg (95% CI)	*P*	Minor allele carriers	Non-minor allele carriers	Adjusted difference mm Hg (95% CI)	*P*
rs1054889	*AA + AG*	*GG*	2.2 (13.7)	2.0 (13.9)	-0.22 (-1.22 to 0.78)	0.67	4.2 (11.8)	4.5 (11.6)	-0.15 (-0.97 to 0.67)	0.73
rs207440	*AA + AG*	*GG*	3.4 (13.8)	2.0 (13.7)	0.93 (-0.57 to 2.43)	0.22	4.5 (11.5)	4.3 (11.8)	0.41 (-0.82 to 1.64)	0.51
rs9308919	*TT + TC*	*CC*	2.2 (13.6)	2.1 (13.8)	-0.35 (-1.27 to 0.57)	0.46	4.1 (11.6)	4.4 (11.9)	-0.32 (-1.08 to 0.43)	0.40
rs12621192	*TT + TC*	*CC*	2.2 (13.7)	2.1 (13.8)	0.26 (-0.65 to 1.16)	0.58	4.1 (11.9)	4.5 (11.6)	-0.78 (-1.52 to -0.04)	0.04
rs732436	*AA + AG*	*GG*	2.1 (13.7)	2.3 (13.9)	-0.25 (-1.14 to 0.64)	0.58	4.1 (11.8)	4.5 (11.8)	-0.34 (-1.04 to 0.43)	0.42
rs17011368	*CC + CT*	*TT*	2.9 (14.6)	2.1 (13.7)	0.52 (-1.33 to 2.36)	0.58	3.9 (11.4)	4.3 (11.8)	-0.29 (-1.80 to 1.21)	0.70
rs2043013	*CC + CA*	*AA*	2.1 (13.7)	2.2 (13.8)	0.23 (-0.73 to 1.18)	0.64	4.0 (11.7)	4.8 (11.8)	-0.99 (-1.77 to -0.20)	0.01*
rs10181969	*CC + CT*	*TT*	1.8 (13.7)	2.2 (13.8)	-0.18 (-1.98 to 1.62)	0.84	4.6 (11.3)	4.3 (11.8)	0.52 (-0.98 to 2.01)	0.49
rs11900892	*AA + AG*	*GG*	2.1 (14.2)	2.2 (13.7)	-0.27 (-1.59 to 1.06)	0.69	3.8 (11.6)	4.4 (11.8)	-0.35 (-1.44 to 0.74)	0.53
rs13398137	*CC + CA*	*AA*	1.7 (13.8)	2.2 (13.7)	-0.39 (-1.66 to 0.89)	0.55	3.9 (11.2)	4.3 (11.9)	-0.28 (-1.34 to 0.77)	0.60
rs11894627	*AA + GA*	*GG*	2.2 (13.8)	2.1 (13.7)	0.03 (-1.15 to 1.20)	0.96	4.3 (11.8)	4.2 (11.4)	-0.60 (-1.57 to 0.37)	0.22
rs45449798	*CC + CT*	*TT*	4.2 (14.6)	2.1 (13.7)	1.92 (-0.38 to 4.22)	0.10	3.4 (12.1)	4.3 (11.8)	-0.42 (-2.31 to 1.47)	0.66
rs45520334	*AG*	*GG*	3.7 (15.7)	2.1 (13.6)	0.91 (-1.68 to 3.49)	0.48	3.8 (13.2)	4.3 (11.7)	-0.03 (-2.14 to 2.09)	0.98
rs3769618	*TT + TC*	*CC*	2.3 (13.9)	2.0 (13.6)	0.24 (-0.67 to 1.14)	0.61	4.4 (11.9)	4.1 (11.7)	0.60 (-0.15 to 1.34)	0.11
rs4407290	*AA + AG*	*GG*	1.7 (13.1)	2.2 (13.8)	-0.38 (-2.22 to 1.47)	0.69	4.8 (11.6)	4.2 (11.8)	0.66 (-0.88 to 2.20)	0.40
rs2236168	*TT + CT*	*CC*	2.2 (13.7)	2.0 (14.2)	0.38 (-0.73 to 1.49)	0.50	4.4 (11.8)	3.9 (11.7)	-0.33 (-1.24 to 0.59)	0.48
rs148756340	*GG + GA*	*AA*	4.8 (15.2)	2.1 (13.7)	1.91 (-0.88 to 4.71)	0.17	2.8 (13.0)	4.3 (11.7)	-0.11 (-2.40 to 2.17)	0.92
rs561525	*CC+ CT*	*TT*	2.8 (14.0)	2.1 (13.7)	0.93 (-1.43 to 3.29)	0.43	4.6 (10.8)	4.2 (11.8)	-0.15 (-2.11 to 1.80)	0.87
rs11904439	*GG + GA*	*AA*	5.0 (15.2)	1.9 (13.6)	2.09 (0.43 to 3.75)	0.01*	3.8 (12.5)	4.3 (11.7)	-0.22 (-1.58 to 1.14)	0.75
rs206857	*TT + TC*	*CC*	2.7 (14.4)	1.9 (13.4)	0.50 (-0.44 to 1.44)	0.30	4.4 (12.0)	4.2 (12.0)	-0.17 (-0.95 to 0.60)	0.66
rs10175754	*CC + CT*	*TT*	2.6 (14.3)	2.0 (13.5)	0.34 (-0.64 to 1.32)	0.49	4.4 (12.1)	4.2 (11.6)	0.04 (-0.77 to 0.84)	0.93
rs206800	*CC + CT*	*TT*	2.3 (14.2)	2.1 (13.5)	0.20 (-0.73 to 1.14)	0.67	4.6 (11.6)	4.1 (11.9)	0.25 (-0.51 to 1.02)	0.52
rs206803	*GG + GT*	*TT*	2.5 (14.5)	2.0 (13.5)	0.17 (-0.82 to 1.16)	0.74	4.9 (11.7)	4.0 (11.8)	0.40 (-0.42 to 1.22)	0.33
rs3769616	*TT + TC*	*CC*	3.6 (14.8)	2.0 (13.6)	1.15 (-0. 83 to 2.68)	0.14	4.6 (12.1)	4.2 (11.7)	0.42 (-0.84 to 1.67)	0.51
rs206804	*AA + AG*	*GG*	2.4 (14.2)	2.0 (13.5)	0.31 (-0.61 to 1.23)	0.51	4.6 (11.7)	4.1 (11.8)	0.18 (-0.58 to 0.94)	0.64

CI, confidence interval. Change is last available follow-up minus baseline measurement of pulse pressure or mean arterial pressure. Difference (95% confidence interval) between minor allele carriers minus non-minor allele carriers (reference), accounted for family clusters and study center modeled as random effects, and were adjusted for sex, age, body mass index, blood pressure (pulse pressure or mean arterial pressure), smoking and drinking, use of female sex hormones (all at baseline), duration of follow-up and three indicator variables coding for starting or stopping antihypertensive drug treatment from baseline to follow-up or remaining on treatment. * Significant with the false discovery rate set at 0.25.

Table S4. Hazard Ratios Relating Risk of Hypertension to Variation in the *XOR* Gene (Hypertension Cohort, *n*=2050)

SNP	Minor allele carriers	Non-minor allele carriers	Adjusted Hazard Ratio (95% Confidence Interval)	*P*
rs1054889	*AA + AG*	*GG*	1.03 (0.88 to 1.21)	0.72
rs207440	*AA + AG*	*GG*	0.93 (0.70 to 1.25)	0.64
rs9308919	*TT + TC*	*CC*	0.90 (0.77 to 1.06)	0.21
rs12621192	*TT + TC*	*CC*	0.97 (0.83 to 1.13)	0.70
rs732436	*AA + AG*	*GG*	0.92 (0.79 to 1.06)	0.25
rs17011368	*CC + CT*	*TT*	0.94 (0.67 to 1.32)	0.73
rs2043013	*CC + CA*	*AA*	1.05 (0.86 to 1.27)	0.64
rs10181969	*CC + CT*	*TT*	1.01 (0.79 to 1.30)	0.92
rs11900892	*AA + AG*	*GG*	1.11 (0.89 to 1.39)	0.35
rs13398137	*CC + CA*	*AA*	0.99 (0.81 to 1.22)	0.93
rs11894627	*AA + GA*	*GG*	1.00 (0.83 to 1.20)	0.98
rs45449798	*CC + CT*	*TT*	1.34 (0.92 to 1.94)	0.13
rs45520334	*AG*	*GG*	1.28 (0.87 to 1.90)	0.22
rs3769618	*TT + TC*	*CC*	1.01 (0.87 to 1.17)	0.93
rs4407290	*AA + AG*	*GG*	0.92 (0.69 to 1.22)	0.55
rs2236168	*TT + CT*	*CC*	1.03 (0.87 to 1.24)	0.79
rs148756340	*GG + GA*	*AA*	1.69 (1.11 to 2.57)	0.01*
rs561525	*CC+ CT*	*TT*	1.20 (0.79 to 1.80)	0.39
rs11904439	*GG + GA*	*AA*	1.31 (1.03 to 1.68)	0.02*
rs206857	*TT + TC*	*CC*	1.14 (0.97 to 1.34)	0.12
rs10175754	*CC + CT*	*TT*	1.11 (0.94 to 1.31)	0.22
rs206800	*CC + CT*	*TT*	1.07 (0.90 to 1.26)	0.46
rs206803	*GG + GT*	*TT*	1.12 (0.93 to 1.34)	0.22
rs3769616	*TT + TC*	*CC*	1.06 (0.85 to 1.33)	0.58
rs206804	*AA + AG*	*GG*	1.08 (0.92 to 1.27)	0.58

Hazard ratios (95% confidence interval) express the risk of minor allele carriers *vs.* non-minor allele carriers (reference), accounted for family clusters and study center modeled as random effects and adjusting for sex, age, body mass index, systolic and diastolic blood pressure, smoking and drinking, and intake of female sex hormones (all at baseline). *Significant with the false discovery rate set at 0.25.

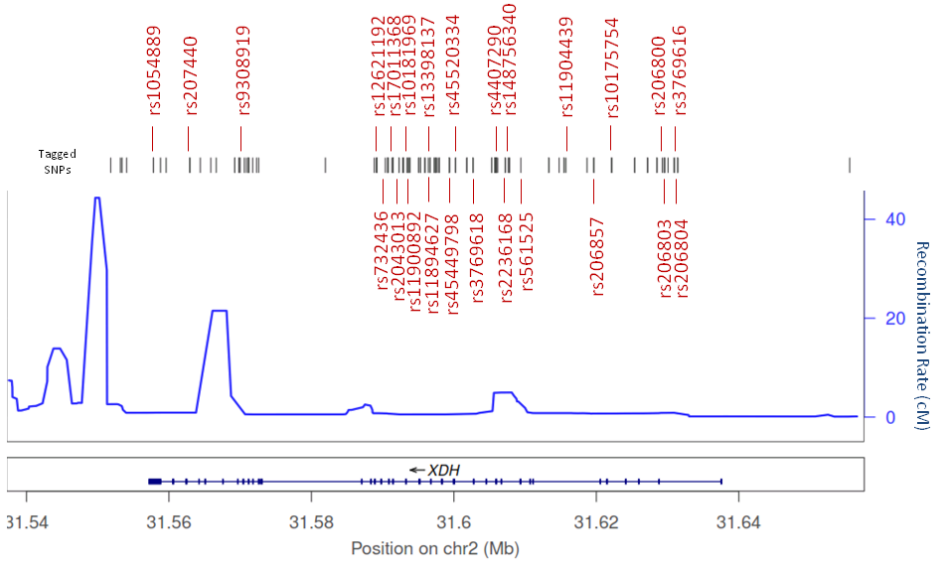

Figure S1. Plot of the *XOR* Gene and Flanking Regions on Chromosome 2.

3

Associations of plasma uric acid and purine metabolites with blood pressure in children: the KOALA birth cohort study

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ABSTRACT

Objective

Elevated serum uric acid concentration has been associated with high blood pressure (BP) and hypertension. A putative underlying mechanism is the accumulation of reactive oxygen species when uric acid is generated by an increased enzyme activity of xanthine oxidase (XO). The aims of the present study were to investigate the associations between plasma uric acid concentration, purine metabolite ratios, as proxies for increased XO activity, and SBP and DBP in school-age children.

Methods

Cross-sectional analyses were performed in 246 children (46% boys; mean age 7.1 years) from the Dutch KOALA Birth Cohort Study. Purine metabolites were determined with ultra-performance liquid chromatography–tandem mass spectrometry. During a home visit, a nurse collected a blood sample and measured BP three times; in addition, parents measured their child's BP on three consecutive days, in the morning and evening. Generalized estimating equations were used for analyses while controlling for variables such as sex, age, body mass index, physical activity, and dietary intake.

Results

In multivariable analysis, uric acid (per SD of 38mmol/L) was associated with DBP z-scores [sß 0.07; 95% confidence interval (CI), 0.01-1.14], but not with SBP *z*-scores. Higher ratios of uric acid/xanthine (per SD of 138) (sß 0.09; CI, 0.01-0.17) and xanthine/hypoxanthine (per SD of 321) (sß 0.08; CI, 0.02-0.17) were associated with higher DBP z-scores, but not with SBP z-scores.

Conclusion

In school-age children, uric acid and the ratios of uric acid/xanthine and xanthine/hypoxanthine were significantly associated with DBP z-scores. Suggesting that, both uric acid concentration and increased XO activity are associated with BP.

INTRODUCTION

Recent meta-analyses show that elevated serum uric acid concentration increase the risk of incident hypertension among adults, independent of traditional risk factors.[1,2] Associations were significantly stronger in younger adults and tended to be larger in women, but studies among individuals younger than 18 years of age were not included in these meta-analyses.[2] During the past several years, a number of studies assessed this association in older children and adolescents, and extended evidence for elevated blood pressure (BP) and hypertension among those with elevated serum uric acid concentration to this age group (summarized in Table 1).

Several mechanisms have been proposed that causally link uric acid with hypertension. For example, uric acid stimulates the renin–angiotensin system, leading to increased production of the vasoconstrictor angiotensin II.[3] Another mechanism not directly related to uric acid concentration, but rather to its production, is the generation of reactive oxygen species (ROS) during the production of uric acid. In the terminal steps of the purine catabolism, the rate-limiting enzyme xanthine oxidase (XO) catalyzes the oxidation of hypoxanthine to xanthine and xanthine to uric acid (Figure 1). The main sources of XO are the liver[4–6] and intestine;[7] but XO can also be released into plasma and circulate to remote sites in which it binds to the surface of endothelial cells.[8] When molecular oxygen acts as the electron acceptor, superoxide radical anion ($O_2^{\bullet-}$),[9] and hydrogen peroxide (H_2O_2) are generated as by-products of the oxidation step. These ROS inhibit endothelial nitric oxide (NO) production and consequently impair the vasodilatory reaction.[8,10,11] In addition, the ROS produced by XO activity induce the adherence of leukocytes to endothelial cells, which in turn leads to more ROS generation and the initiation of proinflammatory events.[8]

We hypothesize that uric acid production, independent of its concentration, contributes to elevated BP and hypertension. As uric acid concentration is not an adequate marker for the production, proxies for uric acid production need to be investigated (Figure 2). As is known that XO is a rate-limiting enzyme, an increased XO activity may lead to a relative decrease in the more upstream metabolites, thus, lower hypoxanthine compared with xanthine and lower xanthine compared with uric acid concentration, resulting in higher ratios of xanthine/hypoxanthine, uric acid/xanthine, and uric acid/hypoxanthine. The latter is based on animal studies and studies examining patients with xanthine oxidoreductase deficiency, a genetic disorder called xanthinuria, who have decreased concentrations of the more upstream metabolites.[12-15]

Table 1. Overview of studies on association between uric acid and blood pressure in children and adolescents

Author (publication), country	Study design & setting, (collection period)	Study population, n (boys %). Age, in years	Uric acid measurement & definition of hyperuricemia	Blood pressure measurement & definition of hypertension	Results	Adjustment confounders
Viazzi, Italy (2016)[34]	Longitudinal intervention study, lifestyle changes (increase of physical activity and dietary modifications). Unit for CV Risk Assessment in Children, (2005–2014). Follow-up: 1.5 years	248 (55). Range: 8.6–13.2	12-h fasting. Median sUA (μmol/L) for NT: 244 [208–285]; PH: 262 [226–309]; HT: 274 [214–309]; HT 99th percentile: 274 [202–333].	Mean of 3 replicates after a 5 minute rest, using an aneroid sphygmomanometer. NT: SBP and DBP <90th (*n*=124); PH: SBP and DBP ≥90th, but both ≤95th (*n*=41); HT: SBP and DBP ≥95th, but both <99th (*n*=59); HT 99th: SBP and DBP ≥99th percentile (*n*=24). Ref. population: NHBPEP.[24]	Baseline sUA associated with SBP *z*-scores at follow-up (β 0.15; CI 0.04–0.26; *P* <0.01), but not with DBP. Baseline sUA associated with HT 99th percentile during follow-up (OR 2.55; CI 1.22–5.34; *P*=0.01), but not with HT (OR 1.26; CI 0.74–2.17; *P*=0.39); after lifestyle changes.	Age, sex, pubertal status, HOMA-index, BMI *z*-scores (at baseline and follow-up)
Sun, Taiwan (2015)[45]	Longitudinal, MJ Health Screening Centre, (1999–2008). Median follow-up: 7.2 years	5,303 (57). Range: 10–15	10-h fasting. Mean sUA (mg/dL) 6.7±1.6 for boys and 5.4±1.2 for girls. Hyperuricemia: sUA >7.6.	Sitting position, by a nurse using a mercury sphygmomanometer. HT: SBP ≥130 and DBP ≥85 mmHg (number unspecified).	Baseline hyperuricemia higher HR for HT at follow-up for boys (HR: 2.34; CI, 1.41–3.75; *P*=0.001); but not for girls (HR: 2.76; CI 0.45–15.26; *P*=0.26).	Confounders unspecified
Gonçalves, Portugal (2015)[50]	Cross-sectional, EPIteen Study, (2006–2007)	1,286 (47). Mean: 17	12-h overnight fasting. Mean sUA (mg/L) for boys 52.6±9.9 and 38.2±8.2 for girls. Categorized in sex-specific quartiles.	Mean of 3 replicates, separate by at least 10 minutes of rest, using a mercury sphygmomano-meter using an auscultatory deivce	No sig. difference in SBP (boys: *P*=0.79; girls: *P*=0.61) and DBP (boys: *P*=0.36; girls: *P*=0.28) among sUA quartiles.	-
Sun, USA (2014)[26]	Cross-sectional, Bogalusa Heart Study, (1987–1988). Including 1,577 white and 1037 black children	Total 2,614 (51). Range: 4–18. Mean: 10.8±3.5 for white and 11.2±3.6 black children	12-h overnight fasting. Mean sUA (mg/dL) for white 4.5±1.4 and black 4.1±1.4 boys; for white 4.1±1.1 and black 3.6±1.0 girls.	Mean of 6 replicates, in a relaxed sitting position, by 2 trained observers using a mercury sphygmomanometer. Outcome MAP: DBP + 1/3 pulse pressure.	sUA associated with MAP in black girls (β 0.11; *P* <0.05), but not in the total study population, black boys or in white boys and girls.	Age and BMI
Pan, China (2014)[51]	Cross-sectional, Xinjiang Congenital Heart Disease survey, (2009–2010)	3,778 (47). Range: 10–15	At least 8-h fasting. Mean sUA (μmol/L) 205.6±65.2. Categorized in quartiles.	Mean of 2 replicates measured in 10 minutes, by doctor/nurse. HT: SBP or DBP≥95th percentile (*n*=352). Ref. population: Chinese.[52]	Increased sUA associated with increased DBP (sig. trend); not for SBP. Participants with sUA in Q3 (OR 1.60; CI, 1.10–2.32; *P*<0.01) or Q4 (OR 1.81; CI, 1.18–2.78; *P* <0.01) higher odds for HT *vs.* Q1.	Age, sex, ethnicity total cholesterol, HDL, LDL, triglycerides, eGFR, fasting glucose, and BMI

Table 1. (continued)

Author (publication), country	Study design & setting, (collection period)	Study population, n (boys %). Age, in years	Uric acid measurement & definition of hyperuricemia	Blood pressure measurement & definition of hypertension	Results	Adjustment confounders
Pan, China (2014)[51]	Cross-sectional, Xinjiang Congenital Heart Disease survey, (2009–2010)	3,778 (47). Range: 10–15	At least 8-h fasting. Mean sUA (μmol/L) 205.6±65.2. Categorized in quartiles.	Mean of 2 replicates measured in 10 minutes, by doctor/nurse. HT: SBP or DBP≥95th percentile (*n*=352). Ref. population: Chinese.[52]	Increased sUA associated with increased DBP (sig. trend); not for SBP. Participants with sUA in Q3 (OR 1.60; CI, 1.10–2.32; *P*<0.01) or Q4 (OR 1.81; CI, 1.18–2.78; *P* <0.01) higher odds for HT *vs.* Q1.	Age, sex, ethnicity total cholesterol, HDL, LDL, triglycerides, eGFR, fasting glucose, and BMI
Viazzi, (2013)[27]	Cross-sectional, San Gerardo Hospital, Unit for CV Risk Assessment in Children, (2004 –2012)	501 (55.9). Range: 6–18 Mean: 10.8±2.4	12-h fasting period. Mean sUA (mg/dL): boys 4.38±1.14 and girls 4.15±0.95. Categorized in quartiles.	Mean of 3 replicates after 5 minute rest, by a nurse using a calibrated aneroid sphygmomanometer NT: SBP and DBP<90th (*n*=156); PH: SBP and/or DBP between 90th–95th (*n*=87); HT: SBP and/or DBP ≥95th percentile (n=136); TH: family paediatrician diagnosed elevated BP but measured as NT (*n*=122). Ref. population: NHBPEP. [24]	1) sUA associated with SBP z-score (*P*=0.03) and DBP z-score (*P*=0.02), in adj. regression (no effect sizes). 2) 1 mg/dL higher sUA (OR 1.40; *P*<0.01) or sUA in Q4 (OR 1.67; *P*=0.05) higher odds for HT *vs.* the entire population. 3) 1 mg/dL higher sUA (OR 1.60; *P*<0.01; and OR 2.25; *P*=0.01) or sUA in Q4 (OR 1.54; *P*=0.01; and OR 2.04; *P*=0.02) higher odds for HT or PH *vs.* NT, respectively.	Age, sex, pubertal status, BMI z-scores, HOMA index
Viazzi, (2013)[27]	Cross-sectional, San Gerardo Hospital, Unit for CV Risk Assessment in Children, (2004 –2012)	501 (55.9). Range: 6–18 Mean: 10.8±2.4	12-h fasting period. Mean sUA (mg/dL): boys 4.38±1.14 and girls 4.15±0.95. Categorized in quartiles.	Mean of 3 replicates after 5 minute rest, by a nurse using a calibrated aneroid sphygmomanometer NT: SBP and DBP<90th (*n*=156); PH: SBP and/or DBP between 90th–95th (*n*=87); HT: SBP and/or DBP ≥95th percentile (n=136); TH: family paediatrician diagnosed elevated BP but measured as NT (*n*=122). Ref. population: NHBPEP. [24]	1) sUA associated with SBP z-score (*P*=0.03) and DBP z-score (*P*=0.02), in adj. regression (no effect sizes). 2) 1 mg/dL higher sUA (OR 1.40; *P*<0.01) or sUA in Q4 (OR 1.67; *P*=0.05) higher odds for HT *vs.* the entire population. 3) 1 mg/dL higher sUA (OR 1.60; *P*<0.01; and OR 2.25; *P*=0.01) or sUA in Q4 (OR 1.54; *P*=0.01; and OR 2.04; *P*=0.02) higher odds for HT or PH *vs.* NT, respectively.	Age, sex, pubertal status, BMI z-scores, HOMA index

Table 1. (continued)

Author (publication), country	Study design & setting, (collection period)	Study population, n (boys %). Age, in years	Uric acid measurement & definition of hyperuricemia	Blood pressure measurement & definition of hypertension	Results	Adjustment confounders
Pacifico, Italy (2009)[33]	Case-control, Obese children (n=120) enrolled at Department of Pediatrics, La Sapienza University of Rome, (2007–2008)	170 (52). Mean: 10.6 for obese and 10.2 for controls	Overnight fast sUA (mmol/L). Mean sUA: obese 0.28 (range 0.27–0.30); controls 0.19 (range 0.17–0.20).	Sitting position, mean of 3 replicates after 10-min rest, during physical examination, using automated oscillatory device,. Elevated BP SBP or DBP>95th. Ref. population: NHBPEP.[24]	sUA associated with SBP (β 0.002; CI 0.0001 – 0.003; *P*<0.05).	Age, sex, Tanner-stage, and creatinine
Jones, USA (2008)[28]	Cross-sectional, American of African (*n*=74), European (*n*=29), and Asian (*n*=1) descent	104 (64). Range: 7–18 Mean: 13.7±2.6	sUA in mg/dL. Elevated sUA>4.9.	ABPM every 20-min in 24-h period, using the auscultatory device (with oscillometric "back-up"). Ref population: German and Hungarian children.[55] HT: BPI>1; for systolic (54%) and diastolic (16%)	sUA associated with 24h-SBP and 24h-DBP (β 1.77; *P*=0.048; and β 1.55; *P*=0.02) Participants with elevated sUA a higher odds for increased 24-h diastolic BPI (OR: 2.09; CI 1.07–4.07; *P*=0.02).	Race, sex, age, and BMI z-score
Gilardini, Italy (2008)[31]	Cross-sectional, Institution of Auxologico (national centre for the study of obesity)	89 (43). Range: 7–18	Plasma UA (μmol/L) Mean: boys: 410.4±107.1; girls and 344.9±83.3.	ABPM every 15-min during day and every 20-min during night time, using oscillometric device. Daytime sustained HT: daytime SBP and/or DBP≥95th (*n*=59). Ref population: German and Hungarian children.[55]	Plasma UA higher in those with night-time HT (6.9±1.5) *vs.* those with normal night-time BP (5.9±1.7) (*P*<0.05). Plasma UA not correlated with any BP parameters (adj. analyses).	BMI and waist circumference
Ford, USA (2007)[56]	Cross-sectional, NHANES, (1992–2002)	1,370. Range: 12–17	sUA (μmol/L) Mean 301.9 ±2.0 (range 113.0 – 719.7). Group sUA ≤291.5 (1: REF), (2) >291.5 - ≤339; and (3) >339.	Mean of 4 replicates in a sitting position at the mobile examination centre, using a mercury-gravity manometer. HT: SBP and/or DBP ≥90th percentile (*n* = 59). Ref. population: NHBPEP.[24]	Elevated sUA was associated with a higher odds for HT. Group 2 (OR: 2.07; CI, 0.90–4.76) and group 3 (OR 3.74; CI 1.86–7.52) *vs.* group 1 (*P*<0.01 for trend). Additionally ad. Association attenuated (OR: 1.48; and 2.05; *P*=0.07 for trend).	Age, sex, race, and concentration of C-reactive protein, MetS. Additionally adj. for waist-circumference and BMI *z*-scores

Table 1. (continued)

Author (publication), country	Study design & setting, (collection period)	Study population, n (boys %). Age, in years	Uric acid measurement & definition of hyperuricemia	Blood pressure measurement & definition of hypertension	Results	Adjustment confounders
Alper, USA (2005)[30]	Longitudinal, Bogalusa Heart Study, including *n* = 334 white and *n* = 243 (42) black children. Baseline (1973–1974); Follow-up (1976–1977) Mean follow-up: 11.4 years (range 7.4 – 15.0)	577 (42); Mean boys: white 13.6±3.2 and black 12.6±3.8 Mean girls: white 13.4±3.1 and black 12.9±3.7	sUA (μmol/L) Mean sUA at baseline: Boys: white 303.3±83.2 and black 261.7±89.2 Girls: white.9±65.4 and black 220.1±71.4 Mean sUA at follow-up: Boys: white 362.8±77.3; black 350.9±77.3 Girls: white 261.7±65.4; black 255.8±136.8.	Mean of 6 replicates taken by each of 2 randomly assigned trained observers, using a mercury sphygmomanometers.	Childhood sUA associated with SBP (r=0.31; *P*<0.01) and DBP (r=0.20; *P*<0.01) in childhood and adulthood SBP (r=0.29; *P*<0.01) and DBP (r 0.28; *P*<0.01). sUA at childhood correlated with childhood and adulthood SBP and DBP (*P*<0.01) in girls, but not in black boys. Change in sUA was associated with SBP (*P*=0.03) and DBP (*P*=0.01).	Childhood analyses adj. for age and sex; Adulthood analyses additionally adj. for childhood BMI, SBP and UA, and change in age and UA
Feig, USA (2003) [29]	Case-control, Paediatric Renal Clinic for hypertension (*n*=125)	165 (61). Range: 6–18. Mean 13.4±3.3	sUA (mg/dL). Mean 3.6±0.8; for controls; 7±1.3 for primary HT; 4.3±1.4 for secondary HT; and.6±0.7 for white coat HT.	Mean of 3 replicates in relaxed seated position, using a Dinamap device. HT: SBP or DBP>95th percentile. Secondary HT: identifiable etiology for elevated BP. White coat HT identified with 24-h ABPM. Ref. population: NHBPEP.[24]	sUA higher in those with primary (*P*<0.01) and secondary HT (*P*<0.01) *vs.* controls. sUA related with SBP (r=0.73, *P*<0.01) and DBP (r=0.61, *P*<0.01) (adj. BMI).	BMI
Goldstein, USA (1993)[57]	Cross-sectional, Cycle III of NHANES	6,268 (52). Range: 1–17	sUA (mg/dL).	BP levels reported as BP_{beg} BP_{end} of the physical examination, measured by nurse-members of the survey team, using a stethoscope mercury sphygmomanometer.	sUA has a predictive effect on SBP_{beg} and DBP_{beg} (*F1*, 64.67, *P*<0.01; *F1*, 7.62, *P*=0.006) and SBP_{end} (*F1* 54.23, *P* <0.01). For white boys similar results, but for white girls only DBP remained sig. Black, association attenuated but trend remained.	Sex, race, age, height, weight, and sexual maturity controlled

CV: cardiovascular risk; sUA: serum uric acid; NT: normotensive; PH: pre hypertension; HT: hypertension; sig. significant; SBP: systolic blood pressure; DBP: diastolic blood pressure; NHBPEP: National High Blood Pressure Education Program; adj: adjusted; HOMA: homeostatic model assessment; OR: odds ratio; BP: blood pressure; HR: hazard ratio; CI: confidence interval; MAP: mean arterial pressure; BMI: body mass index; HDL: high-density lipoprotein; LDL: low-density lipoprotein; eGFR: estimated Glomerular Filtration Rate; TH: transiently elevated blood pressure; RR: relative risk; AMPB: ambulatory blood pressure; NHANES: National Health and Nutrition Examination Survey; POW: percentage of overweight; HbAc1: Haemoglobin A1c; MetS: metabolic syndrome. When UA is classified in group or quartiles, group 1 refers to those with the lowest uric acid concentrations.

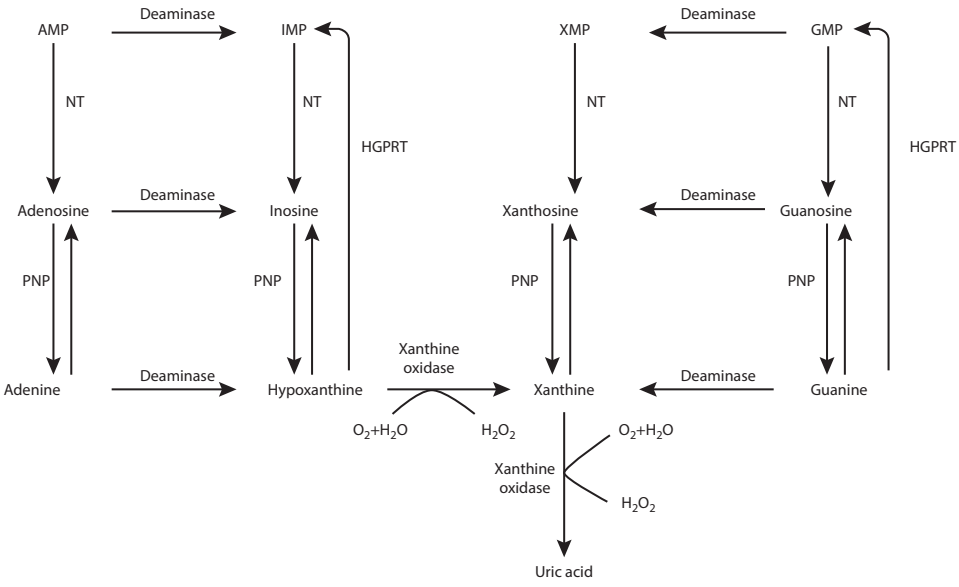

Figure 1. During the final stage of purine metabolism, xanthine oxidase (XO) breaks down hypoxanthine to xanthine, and xanthine to UA, meanwhile generating reactive oxygen species (ROS). These ROS may cause oxidative stress and inactivate the vasodilator nitric oxide. AMP: adenosine monophosphate; IMP inosine monophosphate; XMP: Xanthosine monophosphate; GMP: guanosine monophosphate; NT: nucleotidase; PNP: purine nucleoside phosphorylase: HGPRT: hypoxanthine-guanine phosphoribosyltransferase.

In view of the above, the aims of the present study were to extend existing evidence on the relation between plasma uric acid concentration and BP to school-age children, and to investigate associations between purine metabolite ratios, considered as proxies for an increased XO activity, and BP. Younger individuals are a desirable population in which to investigate this relationship, as common hypertension and metabolic risk factors are less likely to be present. Furthermore, uric acid may be involved in the development of hypertension before vascular damage has occurred but may have less influence on BP levels once vascular damage is permanent.[16]

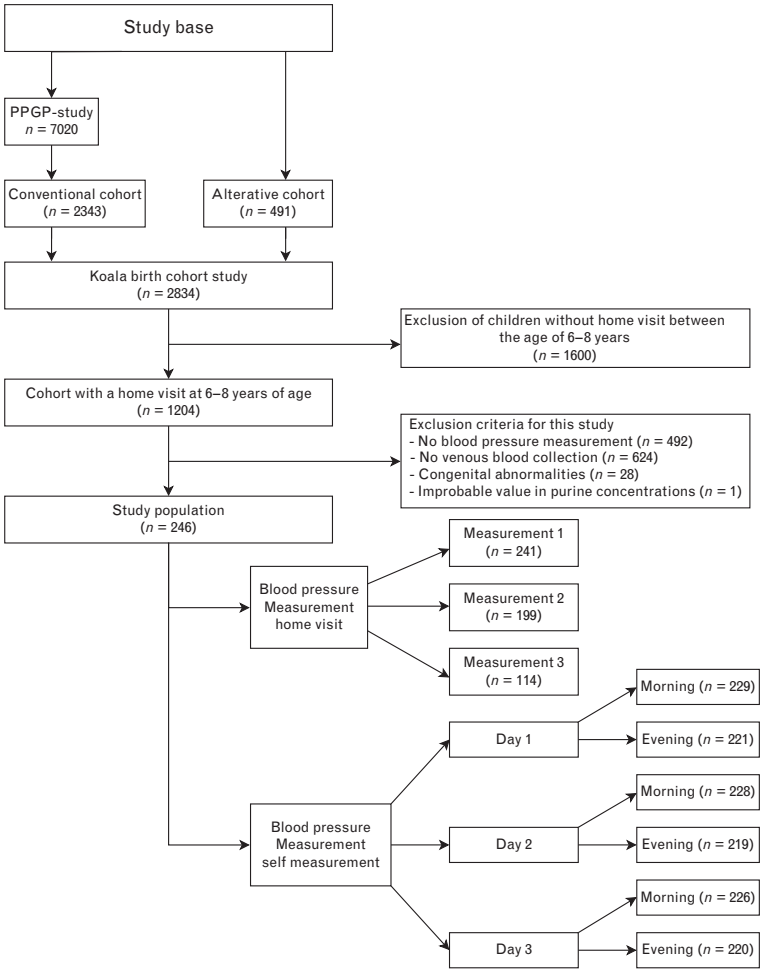

Figure 2. Flow chart of the study population. The KOALA Birth Cohort study included pregnant women with a conventional lifestyle (recruited from the on-going Pregnancy-related Pelvic Girdle Pain study) or those recruited through 'alternative' channels. Reasons for exclusion with numbers are presented.

METHODS

Participants and study design

The current analyses were conducted in the context of the KOALA Birth Cohort Study (A Dutch acronym for child, parents and health: lifestyle and genetic constitution) in The Netherlands.[17] In the years 2000–2002, pregnant women were recruited at 34 weeks of gestation and followed since. The cohort originates from two recruitment groups: healthy

pregnant women with a conventional lifestyle (*n*=2343) and pregnant women recruited through alternative channels (*n*=491) (Figure 2). The women with a conventional lifestyle have been retrieved from an ongoing prospective cohort study on pregnancy-related pelvic girdle pain study in the Netherlands. The second recruitment group consists of pregnant women acquired through anthroposophic doctors and midwives, anthroposophic under-five clinics, posters in organic food shops, magazines for special interest groups, and Steiner schools. The latter group of women was considered to have an alternative lifestyle that could involve dietary habits (vegetarian, organic), child rearing practices, vaccination schemes, and/or use of antibiotics. All parents had signed the informed consent and the study was approved by the Medical Ethics Committee of Maastricht University Medical Centre (MUMC^{+}).

A subgroup of 1204 parents was asked for consent for a home visit for anthropometric measurements, collection of venous blood, and a BP measurement from the child at the age of 6-7 years. This subgroup comprised participants who had home visits for blood collection from the mother during pregnancy and/or the child at age 2 years, and who were still active participants. Venous blood samples were obtained from 600 children and BP measurements from 712 children. Exclusion criteria for the current study were children with congenital heart disease, severe mental disability and/or severe autism, twins, and improbable values in purine concentrations. A total of 246 children that met the criteria were included in the present study (Figure 2).

Information on potential confounders was prospectively collected by parent-completed questionnaires during pregnancy (weeks 14 and 34) and at ages 3, 7, 12, and 24 months, 4–5 years, and 6-7 years of the child. Dietary information was collected using a food frequency questionnaire (FFQ) covering a period of 4 weeks at the age of 4 years. The FFQ was specifically developed to assess childhood energy intake and validated by the doubly labelled water method.[18] Details on collection of dietary information are described elsewhere.[19] During the home visit at 6–7 years, the child's body weight and height were measured. BMI (weight/height^2, kg/m^2) was standardized by recoding it into age-specific and sex-specific BMI z-scores using data from the Dutch reference population.[20] For descriptive purposes, children were classified into normal weight (BMI z-score <1.04, corresponding to the 85th percentile), overweight (BMI z-score between ≥1.04 and <1.65, corresponding to the 85th and 95th percentile), and obesity (BMI z-score between ≥1.65, corresponding to the 95th percentile).[21]

Purine metabolite determination

Venous blood samples were collected in EDTA tubes during the home visit to assess uric acid, xanthine, and hypoxanthine concentrations in plasma. For ethical reasons children were not asked to fast overnight; however, 1.5 h prior to and during the home visit, the children were not allowed to eat or drink, resulting in a mean fast time of 2.2 h (SD 1.2 h). After collection, the samples were stored at -20°C and thawed in a water bath at 37°C prior to analysis and homogenated by vortex mixing. A volume of 30 ml plasma was

mixed with 30 mL of internal standards solution, and while vortex mixing 500 ml of acetonitrile was added and vortex mixed for another 10 s. After centrifugation, the supernatant was transferred to a glass sample vial and evaporated to dryness under nitrogen gas at 45°C for ±15 min. The sample was then taken up in 150 ml 50 mmol/l ammoniumformiate pH 4.00. The purine metabolites were determined with ultra-performance liquid chromatography–tandem mass spectrometry.[22] The ratios of uric acid to xanthine [uric acid (mmol/L)/xanthine (mmol/L)], uric acid to hypoxanthine [uric acid (mmol/L)/hypoxanthine (mmol/L)], and xanthine to hypoxanthine [xanthine (mmol/L)/hypoxanthine (mmol/L)] were calculated.

Blood pressure measurement

Mean arterial BP was measured during the home visit, using a validated Omron 705IT oscillometric automated BP monitor (Omron Healthcare Europe BV, Hoofddorp, The Netherlands).[23] The device automatically provided an estimated SBP and DBP. Up to three measurements were performed by a trained nurse after a 5-min rest, whereas the child was sitting and not allowed to talk or move. BP was measured up to three times on the upper left arm while it was resting on a table. Afterwards, parents were asked to measure their child's BP on three consecutive days, in the morning and in the evening. The parents were instructed by the trained nurse and the same protocol was used. SBP and DBP percentiles and z-scores were calculated according to the nomograms recommended by the National High Blood Pressure Education Program on High Blood Pressure in Children and Adolescents (in the Fourth Report).[24] As BP was measured with an oscillometric device during a single study visit, a formal diagnosis of hypertension is not possible. Therefore, participants were characterized as having 'normal blood pressure' if the mean (measurement performed by the nurse) of SBP and DBP were both less than 95th percentile and 'elevated blood pressure' if the mean SBP or DBP percentile was at least 95th percentile.

Statistical analysis

The characteristics of the participants are given as numbers and proportions for categorical variables and as mean (SD) or median (interquartile range) values for continuous variables with a normal or nonnormal distribution, respectively. Missing values were classified as unknown for categorical variables (maternal smoking during pregnancy *n*=3; passive smoking during pregnancy *n*=6). Participants with missing values on the FFQ (*n*=11) were excluded for the analyses in the third model. Data analysis was performed using SPSS version 20.0 (SPSS Inc., Chicago, Illinois, USA).

Uric acid concentration differs between men and women; therefore, purine values and their ratios were compared between boys and girls with the independent t test or Mann–Whitney test for values with a normal or nonnormal distribution data, respectively.[25] Standardized SBP and DBP z-scores were compared across the three different conditions (home visit measurements versus self-measurement and self-measurements in the

morning versus self-measurements in the evening), using the dependent t test, respectively.

Generalized estimating equation (GEE) models with unstructured correlation structure were used for analysis of the repeated BP measurements [studied as standardized z-scores (adj. for age, sex, and height z-scores)]. Up to nine measurements were available, taken under three different conditions. The BP measurement condition (as the index variable) and recruitment group (conventional; alternative) as confounder were included in the first model. The analyses included only one single purine metabolite or ratio at a time. In the second model, the following confounders were additionally included: age, infant sex (male, female), and BMI z-scores. In the third model, potential confounders that changed the regression coefficient of uric acid and SBP or DBP by more than 10% were added to the GEE model. The following variables met this criterion: place and mode of delivery (vaginal delivery at home, vaginal delivery in hospital, artificial delivery in hospital, and caesarean section in hospital), maternal smoking during pregnancy (no, yes, and unknown), passive smoking during pregnancy (<1h a week, >1 h a week, and unknown), total physical activity at 4-5 years of age (h/week), energy intake (kJ/day), protein intake (%), carbohydrate intake (%), and fibre intake (g/day). By adding dietary variables as confounder in the analyses, we attempt to control for dietary factors that potentially affect BP. The following variables did not meet the criterion and were therefore not included in the third model: prematurity (born before 37 weeks of gestation), maternal pre-pregnancy BMI (kg/m^2), alcohol consumption during late pregnancy (number of consumptions), education level of the mother (lower education, middle education, and high education), birth weight (in grams), duration of breastfeeding (months), and fasting time before blood collection (h).

Finally, we tested whether the association between uric acid concentrations with BP z-scores was modified by sex or BP measurement condition by including an interaction term in the fully adjusted GEE model. The interaction terms were nonsignificant (*P* value >0.05); therefore, the analyses were not stratified by sex or BP measurement condition.

RESULTS

The characteristics of the KOALA cohort (*n*=2834) and the study population are shown in Table 2.

The study population consisted of 246 children, who were on average ages 7.1±0.4 years and of whom 114 (46.3%) were boys. In general, the two populations were similar, except that the study population consisted of less mothers who smoked during pregnancy (active and passive) and more children were delivered naturally at home than in the entire KOALA cohort.

Table 2. Baseline characteristics of the KOALA Birth Cohort Study and the study population

		KOALA Birth Cohort Study (*n*=2834)	Study population (*n*=246)
Gender, boy n (%)		1543 (50.9)	114 (46.3)
Recruitment group, conventional n (%)		2343 (82.7)	207 (84.1)
Age during home visit (years), mean ± SD		-	7.05 ± 0.41
BMI z-score during home visit, mean ± SD		-	-0.24 ± 0.87
Normal weight, n (%)			227 (92.3)
Overweight, n (%)			16 (6.5)
Obese, n (%)			3 (1.2)
Height z-scored, mean ± SD			0.50 ± 0.88
Birth weight (grams), mean ± SD		3504 ± 512	3551 ± 438
Maternal education,[a] n (%)	Low	289 (9.5)	20 (8.1)
	Middle	1060 (34.9)	89 (36.2)
	High	1341 (44.2)	123 (50.0)
	Unknown	343 (11.3)	14 (5.7)
Maternal pre-pregnancy BMI, mean ± SD		23.5 ± 4.5	23.5 ± 4.1
Maternal smoking during pregnancy, n (%)	yes	222 (7.3)	11 (4.5)
	unknown	209 (6.9)	3 (1.2)
Maternal passive smoking during pregnancy (>1h week), n (%)	yes	448 (14.8)	23 (9.3)
	unknown	255 (8.4)	6 (2.4)
Alcohol consumption during pregnancy (number of glasses), mean ± SD		0.3 ± 0.9	0.2 ± 0.7
Duration of pregnancy, n (%)	Mean ± SD	39.8 ± 5.0	39.5 ± 1.1
	<37 weeks	93 (3.1)	0 (0)
	Unknown	202 (6.7)	1 (0.4)
Place and mode of delivery,[b] n (%)	Natural delivery at home	1187 (39.1)	113 (45.9)
	Natural delivery in hospital	924 (30.5)	81 (32.9)
	Artificial delivery at home	1 (0.0)	0 (0.0)
	Artificial delivery in hospital	223 (7.4)	23 (9.3)
	Caesarean section in hospital	311 (10.3)	28 (11.4)
	Unknown	381 (12.6)	1 (0.4)
Duration of breastfeeding (months), mean ± SD		4.91 ± 4.55	5.5 ± 4.2
Total physical activity (hours/week), mean ± SD		9.4 ± 4.5	9.6 ± 4.7
Dietary factors of the child at age four, mean ± SD[c]	Total energy intake, kilojoules	6172 ± 1286	6086 ± 1331
	Energy percentage from fat	29.6 ± 4.2	29.6 ± 3.9
	Energy percentage from protein	14.6 ± 2.1	14.8 ± 2.1
	Energy percentage from carbohydrate	55.8 ± 5.0	55.5 ± 4.7
	Fibre, grams	2.5 ± 0.5	2.5 ± 0.5

* Numbers do not always add up to the total because of missing data. Abbreviations: SD, standard deviation; BMI, body mass index. [a] Low: primary school, preparatory vocational or lower general secondary school, Middle: vocational, higher general secondary or pre-university education, High: higher vocational or academic education. [b] Artificial delivery: Induced labour, as when started with drugs or medical devices. [c] Missing data for 13 participants on variables total physical activity and dietary factors.

Data on purine metabolite concentrations and their ratios are shown in Table 3. The mean plasma uric acid concentration was slightly higher for boys [203.9 mmol/L (SD 39.6)] than for girls [201.8 mmol/L (SD 36.2)] but did not differ significantly (*P*=0.67).

Median hypoxanthine [boys: 2.4 mmol/L (IQR 2.7); girls 2.5 mmol/L (IQR 3.5)] and mean xanthine concentrations [boys: 0.5 mmol/L (SD 0.20); girls: 0.5 mmol/L (SD 0.15)] and the different ratios did not differ substantially by sex.

Table 3. Plasma concentrations of uric acid, xanthine, and hypoxanthine and their ratios

		Total population (*n*=246)	Boys (*n*=114)	Girls (*n*=132)
Purine metabolite				
Uric acid (µmol/L)	Mean ± SD	202.8 ± 37.7	203.9 ± 39.6	201.8 ± 36.2
Xanthine (µmol/L)	Mean ± SD	0.53 ± 0.17	0.53 ± 0.20	0.53 ± 0.15
Hypoxanthine (µmol/L)	Median (IQR)	2.5 (3.2)	2.4 (2.7)	2.5 (3.5)
Ratio				
Uric acid / xanthine	Mean ± SD	417.2 ± 138.4	425.0 ± 154.1	410.5 ± 123.4
Uric acid / hypoxanthine	Median (IQR)	83.9 (94.5)	86.7 (102.5)	81.6 (86.5)
Xanthine / hypoxanthine	Median (IQR)	4.7 (4.8)	4.7 (4.6)	4.6 (4.8)

Abbreviations: SD: standard deviation; IQR: interquartile range

In Table 4, BP data are presented. Elevated BP was found in 64 (26%) of the children. The correlation among the self-measurement ranged from 0.20 up to 0.50. BP z-scores were higher when measurements were performed by the parents than these performed by the nurse (*P*-value <0.001). Furthermore, measurements performed in the evening were higher than these performed in the morning for SBP z-score (*P*=0.001), but not for DBP z-score (*P*=0.96).

Table 4. Blood pressure levels measured by a trained nurse during the home visit and by the parents in the morning and evening

		Systolic BP *z*-scores	Diastolic BP *z*-scores
Home visit [a]			
	Mean ± SD	0.84 ± 0.69	0.49 ± 0.86
Self-measurement			
Morning [b]	Mean ± SD	0.78 ± 0.94	0.49 ± 0.88
Evening [c]	Mean ± SD	0.88 ± 0.94	0.49 ± 0.85

BP: blood pressure. Home visit: BP is measured by a trained nurse, the mean of three replicates. Self-measurement: BP is measured by the parents on three consecutive days, in the morning and evening. Missing for each condition: home visit: *n*=5 (boys *n*=1; girls *n*=4; self-measurement: morning *n*=16 (boys *n*=5; girls *n*=11) and evening *n*=23 (boys *n*=8; girls *n*=15).

Purine metabolite concentrations and blood pressure

Multivariable analysis showed that a 1 SD (38 mmol/L) higher plasma uric acid concentration was associated with a higher DBP z-scores, although statistical significance was only reached in the fully adjusted model [adj. sß 0.07; 95% confidence interval (CI), 0.01–0.14]. No association was found with SBP z-scores (Table 5). Plasma xanthine and hypoxanthine concentrations were not associated with SBP z-scores or DBP z-scores, regardless of adjustment for potential confounders. Although the association

between plasma hypoxanthine with DBP z-scores was close to significance (adj. sß 0.06; CI, -0.01 to 0.13; *P*=0.10). An increase of 0.10 DBP z-score represents a difference of 0.93 mmHg for a 6-year-old boy with a height z-score of 0.00.

Table 5. Association between purine metabolite and their ratios with blood pressure z-scores in children.

		Model 1 sß (95% CI)[a]	*P*	Model 2 sß (95% CI)[b]	*P*	Model 3 sß (95% CI)[c]	*P*
Purine metabolite							
Uric acid	SBP	0.000 (-0.08 to 0.07)	0.99	0.01 (-0.07 to 0.08)	0.90	-0.02 (-0.10 to 0.06)	0.70
	DBP	0.04 (-0.02 to 0.11)	0.17	0.05 (-0.01 to 0.12)	0.08	0.07 (0.01 to 1.14)	0.03
Xanthine	SBP	-0.001 (-0.08 to 0.08)	0.99	0.002 (-0.08 to 0.08)	0.97	-0.03 (-0.10 to 0.05)	0.49
	DBP	-0.03 (-0.10 to 0.05)	0.61	-0.02 (-0.10 to 0.05)	0.54	-0.04 (-1.11 to 0.04)	0.35
Hypoxanthine	SBP	0.01 (-0.01 to 0.10)	0.76	0.01 (-0.07 to 0.09)	0.78	0.01 (-0.08 to 0.10)	0.82
	DBP	0.05 (-0.02 to 0.12)	0.13	0.06 (-0.01 to 0.12)	0.11	0.06 (-0.01 to 0.13)	0.10
Ratio							
Ratio uric acid / xanthine	SBP	-0.01 (-0.10 to 0.08)	0.81	-0.01 (-0.09 to 0.05)	0.83	0.003 (-0.08 to 0.08)	0.93
	DBP	0.06 (-0.02 to 0.14)	0.15	0.07 (-0.01 to 0.15)	0.10	0.09 (0.01 to 0.17)	0.03
Ratio uric acid / hypoxanthine	SBP	-0.002 (-0.04 to 0.04)	0.93	-0.01 (-0.05 to 0.04)	0.28	-0.003 (-0.06 to 0.06)	0.91
	DBP	-0.03 (-0.05 to -0.00)	0.04	-0.03 (-0.06 to 0.01)	0.11	-0.03 (-0.08 to 0.01)	0.17
Ratio xanthine / hypoxanthine	SBP	0.01 (-0.07 to 0.10)	0.74	0.01 (-0.07 to 0.09)	0.79	0.03 (-0.06 to 0.12)	0.50
	DBP	0.06 (-0.00 to 0.12)	0.06	0.06 (-0.00 to 0.12)	0.06	0.08 (0.02 to 0.14)	0.01

CI, confidence interval; BMI, body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure. [a] Adjusted for recruitment group (conventional; alternative). [b] Additionally adjusted for age, sex, and BMI *z*-scores. [c] Additionally adjusted for place and mode of delivery, maternal smoking during pregnancy (active and passive), total physical activity, and nutrition intake at 4 years of age (total energy intake (kcal), energy from carbohydrates (%) and energy from protein (%) and fibre intake (grams). Analyses included 232 children, due to missing values on the FFQ and for total physical activity (*n*=13). The purine metabolites and ratios expressed as standard deviations (uric acid 37.7 µmol/L; xanthine 0.17 µmol/L; hypoxanthine 6.9 µmol/L and the ratios uric acid/xanthine 138.4; uric acid/hypoxanthine 321.4, and xanthine / hypoxanthine 8.8). Outcome presented as standardized blood pressure *z*-scores (adj. for age, gender, and height *z*-scores; ref. population: NHBPEP.[24]

Purine metabolite ratios and blood pressure

The ratios uric acid/xanthine and xanthine/hypoxanthine were associated with DBP z-scores, although statistical significance was only reached in the fully adjusted model (adj. sß 0.09; CI, 0.01-0.17) and (adj. sß 0.08; CI, 0.02-0.14), respectively. None of the ratios was significantly associated with SBP z-scores. The ratio uric acid/hypoxanthine

was associated with DBP z-scores in the crude model but lost significance after adjustment for potential confounders.

DISCUSSION

The present study showed that, in school-age children, elevated plasma uric acid concentration was associated with higher DBP z-scores. Furthermore, elevated ratios of uric acid/xanthine and xanthine/hypoxanthine, considered as proxies for increased XO activity, were associated with higher DBP z-scores. Our findings support the hypothesis that next to an elevated uric acid concentration, an increased XO activity is associated with elevated BP.

Although an association between serum uric acid concentration and BP has been described before in older children and adolescents (summarized in Table 1), we were the first to show an association in school-age children at a mean age of 7 years. The studies performed so far mainly studied older children (with a mean age of 10 years or higher) and adolescents.[26-30] Moreover, we found an effect in a relatively healthy group of children, with respect to both the participants' uric acid concentration [hyperuricaemia (plasma uric acid concentration >5.0 mg/dL) *n*=0], whereas other studies often investigate children from clinical cohorts, who have a relatively high risk of cardiovascular diseases.[28,29,31-34]

To the best of our knowledge, we were the first to study the association between BP and several ratios of purine metabolites as proxies for an increased XO activity in the vasculature. We showed that higher uric acid/xanthine and xanthine/hypoxanthine ratios were associated with higher DBP z-scores. Our hypothesis is based on studies examining the effect of a deficiency in XO on purine metabolite concentrations;[13–15] however, whether an opposite effect in purine metabolite concentrations occurs by an increased XO activity is unknown. Further investigation is required to examine whether increased XO activity leads to a relative increase in the more downstream metabolites and that this will not be compensated by alterations in urinary excretion of uric acid or xanthine, or the degradation of hypoxanthine to inosine monophosphate by hypoxanthine-guanine phosphoribosyltransferase.[15]

In the present study, we found an association with DBP z-scores but not with SBP z-scores. Diastolic pressure and mean arterial pressure, the steady components of BP, reflect peripheral vascular resistance. Particularly in younger individuals (<50 years of age), DBP is the main predictor of cardiovascular risk. Later in life SBP and pulse pressure, the pulsatile components of BP become the main predictors.[35–37] As previously described, there are several plausible mechanisms linking uric acid to vascular resistance.[3] For example, uric acid in the circulation activates angiotensin II, thereby stimulating the nicotinamide adenine dinucleotide (phosphate) (reduced form) (NAD(P)H) oxidase that is the main source of ROS in the vasculature. ROS reduces the bioavailability of vasodilator NO, leading to vascular resistance. In turn, ROS increases the formation of

peroxynitrite, which can increase endothelial NO synthase uncoupling resulting in even more ROS formation.[38,39] Moreover, Landmesser et al. found that angiotensin II substantially increases endothelial XO activation in cultured endothelial cells, subsequent to NAD(P)H oxidase activation, leading to even more ROS formation.[40] Moreover, another mechanism not directly related to uric acid concentration, but rather to its production, is the accumulation of ROS during the degradation of purines by increased XO activity.

The magnitude of the BP change found in our study is relatively small; an increase of 1 SD in uric acid concentration or uric acid/xanthine and xanthine/hypoxanthine ratios was associated with an increase of approximately 0.08 higher DBP z-score. This represents an increase in DBP by 2 percentiles (or 1 mg/dL increase in plasma uric acid leads to an increase in DBP by 3 percentiles). This is probably not clinically relevant during childhood, nevertheless when maintained during adolescence and adulthood, it can have clinical implications.[29,30,34,41,42] Prolonged exposure to increased peripheral vascular resistance and high BP leads to embedding of the vessel structure in a remodelled extracellular matrix.[43,44] Initially, the remodelling of arteries will be adaptive, but it eventually becomes maladaptive and increases stiffness, contributing to cardiovascular complications of hypertension.[43] Furthermore, an increase in uric acid concentration or XO activity may not have clinical implication at the time, but it increases the risk of hypertension later in life.[30,34,45,46] For example, Viazzi et al. showed, among children at high cardiovascular risk, that even a moderately increased serum uric acid concentration at baseline was associated with elevated BP after a mean follow-up of 1.5 years, regardless of appropriate lifestyle changes such as weight loss and diet.[34] Accordingly, among participants in the Bogalusa Heart Study higher childhood serum uric concentration, even within the normal range, was associated with childhood and adult elevated BP (mean follow-up 11.4-year).[30]

In the current study, BP z-scores were higher than these from the reference population (Table 4). We made use of an oscillometric device to measure BP, whereas the available reference values have been obtained by an auscultatory device. Values obtained by an oscillometric device are considerably higher than these obtained by an auscultatory device. In addition, the average BP measured by the parents was higher than that measured by the trained nurse during the home visits, regardless of time of measurement (morning or evening). This same phenomenon has been observed previously in young adolescents and adults (<50 years),[47] but in young children it has not been studied extensively.[48,49] Our results show that these differences already occur in early childhood.

Our study has both strengths and weaknesses that deserve to be commented on. The adjustment for potential confounders, quantification of several purine metabolites, and the repeated BP measurements are major strengths of our study. However, the present study must be interpreted within the context of its potential limitations. First, we would like to emphasize the need for studies examining whether an increased XO activity causes shifts in several purine metabolite concentrations and their ratios. Second, we

used a cross-sectional design, so no conclusions concerning causality can be drawn. Third, an oscillometric device was used to measure mean arterial BP, and SBP and DBP were subsequently estimated. Reference data on oscillometric BP measurements would have been more appropriate to create SBP and DBP z-scores. Unfortunately, referential data on oscillometric BP measurements are limited and insufficiently validated. Further studies are required before a complete substitution can be made from using conventional auscultation method to oscillometry. Last, blood samples to determine the purine metabolites were only collected once. As purine concentrations can vary during the day, multiple measures would have been more accurate.

In conclusion, this study showed that plasma uric acid concentration and the ratios uric acid/xanthine and xanthine/hypoxanthine are associated with DBP z-scores in school-age children. These findings support the role of uric acid as a determinant of BP, already in early childhood, and confirm our hypothesis that an increased XO activity, reflected by increased purine ratios, is associated with BP. These findings highlight the need for further investigation on increased XO activity and BP.

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4

Uric acid and blood pressure: exploring the role of uric acid production in The Maastricht Study

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ABSTRACT

Objective

Accumulation of reactive oxygen species by increased uric acid (UA) production has been suggested as a possible underlying mechanism for the association between UA and high blood pressure. We therefore investigated the association between (i) serum UA concentration (ii) 24-h urinary UA excretion, as proxy for UA production, and ambulatory 24-h blood pressure and hypertension.

Methods

Cross-sectional analyses were conducted among 2555 individuals (52% men, mean age 60.0±8.2 years; 27% type 2 diabetes [by design]) from The Maastricht Study. Multivariable regression analyses were performed to investigate the association of serum UA and 24-h urinary UA excretion with 24-h pulse pressure (PP), 24-h mean arterial pressure (MAP) and hypertension.

Results

After adjustment for traditional hypertension risk factors, serum UA concentration (per standard deviation [SD] of 81 μmol/L) was associated with higher 24-h MAP (β 0.63 mmHg; 95% confidence interval [CI] 0.27-1.00) and positively associated with hypertension (odds ratio 1.43; CI 1.27-1.61). Urinary UA excretion (per SD of 140 mg/day/1.73m^2) was associated with higher 24-h MAP (β 0.79 mmHg; CI 0.46-1.12) and borderline significantly with hypertension (odds ratio 1.13; CI 1.02-1.25). There was no significant association between serum and 24-h urinary UA excretion with 24-h PP. There was no interaction with sex or age for the aforementioned associations.

Conclusion

Higher serum and urinary UA concentrations were associated with higher 24-h MAP and hypertension. These results suggest that serum and 24-urinary UA concentrations, the latter as proxy for UA production are, independent of each other, associated with blood pressure and hypertension.

INTRODUCTION

Uric acid, the final product of purine catabolism, has been associated with blood pressure and hypertension. Recent meta-analyses showed a significant association between serum uric acid and incident hypertension, independent of traditional risk factors.[1,2] Several plausible mechanisms have been proposed that causally link uric acid with elevated blood pressure and hypertension. One mechanism includes the activation of the renin–angiotensin–aldosterone system (RAAS) by elevated concentrations of uric acid, leading to increased production of the vasoconstrictor angiotensin II.[3] Another possible mechanism not directly related to uric acid concentration, but rather to its production, is the generation of reactive oxygen species (ROS) during the production of uric acid. The enzyme xanthine oxidoreductase catalyses the breakdown of hypoxanthine to xanthine and the latter to uric acid. When oxygen is the electron acceptor, superoxide radical anion ($O_2^{\bullet -}$)[4] and hydrogen peroxide (H_2O_2) are generated as by-products of the oxidation step. These ROS directly reduce the bioavailability of the vasodilator nitric oxide and lead to the formation of peroxynitrite, which can increase endothelial nitric oxide synthase uncoupling resulting in even more ROS formation.[5,6]
Studies published so far have focused mostly on serum uric acid and have ignored the distinction between uric acid *concentration* and its *production*. Since the production of uric acid may contribute, independent of uric acid concentration, to the pathogenesis of hypertension, the production should be investigated as well.[7] Facing the problem that it is not possible to directly measure uric acid production in a large population of individuals, and that uric acid concentration is not an adequate marker for production, proxies for uric acid production need to be investigated. Under normal conditions, the body compensates for increased uric acid production by increasing uric acid excretion, so that serum uric acid remains stable and within the normal range of 200-430 μmol/L for men and 120-340 μmol/L for women.[8] Uric acid is predominantly excreted via the urine; therefore we used 24-h urinary uric acid excretion as a proxy for uric acid production.
In view of the above, we aimed to evaluate whether serum uric acid concentration and/or 24-h urinary uric acid excretion, as proxy for uric acid production are, independent of each other, associated with 24-h ambulatory blood pressure and hypertension. We analysed the pulsatile and steady component of blood pressure captured by 24-h mean arterial pressure (MAP) and 24-h pulse pressure (PP), respectively.

METHODS

Study population and design

We used data from The Maastricht Study, an observational prospective cohort study. The rationale and methodology have been described previously.[9] In brief, the study focuses on the aetiology, pathophysiology, complications and comorbidities of type 2 diabetes mellitus (T2DM) and is characterized by an extensive phenotyping approach. Eligible for participation were all individuals aged between 40 and 75 years and living in the southern part of The Netherlands. Participants were recruited through mass media campaigns and from municipal registries and the regional Diabetes Patient Registry through mailings. Recruitment was stratified according to known T2DM status, with an oversampling of individuals with T2DM, for reasons of efficiency. The present report includes cross-sectional data from the first 3451 participants, who completed the baseline survey between November 2010 and September 2013. The examinations of each participant were performed within a time window of three months. The study has been approved by the institutional medical ethical committee (NL31329.068.10) and the Minister of Health, Welfare and Sports of The Netherlands (Permit 131088-105234-PG). All participants gave written informed consent.

For the present study we excluded individuals without data on serum uric acid (*n*=4), creatinine (*n*=6), 24-h urine collection (*n*=164), ambulatory blood pressure (*n*=482), level of education (*n*=77), smoking status (*n*=63), alcohol consumption (*n*=69), body mass index (BMI; *n*=3), estimated glomerular filtration rate (eGFR) (*n*=33), or diabetes duration (*n*=105). We also excluded individuals with type 1 diabetes mellitus (*n*=40) or on uric acid lowering therapy (i.e. allopurinol, benzbromarone) (*n*=67). A total of 2,555 individuals were included in the present analyses (Figure 1).

24-h ambulatory blood pressure measurement

Ambulatory blood pressure was measured with ambulatory 24-h blood pressure monitoring (WatchBP O3; Microlife AG, Widnau, Switzerland). Cuffs were applied to the participants' non-dominant arm. Measurements were done every 15 minutes during daytime ($08:00_{A.M.}$-$11:00_{P.M.}$) and every 30 minutes during the night ($11:00_{P.M.}$-$08:00_{A.M.}$) for a total of 24 hours. Mean 24-h blood pressure measurements were only calculated if there were >14 valid measurements at daytime and >7 valid measurements at night, according to the recommendation of the British Hypertension Society.[10] Mean 24-h systolic blood pressure (SBP) and diastolic blood pressure (DBP) were calculated based on hourly averages.[11] Twenty four-h mean PP was defined as 24-h SBP minus 24-h DBP, and MAP as mean 24-h DBP plus (0.412 × mean 24-h PP).[12] Hypertension was defined as mean 24-h SBP of at least 135 mmHg, 24-h DBP of at least 85 mmHg, or the use of antihypertensive medication. Use of antihypertensive medication was assessed during a medication interview where generic name, dose, and frequency were registered.

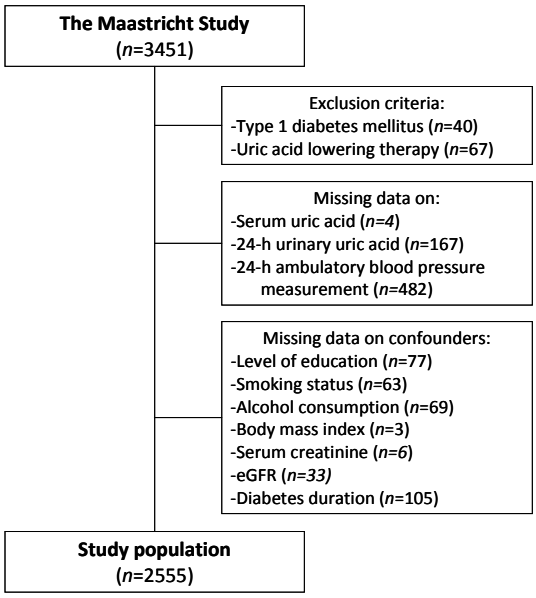

Figure 1. Flow-chart of study participants.

Uric acid determination

After an overnight fast, venous blood samples were collected to assess serum uric acid and creatinine concentrations with standard (enzymatic and/or colorimetric) methods by and automatic analyser (Beckman Synchron LX20; Beckman Coulter Inc., Brea California, USA or the Roche Cobase601 hs-cTnT assay (Roche) on the Cobas6000 analyser for the last 2585 samples) at Maastricht University Medical Centre (The Netherlands).

To assess urinary uric acid and creatinine excretion, participants were requested to collect a 24-h urine sample. Participants were instructed both orally and in writing on the procedure concerning the 24-hour urine collection. Only urine collections with a collection time between 20-h and 28-h were considered valid, in case of violation participants were asked to collect urine once more. Urinary uric acid and creatinine concentration were measured with a standard immunoturbidimetric assay by an automatic analyser (Beckman Synchron LX20, Beckman Coulter Inc., Brea, USA) and multiplied by collection volume to obtain the 24-h urinary uric acid excretion. According to the DuBois and DuBois equation, 24-h urinary uric acid and creatinine excretion were adjusted for body surface.[13]

Covariates

To determine diabetes status, all participants (except those who use insulin) underwent a standardized 2-h 75 g oral glucose tolerance test after an overnight fast as previously

described.[9] Glucose metabolism was defined according to the WHO 2006 criteria into normal glucose metabolism, impaired fasting glucose, impaired glucose tolerance and T2DM.[14] For this study, we defined having either impaired fasting glucose or impaired glucose tolerance as prediabetes.

Weight and height were measured without shoes and wearing light clothing using a scale and stadiometer to the nearest 0.5 kg or 0.1 cm (Seca, Hamburg, Germany). BMI was calculated as body weight (kg) divided by height squared (m^2). Waist and hip circumference were measured in duplicate midway between the lower rib margin and the iliac crest at the end of expiration and at the widest level over the greater trochanters, respectively (Seca, Hamburg, Germany). Waist-to-hip ratio was calculated as the mean of two waist circumference measurements divided by the mean of two hip circumference measurements. As previously described,[9] diabetes duration, education level, smoking status, and alcohol consumption were assessed by means of a self-reported questionnaire. Level of education was self-reported and classified into eight categories: 1) no education; 2) primary education; 3) lower vocational education; 4) intermediate general secondary education; 5) intermediate vocational education; 6) higher general secondary education; 7) higher vocational education; and 8) university. For this study, three groups were created for educational level: low (levels 1–3), middle (levels 4-6) and high (levels 7 and 8). Smoking status was based on self-report of smoking cigarettes, cigars and/or pipe tobacco and divided into three categories, i.e. non-smoker, former smoker and current smoker. Alcohol consumption was self-reported as the number of alcohol consumptions per week. One standard alcohol consumption is equivalent to 10 g (or 13 mL) alcohol. This corresponds to one glass of beer of 250 mL (5% alcohol), one glass of wine of 100 mL (12% alcohol), or one glass of spirit of 35 mL (35% alcohol).[15] For analyses, total alcohol consumption was evaluated in grams of alcohol per week. The patients were grouped into three categories, i.e. non-consumers [0 g of alcohol per week], low consumers [≤70 g of alcohol per week for females and ≤140 g of alcohol per week for males], and high consumers [>70 g of alcohol per week for females and >140 g of alcohol per week for males]." Physical activity was determined from the CHAMPS questionnaire (hours /week). Activities accounted were walking, cycling, gardening, household work, jogging/running, swimming, tennis, team sport, and exercise, regardless whether the activity was on a light or intense. GFR was estimated using the CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) equation based on both serum creatinine and serum cystatin C.[16] Use of uric acid lowering (allopurinol, febuxostat, probenecid, benzbromarone) and glucose-lowering medication were as well assessed during the medication interview.

Statistical analysis

The characteristics of the participants are given as mean values ± standard deviation (SD) for continuous variables and as numbers and proportions for categorical variables. To check if urinary uric acid excretion is an independent marker and does not represent

serum uric acid concentration we created a scatterplot and calculated the R^2 between those two variables.

Multiple linear or logistic regression analyses were used to determine the association between the independent variables serum uric acid concentration and 24-h urinary uric acid excretion with the dependent variables ambulatory 24-h PP, 24-h MAP and the odds ratio (OR) of prevalent hypertension. In the first model, the results of the crude analyses were presented. In the second model, crude results were adjusted for sex, age, glucose metabolism status, smoking status, alcohol consumption, eGFR, level of education, use of diabetes medication (no, oral medication, insulin with or without oral medication), RAAS-inhibitors and other antihypertensive medication (including beta-blockers) that have no known uricosuric properties, losartan (known as a RAAS inhibitor with a uricosuric effect),[17,18] and antihypertensive medication and lipid-lowering medication that may have a uricosuric effect (i.e. secondary uricosurics, amlodipine,[19] atorvastatin,[20] rosuvastatin).[20] In third model, the analyses with serum uric acid were additionally adjusted for 24-h urinary uric acid excretion; and the associations with 24-h urinary uric acid excretion were additionally adjusted for serum uric acid. To explore of serum and urinary uric acid concentrations are, independent of each other, associated with blood pressure and hypertension. The logistic regression models were adjusted for the same covariates, except for the use of antihypertensive medication, which is part of the definition of the outcome. As it has been suggested that uric acid has a more pronounced effect in younger and female individuals, we investigated whether the association between serum uric acid concentration and urine uric acid excretion with ambulatory blood pressure and hypertension differed with gender and age.[21-24] Since an elevated uric acid may be a consequence of a high BMI or waist-to-hip ratio, adjustment for BMI or waist-to-hip ratio in the analyses may lead to overadjustment and these variables were therefore not included in the main analyses. In a sensitivity analysis we additionally adjusted for BMI or waist-to-hip ratio.[25] Due to the large number of missing values on physical activity (*n*=278) this variable was not included as potential confounder in the main analyses. A sensitivity analysis was performed to control for this variable in the subset of participants with complete data. The null hypothesis was rejected for a two-sided *P*-value was less than 0.05, except for the interaction analyses, where a *P*-value less than 0.10 was used. Analyses were conducted using SPSS version 23 for windows (SPSS, Inc.).

RESULTS

The overall study population consisted of 2555 individuals with an average age of 59.8 (SD 8.1) years, 51% of whom were men. Due to oversampling of individuals with diabetes 24% (*n*=606) had T2DM. Table 1 shows the general characteristics of the study population and the individuals excluded from the analyses.

Table 1. Characteristics of The Maastricht Study population and the individuals excluded from the analyses because of missing values

	Study population (*n*=2,555)	Missing	Excluded because of missing values (*n*=896)
Serum uric acid (µmol/L)	329.3 ± 81.3	6	331.8 ± 85.00
Serum creatinine (umol/L)	77.2 ± 16.1	4	77.4 ± 17.7
Urine uric acid excretion (mg/day/1.73m^2)	518.1 ± 139.92	127	507.5 ±181.6
Urine creatinine excretion (mmol/day/1.73m^2)	12.0 ± 2.8	78	11.9 ± 4.1
Fractional uric acid excretion (FE_{UA})	9.3% ± 5.3	173	9.5% ± 10.4
Age (years)	59.8 ± 8.1	0	59.6 ± 8.6
Male sex (n)	51.0% (1303)	0	52.7% (472)
BMI (kg/m^2)	26.8 ± 4.3	5	27.9 ± 5.1
Waist-to-hip ratio	0.94 ± 0.09	4	0.96 (0.10)
Smoking status,(n)		63	
Never	35.9% (916)		28.3% (254)
Past	52.2% (1333)		46.4% (416)
Current	12.0% (306)		18.2% (163)
Alcohol consumption, (n)		69	
No	16.9% (432)		22.0% (197)
Low	56.6% (1446)		47.9% (429)
High	26.5% (677)		22.4% (201)
Educational level, (n)		77	
Low	31.4% (801)		37.1% (332)
Medium	28.6% (731)		24.8% (222)
High	40.0% (1,023)		29.6% (265)
Physical activity (hours/week)*	14.5 ± 8.2	179	13.0 ± 7.8
eGFR (ml/min per 1.73m^2)	88.25 ± 14.50	55	87.7 ± 16.5
Glucose metabolism status, (n)		0	
Normal glucose metabolism	60.6% (1549)		41.9% (375)
Impaired fasting glucose	4.3% (111)		3.6% (32)
Impaired glucose tolerance	11.3% (289)		8.8% (79)
Type 2 diabetes mellitus	23.7% (606)		41.2% (369)
Other type of diabetes	n/a		4.5% (41)
Diabetes treatment among individuals with T2DM, (n)		4	
No medication	25.1% (152)		15.5% (57)
Oral medication	55.0% (333)		58.7% (216)
Insulin with or without oral medication	20.0% (121)		25.8% (95)
Diabetes duration (years), median (range)	8.4 ± 7.2	178	7.4 ± 6.9
Hypertension, (n)	43.2% (1103)	406	59.8% (293)
Use of antihypertensive medication, (n)	37.1% (947)	4	44.6% (353)
RAAS-inihibitors	24.7% (631)		34.3% (307)
Other antihypertensives, no uricosuric effect	20.6% (527)		30.7% (275)
Losartan	2.9 (75)		3.8% (34)
Secondary uricosuric	13.5% (346)		20.4% (182)
Mean arterial pressure 24-h (mmHg)	88.5 ± 7.9	404	89.8 ± 8.5
Pulse pressure 24-h (mmHg)	45.2 ± 8.5	404	47.5 ± 9.8

Data are presented as mean and standard deviation (SD) unless otherwise indicated.
*Missing data for 278 individuals on variable physical activity.

On average the serum uric acid concentration was 329 µmol/L (SD 81) and 24-h urinary uric excretion was 518 mg/day/1.73m^2 (SD 140). In total 1,103 individuals (43%) had hypertension, of these 947 (86%) were on antihypertensive medication. In the

remaining 156 patients (14%), the diagnosis of hypertension relied on the threshold being exceeded for SBP (*n*=49; 31%), DBP (*n*=45; 29%), or both (*n*=62; 40%). Individuals excluded from analyses had a lower level of education, slightly higher BMI, and were more often T2DM or hypertensive patients.
The variance in serum uric acid, while significant (*P*-value <0.001), explains only 0.9% of the variance in urinary uric acid excretion (Figure 2).

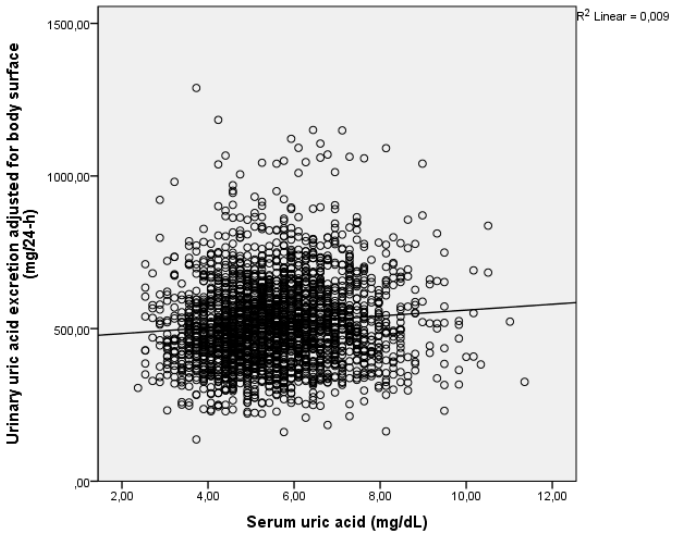

Figure 2. Uric acid excretion in 24-h urine (mg/1.73m^2) as a function of serum uric acid (mg/dL) in 2555 participants from The Maastricht Study.

Serum uric acid and 24-h blood pressure

Crude linear regression analysis showed that a 1 SD (81 µmol/L) higher serum uric acid concentration was associated with 1.73 mmHg (95% confidence interval [CI] 1.44 to 2.02 mmHg) higher 24-h MAP and greater odds for hypertension (OR 1.89; CI 1.73 to 2.07) (Table 2**,** model 1). Further adjustment for sex, age, and glucose metabolism status, smoking status, alcohol consumption, education, eGFR, and use of antihypertensive or diabetes medication, did not materially change the result (model 2). Furthermore, results remained significant with additional adjustment for 24-h urinary uric acid excretion (model 3). The association with 24-h PP was significant in the crude analysis (Table 2, model 1), but lost significance after adjustment for potential confounders (models 2 and 3).

Table 2. Associations between serum uric acid and urinary uric acid excretion with ambulatory mean arterial pressure, pulse pressure and hypertension

	Mean arterial pressure		Pulse pressure		Hypertension[#]	
	β (95% CI)	*P*-value	β (95% CI)	*P*-value	Odds ratio (95% CI)	*P*-value
Serum uric acid*						
Model 1	1.73 (1.44 to 2.03)	<0.001	1.70 (1.37 to 2.02)	<0.001	1.89 (1.73 to 2.07)	<0.001
Model 2	0.70 (0.35 to 1.07)	<0.001	0.09 (-0.28 to 0.45)	0.65	1.45 (1.29 to 1.63)	<0.001
Model 3	0.63 (0.27 to 1.00)	<0.01	0.08 (-0.30 to 0.43)	0.73	1.43 (1.27 to 1.61)	<0.001
Uric acid excretion[^]						
Model 1	1.98 (1.69 to 2.28)	<0.001	1.10 (0.77 to 1.42)	<0.001	1.26 (1.16 to 1.36)	<0.001
Model 2	0.84 (0.51 to 1.16)	<0.001	0.24 (-0.08 to 0.57)	0.15	1.16 (1.04 to 1.28)	<0.01
Model 3	0.79 (0.46 to 1.12)	<0.001	0.24 (-0.09 to 0.57)	0.15	1.13 (1.02 to 1.25)	0.03

* Serum uric acid expressed per standard deviation (81 μmol/L). [^] Uric acid excretion in 24-h urine expressed per standard deviation (140 mg/day/1.73m^2). [#] Hypertension is defined as a 24-h SBP of >135, or DBP>85 or the use of anti-hypertensive medication. In the adjusted analyses, no adjustment for antihypertensive medication. Model 1: Crude. Model 2 adjusted for age, sex, glucose metabolism status (normal, impaired, T2DM), smoking status (never, current, former), alcohol consumption (no, low, high), education (low, middle, high), eGFR, use of diabetes medication (no, insulin with or without oral medication) and antihypertensive medication (no, RAAS inhibitors, diuretics or other beta blockers and calcium), and duration of diabetes. Model 3, analyses with serum uric acid were additionally adjusted for 24-h urinary uric acid excretion; and the associations with 24-h urinary uric acid excretion were additionally adjusted for serum uric acid.

24-h urine uric acid excretion and 24-h blood pressure

Crude linear regression analysis showed that a 1 SD higher 24-h urinary uric acid excretion *(*140 mg/day/1.73m^2) was associated with higher 24-h MAP (β 1.98; CI 1.69 to 2.28 mmHg) and greater odds for hypertension (OR 1.26; CI 1.16 to 1.36) (Table 2, model 1). The associations of serum uric acid with 24-h MAP and hypertension did not materially change after further adjustment for potential confounders (model 2).
Further adjustment for serum uric acid concentration did not materially changes the results (model 3). The association between urinary uric acid excretion and 24-h PP was significant in the crude analysis (β 1.10; CI 0.77 to 1.42 mmHg), but lost significance after adjustment for potential confounders (Table 2, models 2 and 3; *P*-value <0.15).

Effect modification by age or sex

To determine whether the associations between serum uric acid and urine uric acid excretion with ambulatory PP, MAP and hypertension were different across sex and age strata, an interaction term was added to the fully adjusted regression models. No significant interaction with sex or age was identified in any of the investigated associations (*P*-value >0.05).

Sensitivity analyses

Including BMI or waist-to-hip ratio as a covariate to model 3 did not materially alter the associations between serum uric acid and blood pressure or hypertension. However, the

association between urinary uric acid excretion and hypertension was attenuated after including both BMI (OR 0.99; *P*=0.80) and waist-to-hip ratio (OR 1.04; *P*=0.45). Additional adjustment for physical activity did not materially alter the associations of serum uric acid and urinary uric acid excretion with ambulatory PP, MAP and hypertension (data not shown).

DISCUSSION

This study represents a comprehensive analysis of the association of uric acid with ambulatory blood pressure and hypertension in middle-aged individuals. Serum uric acid concentration and 24-h urinary uric acid excretion, as proxy for uric acid production, were independent of each other associated with ambulatory MAP and hypertension. To the best of our knowledge, this study is the first to show an independent association of 24-h urinary uric acid excretion, as proxy for uric acid production, with ambulatory blood pressure and hypertension.

Our results are in line with prior research showing that serum uric acid was associated with hypertension, independent of traditional hypertension risk factors.[2] A meta-analysis performed by Wang et al. in 2014, showed that a 1 mg/dL increase in uric acid was associated with an increased risk of incident hypertension (adj. relative risk 1.15; CI 1.06 to 1.26). In the current cross-sectional study a similar odds ratio of 1.20 (CI 1.10 to 1.31) for each 1 mg/dL higher serum uric acid concentration was found. In the present study, we found no age or sex related difference in the association between serum uric acid and any of the outcomes. This is in contrast with the meta-analysis performed by Grayson at al. which showed that the risk of hypertension among individuals with hyperuricemia was significantly larger in younger individuals and women.[1] This contrasting finding may be attributed to the inclusion criterium of an age of 40 years and older, resulting in a relative high age of our study population (mean age of 60.0 years).

There are no earlier studies examining the association between urinary uric acid excretion, as proxy for uric acid production, and blood pressure. We showed that urinary uric acid excretion was significantly associated with ambulatory MAP and hypertension, after adjustment for potential confounders including serum uric acid. These findings confirm our hypothesis that increased uric acid production is associated with blood pressure and hypertension. Uric acid is produced during the metabolism of endogenous (DNA, RNA and ATP) and exogenous (dietary) purines. Previous studies showed that an increase in uric acid production, either by endogenous or exogenous supply, increases urinary uric acid excretion.[8] Approximately 70% of the produced uric acid is eliminated by the kidney and 30% by the intestine (extra-renal pathway), whether the relative contribution of renal and extra-renal excretion is comparable in case of increased uric acid production needs investigation.[26] In case of decreased extra-renal excretion, serum uric acid and urinary uric acid excretion will increase. Among white individuals approximately 11% (HapMap, CHB, and JPT) has the Q141K mutation in the urate

transport *ABCG2*, which leads to a decrease in extra-renal uric acid excretion, causing an increased serum and urinary uric acid concentrations and eventually gout.[27-29] Theoretically, the "overproducers" in the current concept, those with increased urinary uric acid excretion, may include next to the genuine "uric acid overproducers" also "underexcretors of uric acid via the intestine (extra-renal elimination)".[27] Unfortunately it is not feasible to determine uric acid excretion via the intestine since it is degraded by uricase activity of the intestinal microbiota, leading to an almost complete lack of uric acid in the faeces.[30] However, by excluding patients on uric acid lowering therapy, we have attempted to exclude those with gout and therefore possibly with decreased renal or extra-renal excretion.

In addition, oxidation of hypoxanthine to xanthine does not have to be equivalent to the oxidation of xanthine to uric acid. Several feedback mechanisms can control the supply of xanthine and thereby the corresponding uric acid production.[31] For example, an increased urinary excretion of xanthine[32] or degradation of xanthine to xanthosine monophosphate by certain hypoxanthine-guanine phosphoribosyltransferase[33] decreases the xanthine concentration. If this is the case, increased oxidation of hypoxanthine to xanthine and the associated harmful accumulation of ROS will not lead to an increased uric acid production. In the future a systematic approach integrating the supply of hypoxanthine and xanthine and the involved pathways might be a more accurate way to determine uric acid production.

After adjustment for potential confounders (including serum uric acid) the association between urinary uric acid excretion and hypertension remained significant (Table 2, model 3). Further adjustment for BMI (P=0.80) or waist-to-hip ratio (P=0.45) attenuated the association. This might be explained by the fact that adiposity increases xanthine oxidoreductase activity in mice, thus leading to overproduction of uric acid.[25] Adjustment for BMI or waist-to-hip ratio (measure for abdominal visceral fat), might therefore lead to overadjustment. Further research is required to examine the influence of adiposity and visceral fat on uric acid production and its association with hypertension. The associations of serum uric acid with blood pressure and hypertension were not attenuated after including BMI or waist-to-hip ratio to the fully adjusted model. This is in line with the idea that serum uric acid concentration does not reflect uric acid production, since an increased uric acid production is compensated by increased excretion to maintain serum uric acid within the normal range.

In the present study serum uric acid and urinary uric acid excretion were associated with MAP but not with PP. MAP is the steady component of blood pressure reflecting vascular resistance, which can be increased by inhibiting the vasodilator nitric oxide. As previously described, several plausible mechanisms have been shown to link uric acid with a decrease in the bioavailability of nitric oxide. PP, the pulsatile component of blood pressure, increases as a consequence of prolonged exposure to increased vascular resistance and elevated blood pressure. This results in remodelling of the vascular extracellular matrix,[34,35] stiffening of the arteries and ultimately a loss in vascular compliance. Thus far, no prior studies investigated the association between uric acid and

pulse pressure in depth.[36-41] Previous studies investigated the association between uric acid and arterial stiffness. In line with the current results, a study conducted in a subset of The Maastricht Study population (*n*=614), showed no association of serum uric acid with stiffness of the aorta, or the carotid or femoral artery.[42] Nonetheless, conclusive evidence cannot be provided since the literature shows disparate results.[43-47] We therefore emphasize the need for further studies addressing the role of uric acid on pulse pressure and arterial stiffness. In particular longitudinal studies including middle-aged individuals, since uric acid may lead to early changes in arterial stiffness and subsequently to elevated PP, but may have less influence once vascular damage is permanent.[48]

The use of 24-h ambulatory blood pressure measurements and the adjustment for a large number of carefully measured potential confounders are major strengths of our study. Our study also has some limitations which should be considered. We would like to emphasize the need for further research validating urinary uric acid excretion as a proxy for uric acid production. Furthermore, we proposed that accumulation of reactive oxygen species is a potential underlying mechanism linking increased uric acid production with elevated blood pressure, but from the present study no conclusion can be drawn concerning the actual mechanism. Due to the cross-sectional nature of this study, causal relationships could not be determined. In addition, due to missing data, we had to exclude almost 900 participants. Although we assumed the random nature of these missing because most values were missing due to logistic factors (e.g. the temporary unavailability of ambulatory blood pressure monitors), the excluded individuals had a higher prevalence of (pre)diabetes and hypertension (Table 1). Furthermore, serum uric acid and 24-h urinary uric acid excretion was only determined once. Since uric acid concentrations can vary between days, multiple measures would have been more accurate. Finally, our study population consisted of relatively more individuals with T2DM between 40-75 years of age; therefore, the results might not be representative for the general population.

CONCLUSION

We found evidence for associations between serum uric acid and urinary uric acid excretion with ambulatory MAP and hypertension. By studying urinary uric acid excretion, we aimed to investigate whether an increased uric acid production was, independent of serum uric acid, associated with blood pressure. Finding significant associations supports our hypothesis, highlighting the need for further investigation on increased uric acid production and its effect on urinary uric acid excretion and blood pressure.

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PART II

Gout management by the patient and general practitioner

5

Medication adherence among gout patients initiated allopurinol: a retrospective cohort study in the Clinical Practice Research Datalink

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To be submitted

ABSTRACT

Objective

To assess medication adherence, including non-persistence, non-adherence and restarting therapy, among newly diagnosed gout patients initiated allopurinol and to identify factors associated with poor medication adherence.

Methods

A retrospective cohort study was conducted within the UK Clinical Practice Research Datalink (1987-2014) among incident gout patients, aged ≥40 years and initiating allopurinol (*n*=48,280). Medication adherence for allopurinol treatment was described by non-persistence (occurrence of a first gap of ≥90 days), non-adherence (proportion of days covered [PDC] <80% over observation period), restart of therapy after the first gap and subsequent medication adherence. Kaplan Meier survival and multivariable Cox- and logistic regression were used to estimate non-persistence and median time until discontinuation, and factors associated with non-persistence or non-adherence, respectively.

Results

Non-persistence increased from 38.5% (95% confidence interval [CI] 38.1-38.9) to 56.9% (CI 56.4-57.4) after 1 and 5 years of initiation, respectively. Median survival time until a first 90-day gap was 225 days (CI 220-231). Median PDC was 0.67 (IQR: 0.65) and 62% of the patients were considered non-adherent. After the occurrence of a first gap, 43.3% (CI 42.7-43.9) restated therapy within 1 year, yet only 52.3% (CI 51.4-53.1) persisted for 1 year. Females and current smokers had an increased risk for non-persistence and non-adherence, while older age, overweight, receiving antihypertensive medication or colchicine, and suffering from dementia, diabetes and dyslipidaemia decreased the risk.

Conclusion

Medication adherence among gout patients on allopurinol therapy is poor, particular among females, younger, and those with less comorbidities.

INTRODUCTION

Gout is a painful inflammatory condition, determined by the deposition of monosodium urate crystals within the joint and surrounding tissues. A worldwide prevalence of 0.6% has been described, but large variations across regions and sexes are acknowledged.[1] In the UK, approximately 4% of men and 1% of women are affected.[2] Both prevalence and incidence were significantly higher in 2012 than in 1997; with a 64% increase in prevalence and 30% increase in incidence over this period.[2] Gout and its main risk factor hyperuricemia have been associated with hypertension, cardiovascular disease, and chronic kidney failure.[3] In the long-term, gout and its accompanied comorbidities can result in impaired function and ultimately a decline in health-related quality of life.[4]

In patients with recurrent gout flares or tophi, it is recommended to start long-term uric acid lowering therapy to reduce the number of gout flares and resolve tophi.[5] Despite proven efficacy of these drugs,[6,7] a substantial subgroup of patients fails to achieve optimal clinical benefit, partly because of poor medication adherence. Previous studies showed that non-adherence, usually defined as 80% or less of the total observation time that is covered by medication, is associated with higher serum uric acid concentrations and more gout flares.[8-13] While non-persistence, occurrence of a gap in therapy use over time, may have little clinical effect in certain chronic diseases, a gap in uric acid lowering therapy might actually trigger or prolong a gout flare.[12] Hence, poor medication adherence may therefore lead to a more severe disease for the patient and to an increase in gout-related healthcare costs for society.[11,14]

Recently, a systematic review confirmed poor medication adherence among gout patients using uric acid lowering therapy.[15] Adherence ranged from 18% to 44% and persistence from 12% to 44%.[15] Twelve out of 16 studies included were conducted in the US, hampering transferability to a European setting where general practitioners are often the main healthcare provider. Data were often derived from electronic prescription records databases and managed health care plans, which only include insured residents and had, as a result, limited validity. Furthermore, most studies focused on non-adherence, but little is known about non-persistence, time until non-persistence, restart of therapy and subsequent medication adherence after first discontinuation.[16] In order to identify patients at high risk, factors associated with poor medication adherence should be determined. Several factors such as older age or suffering from certain comorbidities like hypertension and diabetes, were associated with higher adherence rates,[15] however, yet these factors have not or poorly been studied as determinants of non-persistence.

The objective of the current study was therefore to assess among newly diagnosed gout patients (i) rates of non-persistence and non-adherence with allopurinol therapy, (ii) determinants of non-persistence and non-adherence, and (iii) to describe the number of patients restarting therapy and subsequent medication adherence after the first occurrence of a gap in therapy.

METHODS

A retrospective cohort study was conducted using the UK Clinical Practice Research Datalink (CPRD), including more than 11.3 million individuals from 674 general practices in England, Northern Ireland, Scotland and Wales, representing 6.9% of the British population.[17] The database provides detailed information on demographics, drug prescriptions, clinical events, specialist referrals, and hospital admissions.[17] For this study we used data from 1987 through June 2014.

Study population

The study population consisted of individuals aged 40 years or older, with a first ever Read code of gout and a prescription of allopurinol during the period of valid data collection. The date of the first prescription of allopurinol after start of valid data collection defined the index-date. Follow-up was defined from the index-date to either the end of data collection (30th of June 2014), the date of transfer of the patient out of the practice area, or the patient's death, whichever came first. Patients who switched to other uric acid lowering therapies (probenecid or febuxostat) during follow-up were censored for analyses. Patients with a prescription of uric acid lowering therapy prior to the diagnosis of gout and those who had a follow-up less than 90 days after completion of the first prescription were excluded from analyses.

Measures of medication adherence

Non-persistence was defined as the occurrence of a gap of at least 30 or 90-days after the end of each prescription, in the case of overlap between two prescriptions (i.e. a repeat prescription within the duration of use of a previous prescription), the overlap days was added to the duration of treatment time. In case of a missing length, the median value of all allopurinol prescriptions was assigned. Non-persistence rate was measured by calculating the proportion of patients who discontinued treatment, a gap of at least 30-days or 90-days, over time.

Non-adherence was defined by the proportion of days covered (PDC). This was calculated as the number of days of prescribed medication divided by the total duration of follow-up, while truncating the possible overlap between two prescriptions. A PDC <0.80 was considered as non-adherence. Additionally, the medication possession ratio (MPR) was determined, calculated as number of days medication prescribed divided by the total duration of follow-up. In case of overlap between two prescriptions the overlap will be added to the number of days prescribed, this could lead to a MPR >1.0.

To improve insight into medication patterns, the proportion of patients restarting allopurinol therapy after the occurrence of a first 90-day gap was calculated. Subsequently, non-persistence estimates and non-adherence was once described in those patients restarting allopurinol therapy after a 90-day gap.

Potential determinants of non-adherence and non-persistence

Several determinants were explored for their association with non-persistence and non-adherence. These factors related first to patient characteristics at baseline including gender, age (40-49, 50-59, 60-69, 70-79, and ≥80 years), body mass index (<20.0, 20.0-24.9, 25.0-29.9, 30.0-34.9, and ≥35.0 kg/m^2), smoking status (never, current, former), alcohol consumption (yes, no), socio-economic status (low, low-medium, medium, medium-high, high), year start allopurinol (1987-1999, 2000-2005, 2006-2009, and 2010-2014 year), and the number of days between gout diagnosis and initiation of allopurinol. Additionally, the number of visits to general practitioners 12 months prior to the index date was considered (0, 1-9, 10-19, 20-29, ≥30 visits).
Second, comorbidities prior to the index date were considered, including alcoholism, chronic obstructive pulmonary disease (COPD), dementia, depression, diabetes mellitus, hypertension, ischemic heart disease, myocardial infarction, osteoarthritis, renal calculi, and stroke. In the case of multiple records, the record most prior to the index-date was used. In addition, renal function was evaluated by reviewing laboratory test data (estimated glomerular filtration rate (eGFR) [MDRD] where possible), and CPRD read codes which describe the stage of renal function. In the case of multiple eGFR values on the same day the mean value was used. CPRD read codes were prioritized if there was a laboratory test on the same day.
Third, medication prescriptions six months prior to the index-date for hypertension, use of statins (a proxy indicator for dyslipidaemia), and acute gout medication (non-steroidal anti-inflammatory drugs [NSAIDs], colchicine, and oral corticosteroids) were considered.

Statistical analysis

Descriptive statistics were used to characterize the study population. Kaplan-Meier life table analyses were used to present non-persistence (30-day or 90-day gap) estimates with a 95% confidence intervals (CI) for 1 and 5 years after initiation of allopurinol. In addition, the median number of days until non-persistence was calculated. In a second analysis, Kaplan-Meier life table was applied to estimate the proportion of patients restarting allopurinol therapy after first 90-day gap. In addition, the median number of days until restarting allopurinol therapy and subsequent medication adherence were once more calculated.
Cox-regression analyses were performed to study the strength of the association between determinants with non-persistence (90-day gap), by entering all covariates into the regression model. The proportional hazards assumption was tested by including time interaction terms into the model. In case of violation (*P*-value <0.05), hazard ratios for the association between that covariate and non-persistence were calculated for the first year after the index date. Multivariable logistic-regression analyses were performed to study the strength of the association between determinants with non-adherence (PDC <0.80).

All analyses were conducted using SAS software (Version 9.3; SAS Institute Inc., Cary, North Carolina).

RESULTS

Study population

Of the 131,565 newly diagnosed gout patients a total of 48,438 (38.8%) patients initiated allopurinol as the first uric acid lowering therapy during observation period. Of the 48,438 allopurinol users, 158 patients were excluded for analyses because follow-up was shorter than 90 days after completion of the first prescription, resulting in a study population of 48,280 gout patients.

Table 1 shows the general characteristics of the study population. The mean age of the study population was 64.6 (SD 13.2) years and 76% were male. Comorbidities were common, the most common being hypertension (51%), dyslipidaemia (30%), osteoarthritis (24%), and depression (16%), and 41% received antihypertensive medication.

Table 1. Baseline characteristics of allopurinol users with gout (*n*=48,280)

Male	75.7 (36,169)
Age (years), mean ± SD	64.6 ± 13.2
40-49 years	16.1 (7,750)
50-59 years	21.3 (10,305)
60-69 years	24.2 (11,684)
70-79 years	23.6 (11,375)
80+ years	14.8 (7,166)
BMI most recent prior to index date	
BMI (kg/m^2), mean ± SD	29.7 ± 5.4
<20.0	1.1% (536)
20.0-24.9	14.4 (6,969)
25.0-29.9	38.1 (18,393)
30.0-34.9	25.5 (12,311)
≥35.0	13.4 (6,467)
Missing	7.5 (3,604)
Smoking status	
Never	42.6 (20,573)
Current	12.1 (5,865)
Ex	43.5 (20,998)
Missing	1.8 (844)
Alcohol use	
No	15.1 (7,276)
Yes	78.0 (37,637)
Missing	7.1 (3,367)
Alcoholism	4.5 (2,150)

Table 1. (continued)

Calendar year start allopurinol, index date	
1987-1999	17.5 (8,430)
2000-2005	29.1 (14,071)
2006-2009	32.0 (15,464)
2010-2014	21.4 (10,315)
Days between gout diagnosis and initiation allopurinol, mean ± SD	605 ± 1039
0	24.3 (11,717)
1–90	26.7 (12,903)
91–365	14.1 (6,789)
>366	34.9 (16,871)
Number of GP visits in year prior to index date	
0	18.8 (9,062)
1–9	21.9 (10,576)
10–19	22.7 (10,975)
20–30	17.1 (8,268)
≥30	19.5 (9,399)
Socio-economic status	
Low	13.6 (6,557)
Low-medium	14.9 (7,177)
Medium	11.9 (5,760)
Medium - high	10.3 (4,973)
High	8.0 (3,875)
Missing	41.3 (19,938)
History of drug use within 6 months before index date	
Antihypertensive	40.6 (19,362)
Acute gout treatment	
Colchicine	22.2 (10,714)
Corticosteroid	7.6 (3,647)
NSAIDs	67.0 (32,332)
History of comorbidity ever before index date	
COPD	15.1 (7,296)
Dementia	0.5 (250)
Depression	15.5 (7,480)
Diabetes	10.3 (4,950)
Dyslipidaemia*	30.2 (14,603)
Hypertension	51.4 (24,830)
Ischemic heart disease	17.7 (8,561)
Myocardial infarction	8.9 (4,304)
Osteoarthritis	23.8 (11,507)
Renal calculi	1.1 (517)
Stroke	6.4 (3,111)
Most recent eGFR measurement	
(ml/min/1.73m^2), mean ± SD	66.5 ± 22.2
CKD 1	12.9 (6,212)
CKD 2	38.6 (18,644)
CKD 3	35.8 (17,299)
CKD 4	2.9 (1,420)
CKD 5	0.3 (141)
Missing	9.5 (4,564)

Data are presented as percentage and number (n) unless otherwise indicated. Abbreviations: BMI: body mass index; CKD: chronic kidney disease; GP: general practitioner; NSAIDs: non-steroidal anti-inflammatory drug; COPD: chronic obstructive pulmonary disease; eGFR: estimated glomerular filtration rate. CKD 1: eGFR≥90; CKD 2: eGFR 60<90; CKD 3: eGFR 30<60; CKD 4: eGFR 15<30; CKD 5: eGFR<15

Medication adherence

Non-persistence estimates for treatment with allopurinol are displayed in Figure 1. Considering a gap-length of 30-days, non-persistence with allopurinol therapy increased from 57.8% (95% CI 57.3-58.2) at year 1 to 80.9% (95% CI 80.4-81.2) at 5 years following initiation (Table 2). The median time until discontinuation was 225 days (95% CI 220-231). Increasing the gap length up to 90-days non-persistence estimates were 38.5% (95% CI 38.1-38.9) at year 1 and 56.9% (95% CI 56.4-57.4) at 5 years following initiation, with a median time until discontinuation of 1029 days (95% CI 988-1078).

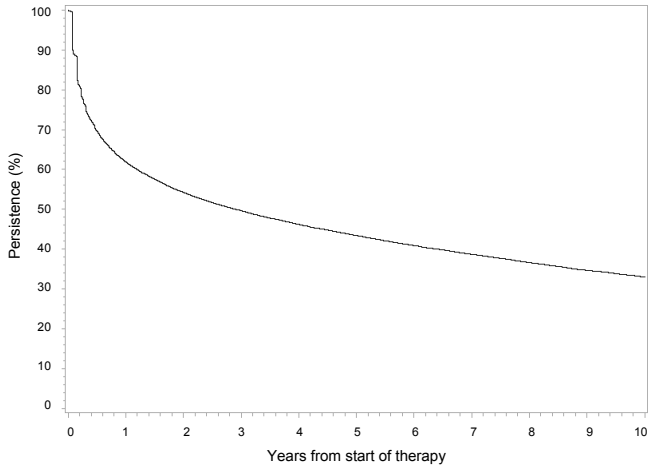

Figure 1. Kaplan-Meier curves for persistence to treatment with allopurinol medication in a the total study population

The vast majority of patients were non-adherent to allopurinol therapy (61%). The PDC was on average 0.57 (SD 0.43), indicating that patients had an allopurinol prescription for 57% of the observed days. The MPR was on average 0.66, and 47.6% of the patients had an MPR>0.80 and 23.2% even reached a MPR>1.00.

Of the 26,235 patients who experienced a 90-day gap, 15,013 (57%) restarted allopurinol therapy. Of all patients who had a gap of at least 90-days, 43.3% (95% CI 42.7-43.9) restarted therapy within 1 year, and this increased to 64.2% (95% CI 63.5-64.9) within 5 years (Figure 2). Median time until restart was 643 days (95% CI 617-678). Among patients who restarted therapy, 75.7% (95% CI 75.0-76.4) experienced a 30-day gap and 52.3% (95% CI 51.4-53.1) experienced a 90-day gap in the first year after re-initiation. Here, the median time until discontinuation was 88 days (95% CI 84-97) for a 30-day and 319 days (95% CI 301-340) for a 90-day gap (Figure 3). After restarting allopurinol, the average PDC was 0.49 (SD 0.31) and only 10.3% were considered adherent.

Table 2. Medication adherence among gout patients initiated allopurinol (*n*=48,280)

Non-persistence*	**30-day gap**		**90-day gap**	
First year	57.8	(57.3 - 58.2)	38.5	(38.1 - 38.9)
First five years	80.8	(80.4 - 81.2)	56.9	(56.4 - 57.4)
Time until discontinuation (days), median (95% CI)	225	(220 - 231)	1029	(988 - 1078)
Adherence (PDC)				
Mean ± SD	0.57 ± 0.34			
Median (IQR)	0.67 (0.65)			
Categories, n (%)				
<0.20	23.1		(11,136)	
0.20–0.40	10.9		(5,242)	
0.40–0.60	11.9		(5,756)	
0.60–0.80	15.8		(7,611)	
0.80–1.00	38.4		(18,535)	
Estimates for restarting therapy after a gap of at least 90-days				
First year	43.3		(42.7 - 43.9)	
First five years	64.2		(63.5 - 64.9)	
Time until restart (days), median (95% CI)	643		(617 - 678)	
Medication adherence after restarting therapy (*n*=14,084)				
Non-persistence*	**30-day gap**		**90-day gap**	
First year	75.7	(75.0 - 76.4)	52.3	(51.4 - 53.1)
First five years	91.1	(90.5 - 91.7)	71.6	(70.7 - 72.5)
Time until 2nd discontinuation (days), median (95% CI)	87	(84 - 94)	313	(294 - 334)
Adherence (PDC)				
Mean ± SD	0.49 ± 0.31			
Adherence (PDC≥0.80)				
no	76.8		(10,819)	
yes	23.2		(3,265)	

Data are presented as percentage and number (n) unless otherwise indicated. *Kaplan-Meier estimates for non-persistence at (%) at different time periods following initiation, by 30-day and 90-day gap lengths. CI: confidence interval; IQR: interquartile range; PDC: proportion days covered; SD: standard deviations.

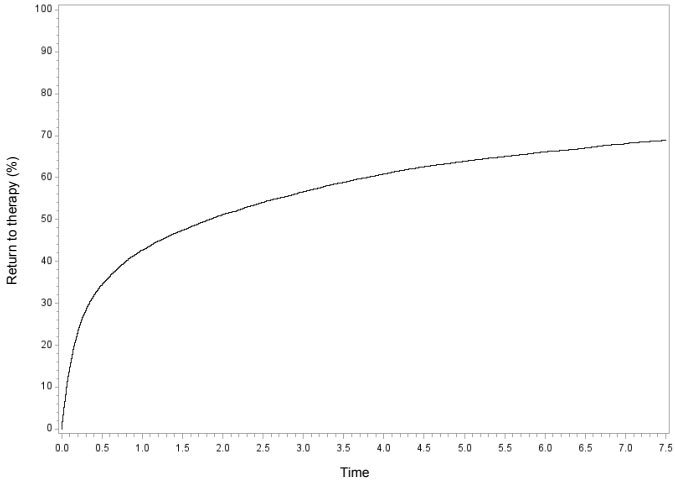

Figure 2. Kaplan-Meier curve for cumulative incidence of restart with allopurinol medication after first gap of 90 days.

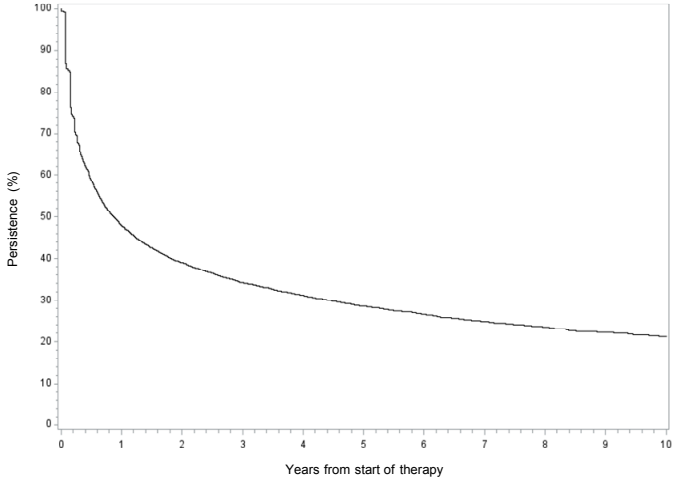

Figure 3. 1 – Kaplan-Meier curve for persistence to treatment with allopurinol medication after restarting therapy after a first 90-day gap.

Factors associated with medication adherence

Factors associated with non-persistence or non-adherence are presented in Table 3. In the multivariable adjusted model, female gender and current smoking status increased the risk of non-persistence and non-adherence. While older age, overweight, former smoking, use of colchicine, and suffering from dementia, diabetes mellitus, or dyslipidaemia decreased the risk of non-persistence and non-adherence.

The effect of use of antihypertensive medication, year start allopurinol, and kidney function on non-persistence, however, was not constant over time (p value interaction <0.05). When follow-up was restricted to the first 365 days following initiation, the use of antihypertensive medication decreases the risk to of non-persistence (HR: 0.73; 95% CI 0.70-0.77). Patients starting allopurinol between 2010 and 2014 were more likely to be persistent compared to patients who started between 1987 and 1999 (HR: 0.96; 95% CI 0.87-0.99). Kidney function was not significantly associated with non-persistence in the first year after initiation.

Table 3. Determinants of non-persistence (gap of ≥90-days) and non-adherence (PDC<0.80)

Characteristics	Non-persistence (90-day gap)				Non-adherence (PDC < 0.80)			
	Age and sex adjusted		Fully adjusted*		Age and sex adjusted		Fully adjusted*	
	HR	95% CI	HR	95% CI	OR	95% CI	OR	95% CI
Females (ref: male)	1.14	(1.11 – 1.18)	1.18	(1.14 – 1.23)	1.18	(1.13 – 1.23)	1.21	(1.14 – 1.28)
Age (ref: 40-49 years)								
50-59 years	0.74	(0.71 – 0.77)	0.81	(0.78 – 0.84)	0.60	(0.56 – 0.64)	0.70	(0.65 – 0.75)
60-69 years	0.59	(0.57 – 0.62)	0.70	(0.67 – 0.73)	0.41	(0.38 – 0.43)	0.55	(0.51 – 0.59)
70-79 years	0.60	(0.58 – 0.62)	0.74	(0.71 – 0.78)	0.40	(0.38 – 0.43)	0.57	(0.53 – 0.62)
80+ years	0.59	(0.56 – 0.62)	0.70	(0.66 – 0.74)	0.40	(0.37 – 0.43)	0.55	(0.51 – 0.60)
BMI (ref: 20.0-24.9 kg/m^2)								
<20.0 kg/m^2	1.19	(1.06 – 1.34)	1.10	(0.98 – 1.24)	1.30	(1.07 – 1.57)	1.15	(0.95 – 1.39)
25.0-29.9 kg/m^2	0.88	(0.85 – 0.91)	0.93	(0.89 – 0.96)	0.84	(0.80 – 0.89)	0.92	(0.87 – 0.98)
30.0-34.9 kg/m^2	0.81	(0.78 – 0.84)	0.89	(0.85 – 0.93)	0.73	(0.69 – 0.78)	0.86	(0.81 – 0.92)
≥35.0 kg/m^2	0.73	(0.69 – 0.76)	0.84	(0.80 – 0.88)	0.62	(0.58 – 0.67)	0.79	(0.73 – 0.85)
Smoking status (ref: never)								
Current	1.14	(1.10 – 1.19)	1.15	(1.11 – 1.19)	1.19	(1.12 – 1.27)	1.19	(1.12 – 1.27)
Ex	0.89	(0.87 – 0.92)	0.97	(0.94 – 0.99)	0.82	(0.79 – 0.85)	0.94	(0.90 – 0.98)
Alcohol use (ref: no)								
Yes	0.94	(0.91 – 0.97)	0.92	(0.89 – 0.95)	0.93	(0.89 – 0.99)	0.91	(0.86 – 0.96)
Calendar year initiation allopurinol# (ref: 1987 – 1999)								
2000-2005	0.94	(0.90 – 0.98)	1.02	(0.97 – 1.08)	0.80	(0.76 – 0.85)	0.91	(0.84 – 0.98)
2006-2009	0.91	(0.87 – 0.95)	1.06	(1.00 – 1.12)	0.69	(0.65 – 0.73)	0.86	(0.80 – 0.94)
2010-2014	0.78	(0.75 – 0.82)	0.93	(0.87 – 0.99)	0.59	(0.56 – 0.63)	0.75	(0.69 – 0.82)
Days between gout diagnosis and initiation allopurinol (ref: 0)								
1–90	0.88	(0.85 – 0.91)	0.89	(0.86 – 0.93)	0.84	(0.80 – 0.89)	0.87	(0.82 – 0.92)
91-365	0.91	(0.88 – 0.95)	0.94	(0.91 – 0.98)	0.86	(0.86 – 0.92)	0.90	(0.85 – 0.96)
>366	0.93	(0.90 – 0.96)	0.97	(0.94 – 1.00)	0.86	(0.82 – 0.91)	0.93	(0.89 – 0.98)
Number of GP visits (ref: 0)								
1–9	1.06	(1.02 – 1.10)	1.11	(1.06 – 1.16)	1.05	(0.98 – 1.11)	1.17	(1.09 – 1.27)
10–19	0.89	(0.85 – 0.92)	1.06	(1.01 – 1.11)	0.77	(0.73 – 0.81)	1.06	(0.99 – 1.15)
20–29	0.81	(0.77 – 0.84)	1.01	(0.96 – 1.06)	0.67	(0.63 – 0.71)	0.99	(0.91 – 1.08)
≥30	0.77	(0.74 – 0.80)	0.99	(0.94 – 1.05)	0.62	(0.59 – 0.66)	0.96	(0.88 – 1.04)
Socio-economic status (ref: medium)								
Low	0.99	(0.95 – 1.04)	0.99	(0.94 – 1.04)	1.04	(0.96 – 1.12)	1.04	(0.96 – 1.12)
Low-medium	0.95	(0.90 – 0.99)	0.96	(0.91 – 1.00)	0.93	(0.87 – 1.00)	0.94	(0.88 – 1.02)
Medium - high	1.01	(0.96 – 1.06)	1.02	(0.97 – 1.07)	1.02	(0.94 – 1.10)	1.03	(0.95 – 1.12)
High	1.04	(0.99 – 1.10)	1.05	(0.99 – 1.10)	1.08	(0.99 – 1.17)	1.08	(0.99 – 1.18)

Table 3. (continued)

Characteristics	Non-persistence (90-day gap)				Non-adherence (PDC < 0.80)			
	Age and sex adjusted		Fully adjusted*		Age and sex adjusted		Fully adjusted*	
	HR	95% CI	HR	95% CI	OR	95% CI	OR	95% CI
Medication (ref: no)								
Antihypertensive[#]	0.65	(0.63 – 0.67)	0.73	(0.70 – 0.77)	0.54	(0.52 – 0.56)	0.67	(0.63 – 0.71)
Acute gout treatment								
Colchicine	0.86	(0.83 – 0.89)	0.94	(0.90 – 0.97)	0.81	(0.78 – 0.85)	0.95	(0.91 – 1.00)
Corticosteroid	0.93	(0.89 – 0.98)	0.98	(0.93 – 1.03)	0.92	(0.85 – 0.98)	0.99	(0.92 – 1.07)
NSAIDs	1.08	(1.05 – 1.11)	1.05	(1.02 – 1.08)	1.10	(1.06 – 1.14)	1.04	(0.99 – 1.08)
Comorbidities (ref: no)								
Alcoholism	1.04	(0.98 – 1.10)	1.06	(0.99 – 1.13)	1.04	(0.95 – 1.14)	1.06	(0.99 – 1.13)
COPD	0.99	(0.96 – 1.03)	1.02	(0.98 – 1.06)	1.01	(0.96 – 1.07)	1.02	(0.98 – 1.06)
Dementia	0.64	(0.50 – 0.81)	0.58	(0.46 – 0.74)	0.63	(0.49 – 0.82)	0.56	(0.43 – 0.72)
Depression	0.99	(0.95 – 1.02)	1.03	(0.99 – 1.07)	0.96	(0.91 – 1.01)	1.03	(0.97 – 1.08)
Diabetes	0.77	(0.74 – 0.81)	0.94	(0.89 – 0.98)	0.68	(0.64 – 0.73)	0.92	(0.86 – 0.98)
Dyslipidaemia[‡]	0.73	(0.71 – 0.75)	0.87	(0.84 – 0.90)	0.62	(0.60 – 0.65)	0.84	(0.80 – 0.89)
Hypertension	0.77	(0.75 – 0.79)	1.00	(0.96 – 1.03)	0.66	(0.63 – 0.68)	0.95	(0.91 – 1.01)
Ischemic heart disease	0.94	(0.90 – 0.97)	1.04	(1.00 – 1.08)	0.92	(0.88 – 0.97)	1.05	(0.99 – 1.11)
Myocardial infarction	0.90	(0.86 – 0.94)	0.99	(0.94 – 1.05)	0.87	(0.81 – 0.93)	0.99	(0.92 – 1.07)
Osteoarthritis	0.97	(0.94 – 1.00)	1.00	(0.97 – 1.03)	0.96	(0.91 – 1.00)	1.02	(0.97 – 1.07)
Renal calculi	0.94	(0.83 – 1.06)	1.00	(0.88 – 1.13)	0.83	(0.70 – 0.99)	0.92	(0.77 – 1.11)
Stroke	0.96	(0.91 – 1.01)	0.99	(0.94 – 1.05)	1.02	(0.95 – 1.10)	1.07	(0.99 – 1.15)
Renal function[#] (ref: CKD 1)								
CKD 2	1.02	(0.97 – 1.08)	0.98	(0.93 – 1.03)	0.95	(0.86 – 1.05)	1.00	(0.94 – 1.07)
CKD 3	1.07	(1.02 – 1.13)	1.05	(0.99 – 1.11)	0.95	(0.86 – 1.06)	1.04	(0.97 – 1.12)
CKD 4	1.00	(0.90 – 1.11)	1.01	(0.90 – 1.12)	1.01	(0.88 – 1.15)	1.10	(0.97 – 1.25)
CKD 5	1.07	(0.81 – 1.41)	1.14	(0.86 – 1.50)	1.22	(0.90 – 1.64)	1.45	(1.01 – 2.07)

PDC: proportion of days covered; HR: hazard ratio; OR: odds ratio; CI: confidence interval; BMI: body mass index; GP: general practitioner; NSAIDs: non-steroidal anti-inflammatory drug; COPD: chronic obstructive pulmonary disease; eGFR: estimated glomerular filtration rate. * Adjusted for sex, age, BMI, smoking status, alcohol use, socio-economic status at index date, calendar year start allopurinol, number of GP visits 12 months prior the index date, use of antihypertensive, colchicine, corticosteroids, NSAIDs, and statins in the 6 months prior to the index date, and comorbidities ever before the index date (COPD, dementia, depression, diabetes, hypertension, ischemic heart disease, myocardial infarction, osteoarthritis, stroke and renal function), except the variables which is the explanatory variable in the analysis. [#] Follow-up was restricted to 1 year for the Cox-regression analyses, because of violating the proportional hazard assumption.

DISCUSSION

The present study showed poor medication taking behaviour among incident gout patients initiated allopurinol. Non-persistence (90-day gap) with allopurinol therapy increased from 57% at 1 year to 77% at 5 years following initiation. After the occurrence of a gap 54% of the patients restarted therapy; yet less than half of the patients persisted with treatment for 1 year following restarting therapy. During the entire observation period only 38% of the patients were considered adherent.

Non-persistence estimates found in the present study were comparable to the few previous conducted studies, which showed rates between 56% and 88%.[16,18-20] All studies used pharmacy-claims data and three out of four were conducted in the USA.[16,19,20] Despite these differences in population and design, it seems that the variation in non-persistence rates mainly depends on the permissible gap-length and observation period. For example, Solomon et al. showed among mainly female (~80%) and American gout patients that 56% experienced a gap of at least 30-days in the first year after starting allopurinol.[20] Another study conducted among 15,908 Irish gout patients found that 81% experienced a gap of 5 weeks and 54% a gap of 9 weeks in the first year after initiation of uric acid lowering therapies.[18]

The dynamics in medication use is less frequently studied, such as whether patients return to therapy after the occurrence of a gap and if they remain on therapy after restarting. We found that almost half of the patients returned to therapy after a 90-day gap in the first year. This finding is in line with a study conducted by Harrold et al. who showed that 70% had a gap of at least 60-days in therapy and among those with a gap an estimated 50% returned to treatment within 8 months, and by 4 years, 75% had restarted.[16] Although it is encouraging that many patients return to therapy after the occurrence of a first gap, we found that only 50% remained on therapy in the first year after reinitiating. Multiple and extended gaps in therapy lead to less efficiency in achieving the target level of serum uric acid, required to dissolve existing monosodium urate crystals and to prevent further gout flares.[21]

In contrast to persistence, adherence is a measure of medication adherence during the entire observation period. In the present study only 38% of the gout patients were considered adherent. Although, our reliance on a PDC of more than 0.80 to be considered adherent is arbitrary, the mean PDC was 0.57, which is far from optimal. In 2012, a systematic review reported mean adherence rates all below 0.80 and the proportion of adherent patients ranged from 10–46%.[15] This was based on 10 claims/electronic records of which nine were conducted in the US.[15] Recently, two European studies have been conducted. Among Irish patients 35.8% had an MPR ≥0.80 in the first 12 months, using pharmacy claims data.[18] In an Italian study the rates were even more alarming, 10% of the 3727 gout patients were adherent to allopurinol in the first 150 days and just 3% in the first year.[10] Our data from the UK and the studies conducted in Ireland[18] and Italy,[10] indicate that poor medication adherence among gout patients not only occurs in patients from the US but also in Europeans.

To improve medication adherence, patients at-risk should be identified. In our study, females were more prone to have poor medication adherence; this is in line with other previous research,[13,18] while others found the opposite,[8,22,23] or no effect of gender on persistence or adherence.[10,20] In agreement with our findings, older patients and those being overweight[13] and suffering from comorbidities such as, diabetes, and hypertension were more likely to be persistent and adherent.[8,10,13,19,22,24] Although not investigated, these patients appear to be less healthy and might have more severe gout or are aware of the negative consequences of poor medication adherence on their health than younger and healthier gout patients.[11,19,20,25] For example, we showed that users of antihypertensive medication had a 27% lower hazard to be non-persistent (90-day gap) and a 33% lower odds to be non-adherent, compared to non-users. Suffering from overweight appears to decrease the risk by approximately 11% and 14%, for dyslipidaemia 13% and 16%, and for diabetes 6% and 8%, to become non-persistence and non-adherence, respectively. Somewhat contra-intuitively, persons registered with dementia had a lower risk of non-persistence (HR: 0.58) and non-adherence (OR: 0.56), most likely because they will be supervised by taking their medication. Likely, general practitioners register dementia only in the clear and more severe cases. Therefore, it cannot be excluded that person with mild dementia also have a lower risk for poor medication adherence.

The reason for poor medication adherence among gout patients is complex and is probably also attributable to poor illness perception and incorrect medication beliefs.[26-29] Herein, the care provider can contribute by explaining the pathophysiology of the disease, the short- and long-term side effects of urate lowering therapy and the role of life style and diet changes. Nevertheless, the awareness and support from the health providers in medication adherence in those with gout is far from optimal.[30] Up to now, there is only one intervention study in gout patients which investigated the effect of nurse delivered 'package of care', including patient education, lifestyle advice and uric acid lowering therapy, on medication adherence and the recommended treatment target of serum uric acid <360 μmol/L. More than 90% of the gout patients (*n*=106) succeeded in achieving the therapeutic target during 12 months of follow-up.[31] Furthermore, excellent adherence and persistence of four years after the responsibility of treatment is taken over by the patient's general practitioner was achieved.[32] Emphasizing that, as in other chronic diseases, nurse delivery of care is likely to be effective in improving medication adherence and achieving therapeutic targets.[32-34]

This study represents a comprehensive analysis of medication adherence among newly diagnosed gout patients from a representative primary care data-set from the UK. There were certain limitations to this study. First, medication adherence was estimated retrospectively by analysis of allopurinol prescribed by the general practitioner. Therefore, we could not ascertain if patients actually purchased and took the prescribed medication. On the other hand, by using prescription data any distortion caused by patient recall or desire to give social accepted answers can be eliminated[35] and is an acceptable and accurate measure of persistence and adherence.[36] Second, the reason

for stopping or interrupting allopurinol use could not be traced and might be physician-directed. Since the chronic nature of the disease and the low prevalence of the side-effects we do not expect big shifts in our findings. Third, only patients initiated allopurinol were included. A low number of patients switching to probenecid or febuxostat were censored. Likely these drugs are prescribed to patients with different health characteristics, and might receive more attention in the healthcare system influencing their adherence to medication. Third, adherence was defined as a PDC of ≥0.80. Even though the dichotomization of adherence is widely used in the literature, this cut-off is arbitrary and not clinically validated.

In conclusion, our study showed poor medication adherence among incident gout patients initiated allopurinol. The results of the present study further add to the literature, that although patients experiencing a gap in therapy are likely to return, the changes of a backlash are as likely. Highlighting the need for additional research focusing on improving medication adherence among those initiated allopurinol therapy, but also among those restarting therapy after the occurrence of a gap. Clear guidelines and studies investigating means to improve medication adherence are urgently needed to tackle this complex problem. Target groups for intervention may be those who are younger and considered healthier, so with a normal weight and those not on antihypertensive medication and suffering from comorbidities like dementia, depression, dyslipidaemia, and hypertension.

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6

Medication adherence among patients with gout: a systematic review and meta-analysis

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Under revision

ABSTRACT

Objective

In the management of chronic gout, a large proportion of patients need long-term management with urate lowering therapy (ULT). This study reviews medication adherence to ULT and summarizes factors associated with adherence.

Methods

We performed a systematic literature search for studies on adherence to ULT among gout patients in PubMed, Embase, CINAHL, and PsycINFO. We conducted meta-analysis, with a random effect model, for the studies reporting the proportion of patients considered adherent to at least 80% of prescribed medication or time taken. We explored potential sources of heterogeneity, including geographic area and measure of adherence. Narrative summaries were made for data on adherence assessed/defined by Medication Event Monitoring System (MEMS)/pill-count or patient-reported, occurrence of a gap in therapy ≥30 days (non-persistence), and factors associated with adherence.

Results

Of the 23 studies, 15 assessed adherence using prescription/claims data, two by the MEMS or pill count, and six by patient-reported data. The pooled proportion of adherent patients (*n*=13) was 46.0% (95% CI 40.8-51.2); 45% across studies conducted in the USA (*n*=8) and 48% in other countries (*n*=5). Adherence assessed by MEMS/pill count and patient-reported was much higher than by studies using prescription/claims data. Non-persistence (*n*=5) ranged from 56% to 87%. Factors associated with adherence were investigated in 18 studies, strong evidence for a positive association of older age, more comorbidities, and the presence of diabetes or hypertension was found.

Conclusion

Medication adherence to ULT among gout patients was poor. Better insight into reasons and consequences or poor adherence is needed.

INTRODUCTION

Gout is the most common type of inflammatory arthritis, affecting approximately 1–3% of the population worldwide.[1,2] Hyperuricemia, the main risk factor for gout, can be successfully treated with urate lowering therapy (ULT). The initiation of ULT is recommended in patients with recurrent gout attacks or with tophi, as these might lead to debilitating long-term consequences such as joint damage.[3] Despite availability of effective treatments, a substantial proportion of patients experiences multiple gout flares and develops tophi, signifying insufficient control of the disease.[4] One of the barriers towards optimal dug treatment of gout is poor medication adherence of the patient to ULT.

Medication adherence refers to the process by which patients take their medications as prescribed.[5] It is a complex construct and contains several components, including *adherence* and *non-persistence*. Adherence is usually defined as the extent to which a patient's actual medication intake corresponds to the prescribed dosing regimen during observation time. Non-persistence is defined as the occurrence of a gap in taking therapy (e.g. for at least 30- or 60-days), or stopping.

In 2014, a systematic review of 16 studies by De Vera et al. revealed poor overall medication adherence to ULT among gout patients.[6] The proportion of patients considered adherent ranged from 56% to 72%. Non-persistence ranged from 56% to 88%.[6] However, 12 of 16 studies were conducted in the USA, hampering transferability of results to the European setting. Furthermore, data were often derived from electronic prescription records and managed health care plans, which might have introduced selection bias and misclassification. Since then, several new studies were published, also from outside the USA and using others methods than prescription/claim data to assess medication adherence.

The aims of the present study were (i) to perform a systematic review and meta-analysis on studies that assess medication adherence and/or non-persistence among gout patients treated with ULT, (ii) to perform subgroup analyses on demographical and methodological characteristics that might explain heterogeneity, and (iii) to present an overview of studies addressing the association of medication adherence with explanatory variable factors and with gout related clinical outcomes.

METHODS

A systematic literature review was conducted of primary studies investigating medication adherence to ULT in patients with gout. The search was performed in PubMed, Embase, PsycINFO, and CINAHL databases (inception to August 15th 2016) and combined free text- and keyword-search terms relating to gout and hyperuricemia, ULT, and medication adherence (Appendix 1: search strategy). In addition, a hand search of references of relevant articles was performed. Studies not published in English language,

letters, editorials, reviews, case reports, opinion articles, congress abstracts and randomized controlled trials were excluded. The latter are assumed to not reflect accurately real-world patterns of patient medication adherence. For final inclusion, studies had to meet the following criteria: (i) presenting primary data from an original research study; (ii) concerning patients with evidence of gout or hyperuricemia; (iii) being prescribed or taking ULT; and (iii) reporting on at least one measure of medication adherence. No restrictions on the methods to assess medication adherence was made.
A two-stage screening process was performed. First, titles, index terms, and/or abstracts were screened to identify potentially relevant articles. If in doubt, the full article was evaluated in the second step. Both steps were independently performed by two reviewers (LS and MO) using a standardized protocol and reporting form. Difference in assessment was resolved by arbitration of a third reviewer (AB), and consensus was reached after discussion.

Data extraction

Data extraction was performed by one reviewer using a standardized data extraction form (LS), and was checked by a second reviewer (MO). Data extraction included: study identification (first author, year of publication), study characteristics (study design, setting, recruitment site, country, inclusion period, inclusion criteria, sample size, observation time for adherence), patient characteristics (average age of patients, percentage of males), case definition of gout, type of ULT, and medication adherence.
With regard to the assessment of medication adherence the following information was extracted, the method for assessing medication adherence (using prescription/claims data, patient-reported etc.), the component of medication adherence studied (adherence, non-persistence, time until non-persistence etc.), the type of measurement used (medication possession ratio (MPR) [sum of days' supply / days of observation], proportion of days covered (PDC), which is more conservative that the MPR since a possible overlap between two prescriptions is truncated),[7] and estimate and corresponding threshold to define the outcome (mean adherence, proportion of patients with a PDC or MPR of at least 80%, gap of at least 30 days etc.).

Quality assessment / risk of bias

Quality scoring of observational studies is controversial because the proposed criteria may not be relevant for the study question or for the actual quality of the study.[8] Moreover, the current review included 16 prescription/claims studies, which have a similar risk of bias with regard to generalisability, absence of information on drop-out, misclassification of disease and exposure and assessment of outcome. Therefore, we have chosen to discuss the results in the light of aspects related to risk of bias and factors which might influence medication adherence such as study population and the method to assess medication adherence.

Meta-analysis

Since for the majority of studies the proportion of patients adherent to ULT was based on a threshold of 80%, the results could be pooled by performing a meta-analysis. To ensure consistency in the interpretation of the outcome, the figures of studies reporting proportion non-adherent were converted to present proportion non-adherent to ULT. Heterogeneity was tested by Cochran's Q test and the I^2 statistic, the latter describing the percentage of variation across studies.[9] Whenever heterogeneity was high (I^2 >50%), random effects models were used.[10] Potential sources of heterogeneity were further explored by stratification for (1) studies conducted in the USA opposed to other countries and (2) differences in the measure of adherence (PDC or MPR).
Further narrative summaries were provided for studies reporting adherence using another type of measure/numeric value and for the other outcomes, including non-persistence, the association between medication adherence and factors and clinical gout related outcomes.

Factors associated with medication adherence

If available, information on factors associated with medication adherence was extracted from each study. Consistent with the World Health Organization (WHO) framework, factors were categorized into patient-, condition-, therapy-, social/economic-, and health care system factors. Factors were considered associated with medication adherence based on significance (P-value <0.05) and the direction of association was reported as being positively, negatively or no association. The strength of evidence across studies was classified into four levels of evidence, according to recommendations by Van Tulder et al.[11] The level of evidence was summarized as follows: 1) strong: ≥four studies and ≥75% of all studies considered report findings in the same direction; 2) moderate: three studies and two report findings in the same direction; 3) limited: only one or two studies and ≥75% of all studies reported findings in the same direction; 4) conflicting: ≥75% of the studies report inconsistent findings.
Furthermore, the association between medication adherence and gout related outcomes, including serum uric acid and the number of gout flares was registered.

RESULTS

The search resulted in 1991 potentially relevant articles (Figure 1). After excluding duplicates and screening of title, index terms, and/or abstract, 98 articles remained for full-paper review. After reading the full-text, a total of 23 articles were included. Hand search of references did not reveal additional articles. Because of the different methods to assess medication adherence, the characteristics of the studies were presented according to the type of method, i.e. prescription/claims, MEMS/pill count or patient-reported data (Table 1).

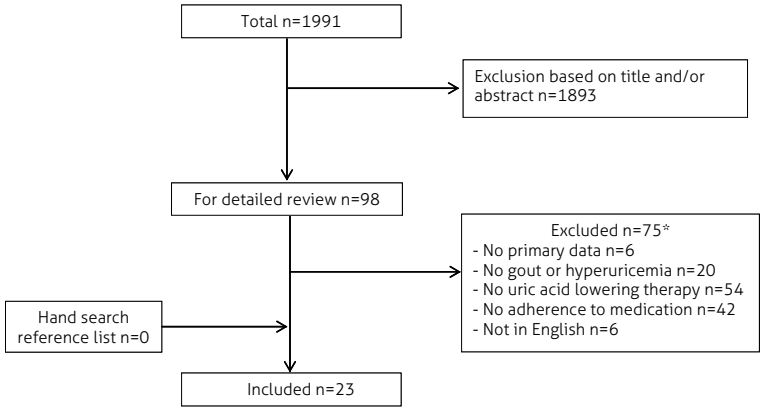

Figure 1. Flow-diagram of systematic literature search
* Numbers are not mutually exclusive

Characteristics of included studies

Prescription/claims data

Sixteen studies used prescription/claims data, 10 (64%) were conducted in the USA, one in Israel,[24] one in New Zeeland[12] and three in Europe (UK,[2] Italy,[13] and Ireland[14]). All of them made use of claims or pharmacy data, except the studies from Italy and UK which used records in primary care databases. Patients were classified as having gout by either a gout diagnosis (ICD- or read-codes) or a dispensing record for ULT.[12,14] Study sample sizes ranged from 242[15] to 49,395 patients.[2] Adherence was defined as either a MPR (*n*=7) or PDC (*n*=7) of more than 80%.

Electronic monitoring device or pill count

Of the two studies retrieved, one was conducted in The Netherlands and used MEMS to assess mean adherence (number of patients=17). The diagnosis of gout was based on physician opinion.[16] The other study, conducted in Korea, used pill count by the nurse to assess adherence, (number of patients=132). The diagnosis of gout was based on a physicians' assessment of the Wallace classification criteria.[17] Adherence was defined as at least 80% of the prescribed dose was not returned. Both studies included patients from a Rheumatology centre.

Table 1. Characteristics of the studies included in the systematic review

Author, Country, (Publication date)	Study design and setting	Recruitment site	Inclusion period (years)	Inclusion criteria	Observation time	Case definition of gout	Medication adherence		
							Adherence	Non-persistence	According to gout related outcome
Prescription / claims database									
McGowan, Ireland (2016)[14]	Cohort (retrospective pharmacy claims database)	Irish Health Service Executive - national Pharmacy claims database	2008 - 2012 (5)	No ULT prescription 12 months prior to index date	First 6 and 12 months after initiation	Dispensing record of allopurinol, febuxostat, probenecid, or sulfinpyrazone	MPR ≥0.80	Gap of 5 or 9 weeks	-
Rashid, USA (2015) (A)[28]	Cohort (retrospective claims database)	Kaiser Permanente Southern California (KPSC)	2007 - 2011 (5)	Age ≥18 years, no allopurinol dispensing 12 months prior to index date	12 months after initiation	Two ICD-code for gout more than ≥30 days apart at any out- or inpatient visit	PDC ≥0.80	-	-
Rashid, USA (2015) (B)[32]	Cohort (retrospective claims database)	Kaiser Permanente Southern California (KPSC)	2007 - 2010 (4)	Age ≥18 years, no ULT prescription 12 months prior to index date	12 months after initiation	Two outpatient ICD-codes for gout ≥30 days apart or one inpatient gout diagnosis	-	-	Percentage of adherent (PDC ≥0.80) according to number of flares
Mantarro, Italy (2015)[13]	Cohort (retrospective general practitioner database)	General practitioner database	2002 - 2011 (10)	Age ≥18 years, 1 year medical history recorded and ≥6 months of FU	Up to 12 months of FU	ICD-code for gout *or* tophi in verbatim in free-text	PDC ≥0.80 for 3 semesters: (1) day 30 - 89; (2) day 90 - 149; (3) day 150 - 365	-	-
Horsburgh, New Zeeland (2014)[12]	Cohort (retrospective community pharmacy database)	Community pharmacies in the Gisborne region	2005 - 2006 (1)	Age ≥25 years, receive allopurinol in the first 6 months of FU and supply of >90 days	12 months	Dispensing records of allopurinol	MPR ≥0.80	-	-
Kuo, UK (2014)[2]	Cohort (retrospective general practitioner database)	Medical records from primary care	1997 - 2012 (16)	All participants who contributed to the CPRD database	Each calendar year	Read codes for gout	PDC ≥0.80	-	-

Table 1. (continued)

Author, Country, (Publication date)	Study design and setting	Recruitment site	Inclusion period (years)	Inclusion criteria	Observation time	Case definition of gout	Medication adherence		
Zandman-Goddard, Israël (2013)[24]	Cohort (retrospective claims database)	HMO	2002 - 2009 (7)	Age ≥25 years, incident gout patients	At least 12 months	ICD-code for gout and diagnosed by a rheumatologist	PDC ≥0.80	Time to insufficient supply to cover 80% of FU + 30 days supply	Risk of non-adherence according to sUA level
Park, USA (2012)[15]	Cohort (retrospective claims database)	Commercial and Medicare members	2005 - 2010 (5.5)	Age ≥18 years, first gout-related pharmacy claim	12 months after initiation	ICD-code for gout or pharmacy claim for gout with ≥2 sUA tests within 1 year	-	-	Time to a 30- or 60-day gap according to sUA level
Halpern, USA (2009)[29]	Cohort (retrospective claims database)	Managed care enrolees	2002 - March 2004 (approx. 2)	Enrolment ≥12 months before and after index date (first gout- related claim)	12 months	≥2 ICD-codes for gout *or* ≥1 code and a pharmacy claim for allopurinol, probenecid, colchicine, or sulfinpyrazone	MPR ≥0.80	-	Adherence according to sUA level
Harrold, USA (2009) (A)[40]	Cohort (retrospective claims database)	HMO	2000 - 2006 (6.5)	Age ≥18 years at time of first ULT dispensing and no of ULT 6 months prior investigation	12 months	ICD-code for gout	MPR ≥0.80	-	-
Harrold, USA (2010) (B)[31]							-	Gap of 60-days; Time until non-persistence; Percentage returning to ULT after gap	-
Solomon, USA (2008)[27]	Cohort (retrospective claims database)	Enrolled in Medicare system + pharmacy benefit for older low-income adults in state Pennsylvania	Unspecified	Age ≥65 years, no ULT pharmacy claim 12 months prior to index date	Up to 12 months after initiation	Filling record for allopurinol, probenecid, or sulfinpyrazone	PDC ≥0.80	Gap of 60-days; Time until non-persistence	-

Table 1. (continued)

Author, Country, (Publication date)	Study design and setting	Recruitment site	Inclusion period (years)	Inclusion criteria	Observation time	Case definition of gout	Medication adherence		
Briesacher, USA (2008)[25]	Cohort (retrospective claims database)	MarketScan, employers sponsored medical care	2001-2004 (3)	No pharmacy claim 12 months prior to index date	At least 12 months	ICD-code for gout	MPR ≥0.80	-	-
Sarawate, USA (2006)[30]	Cohort (retrospective claims database)	HMO	2000-2003 (3)	Age ≥18 years, no pharmacy claim 12 months prior to index date	At least 12 months	≥2 visits with ICD-code for gout or ≥1 gout pharmacy claim	MPR ≥0.80	No refill with 1.5 day's supply previous prescription; Time to non-persistence; Percentage of reinitiating	
Riedel, USA (2004)[26]	Cohort (retrospective claims database)	HMO, employers sponsored medical care	1997-1998 (1)	Continuous enrolment for ≥24 months after index date and ≥2 allopurinol prescriptions (study population)	At least 24 months	ICD-code for gout *or* 1 prescription for ULT or colchicine	Taking medication ≥0.80	-	-
Electronic monitoring devices / pill count									
Lee, Korea (2016)[17]	Cohort (prospective observational cohort)	Rheumatology centre	Unspecified	Male, age ≤75 years, no ULT prescription 12 months prior to index date	12 months	Wallace classification criteria	Pill count by nurse. Adherent: ≥80% of pills taken	Gap of 30-days	-
De Klerk, The Netherlands (2003)[16]	Observational prospective cohort study	Rheumatology outpatient clinical	Unspecified	First prescription of ULT	12 months or less if patient or rheumatol-ogist stopped medication	Rheumatologist diagnosed gout	MEMS®: Mean adherence: (referred to as taking compliance), the percentage of prescribed dose taken	-	-

Table 1. (continued)

Author, Country, (Publication date)	Study design and setting	Recruitment site	Inclusion period (years)	Inclusion criteria	Observation time	Case definition of gout	Medication adherence		
Patient-reported adherence									
Singh, USA (2016)[23]	Cross-sectional study	Visitors of website gouteducation.org	Aug 2014 - Apr 2015 (8 months)	Patient reported that gout was diagnosed by a physician	1 month	Patient reported that gout was diagnosed by a physician	Patient-reported Adherent: >80% medication used in last 30 days	-	-
Van Onna, The Netherlands (2014)[22]	Cross-sectional study	Rheumatology outpatient clinical and primary care practices	Sep 2013 - Feb 2014		Unspecified	Physician diagnosed	Semi-structured interview. Adherent: medication taken on a daily routine	-	-
Martini, New Zealand (2012)[20]	Cross-sectional data	Community pharmacies, region Auckland and Waikato (mainly from Māori and Pacific)	2010 (July)	Patient believed he/she had gout and could communicate in English	Unspecified	Prescription allopurinol / colchicine + patient believed he/she had gout	Semi-structured interview. Adherent: not specified	-	-
Dalbeth, New Zealand (2012) (A)[18]	Cross-sectional data from a cohort study	Community advertising (primary and secondary care clinics)	Unspecified	First gout attack in <10 years	Information on long-term regimes	Physician diagnosed and gout defined by the Wallace criteria		-	Mean MARS score (10-item questionnaire) according to sUA level
Dalbeth, New Zealand (2011) (B)[19]	Cross-sectional data from a cohort study	Community advertising (primary and secondary care)	Unspecified	First gout attack in <10 years	Information on long-term regimes	Wallace criteria and EULAR recommendations (2006)	Mean MARS score: 10-item questionnaire	-	-
Silva, Spain (2010)[21]	Observational prospective study	Participants in a prospective study on ultrasound evaluation of gout	2008 - 2010 (2)	FU at 1, 3, 6, 9 and 12 months from the first visit (required for primary study aim)	During 12 months after 1st evaluation	Wallace classification criteria	Patient-reported. Adherent: taking medication regularly, as prescribed.	-	Non-adherence according to sUA level

Approx.: approximately; CPRD: clinical practice research datalink; EULAR: European League Against Rheumatology; FU: follow-up; HMO: health maintenance organization; ICD: international classification of diseases; KPCS: Kaiser Permanente Southern California; MARS: medication adherence report scale; MEMS®:medication event monitoring system; MPR: medication possession ratio; PDC: proportion days covered; sUA: serum uric acid; ULT: uric acid lowering therapy.

Patient-reported data

Six studies used patient-reported data to assess medication adherence, three were conducted in New Zeeland,[18-20] two in Europe (Spain[21] and The Netherlands[22]), and on in the USA.[23] The diagnosis of gout was either patient self-reported[20,23] or physician reported. The latter could either be based on the physicians opinion[22] or by using of the Wallace classification criteria.[18,19,21] Sample sizes were considerably smaller, ranging from 15[22] to 251.[23] To measure medication adherence, the two studies from New Zealand had administered the validated nine-item Medication Adherence Report Scale (MARS) questionnaire, which provides a mean adherence estimate.[18,19] A higher score (max 45) represents better adherence. Adherence for the four remaining studies was reported as the proportion of patients indicating to take medication as a daily routine (*n*=2),[21,22] taking at least 80% of their medication (*n*=1),[23] or considered himself adherent (yes/no) (*n*=1).[20]

Medication adherence

Adherence

In 13 studies the proportion of patients considered adherent was based on the threshold of being covered for at least 80% during observation time. Adherence ranged from 17%[24] to 78%[12] (median=37%[25]) in 11 prescription/claims studies, was 71%[17] in the study using pills count by the nurse, and was 79% in the study using patient-reported adherence.[23] In the meta-analysis, the pooled proportion of patient adherent, in the random effects model was 46.0% (95% confidence interval (CI) 40.8-51.2) with a substantial heterogeneity observed (Q=3551, *P*-value <0.001, I^2=99%) (Table S1). Figure 2 shows a forest plot with the estimated proportion of patients considered adherent. In a stratified meta-analysis (Table S2), the pooled proportion of patients adherent was similar across studies conducted in the USA (*n*=8; 45.2%, CI 39.8-50.6) and other countries (*n*=5; 47.5%, CI 36.5-58.6). The pooled proportion of patients adherent was higher in studies using MPR (*n*=6; 44.1%, CI 37.6-50.7) compared to studies using PDC (*n*=4; 33.5%, CI 24.4-43.2). Three of the 13 studies did not specify the type of measure (PDC/MPR) and were therefore not included in the stratified meta-analysis according to PDC or MPR.[17,23,26]

Mean adherence was reported in seven studies and ranged from 54%[26] to 88%.[12,16,25-29] Because of the low number of studies presenting mean adherence and often missing data on the according standard deviations, we did not pool the results for mean adherence.

For the five remaining studies the proportion of patients adherent was not based on the threshold of 80%, and were therefore not included in the meta=analysis. Adherence was defined as taking medication "on a daily routine",[22] "on a regular basis as prescribed"[21] and one study did not define when a patient was considered adherent.[20] Adherence rates in these studies ranged from 40%[20] to 79%.[19] Finally, two studies used the MARS

questionnaire to assess adherence, but only the mean scores were presented ranging from 39.0 (SD 7.4) to 4.2 (SD 2.7), 45 is considered highly adherent.[18,19]

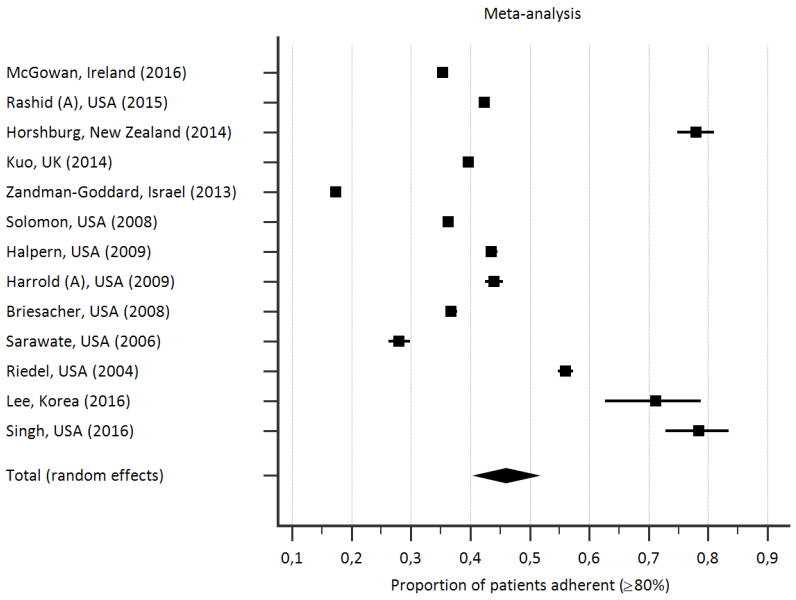

Figure 2. Forest plot: pooled estimate of proportion of patients adherent

Non-persistence

Five studies reported non-persistence, and rates ranged from 56%[27] to 87%.[14,17,30,31] Time until non-persistence was investigated in three studies and ranged from 4 to 12 months.[24,30,31] The variation in outcomes is mostly attributable to the differences in the permissible gap length and observation period (Table 2). Due to large differences in defining non-persistence and the low number of studies, it was inappropriate to create a pooled estimate.

Factors associated with medication adherence

Table 3 summarizes factors associated with medication adherence among 18 studies. We found strong evidence for better medication adherence in patients who are older, had a higher number of comorbidities, and suffered from diabetes or hypertension. In addition, patients from African-America and Māori descent, were more likely to have poor medication adherence (moderate evidence).[12,27,28] Several factors were infrequently studied and therefore no conclusions on the role of factors as smoking status, body mass index, socio-economic status could be made. The same applies to other condition- or patient-related factors, such as perceived health status or understanding illness, these aspects were only investigated by single studies.[16,17,19,23]

Table 2. Adherence to medication, including adherence, non-persistence and the association with uric acid concentration or gout flares

Author	Sample size (male)	Mean age years (SD)	Drugs studied	Medication adherence		
				Adherence	Non-persistence	Medication adherence according to gout related outcomes
Prescription/claims data						
McGowan[14]	(i) Gout patients: *n*=34,634 (73%) (ii) Study population: *n*=15,908	(i) 65.2	Allopurinol, febuxostat, probenecid, and sulfinpyrazone.	Adherent (MPR>0.80): 46% first 6 months of FU 36% first 12 months of FU	Non-persistence (5-weeks gap): 59% first 6 months of FU 81% first 12 months of FU Non-persistence (9-weeks gap) 54% first 6 months of FU; 77% first 12 months of FU	
Rashid (A)[28]	(i) Gout patients with ≥1 prescription: *n*=13,341 (78%) (ii) ≥2 prescriptions: *n*=10,991 (iii) sUA measurement available at baseline and follow-up: *n*=9,581	(i) 60 (14)	Allopurinol	Adherent (PDC>0.80): (i) 33%; mean 0.65 (SD 23) (ii) 42%; mean 0.74 (SD 21)		Achieving sUA <6.0 mg/dL: Adherent patients were 2.5-fold more likely than non-adherent patients to achieve the target
Rashid (B)[32]	8,828 (80%)	Unspecified	Allopurinol, febuxostat, and probenecid.			Percentage of patients adherent (PDC >0.80) according to number of flares: 0 flares: 74% 1 - 2 flares: 39% >3 flares: 28%
Mantarro[13]	(i) Gout patients: *n*=3,727 (80%) (ii) Study population: *n*=3,570	(i) 65	Allopurinol	Adherent (PDC>0.80) according to FU time in 1st year: 0–29 days: 45.9% 30–89 days: 16.7% 90–149 days: 10.0% 150–365 days: 3.2%		Odds ratio for risk of hyperuricemia among adherent patients, according to FU time in 1st year: 0 - 29 days: OR 0.49 (CI 0.33 – 0.73) 30 - 89 days: OR 0.40 (CI 0.24 – 0.67) 90 - 149 days: OR 0.23 (CI 0.15 – 0.34)
Horsburgh[12]	(i) Gout patients: *n*=953 (80%) (ii) Study population: *n*=732	Unspecified	Allopurinol	Adherent (MPR>0.80): 78%; mean MPR: 0.88		
Kuo[2]	1997: unspecified 2012: *n*=49.395	Unspecified	Allopurinol, febuxostat, Benzbromarone, probenecid or sulfinpyrazone.	Adherent (PDC≥0.80) in 1997: 28% (CI 27.3–29.3) Adherent (PDC≥0.80) in 2012: 39.7% (CI 39.1–40.2) PDC 0.20–0.80: 43% PDC<0.20: 18%		

Table 2. (continued)

Author	Sample size (male)	Mean age years (SD)	Drugs studied	Medication adherence		
				Adherence	Non-persistence	Medication adherence according to gout related outcomes
Zandman-Goddard[24]	7.644 (72%)	Unspecified	Allopurinol	Adherent (PDC≥0.80): 17% PDC>0.20–0.80: 36% PDC<0.20: 47%	Days to non-persistence (30-day gap): Men: 358 Women: 379	Per 1 mg/dl increase in sUA lower risk of non-adherence (PDC ≥0.80) (OR 0.99; 95% CI 0.99 - 0.99)
Park[15]	(i) Gout patients: *n*=352 (72%) (ii) Study population: *n*=242	61 (?)	Allopurinol, febuxostat, probenecid, colchicine, probenecid /colchicine or fixed-dose combination		Days to non-persistence according to sUA level: 30-day gap (*P*-value: 0.032) 1) 215 (SD 150) 2) 141 (SD 133) 3) 139 (SD 132) 60-day gap (*P*-value: 0.008) 1) 255 (SD 136) 2) 164 (SD 136) 3) 178 (SD 136)	Adherent (PDC ≥0.80) according to sUA level: 1) sUA <6 mg/dL: 56%; mean 0.73 2) sUA 6 – 9.0 mg/dL: 24%; mean 0.46 3) sUA ≥9.0 mg/dL: 22%; mean 0.50
Solomon[27]	9,823 (28%)	79 (7)	Allopurinol, probenecid, sulfinpyrazone	Adherent (PDC≥0.80): 36% PDC<0.65: 56% PDC<0.50: 46% Mean: 0.54 (SD 0.36) Known history of gout (*n*=1,391): Mean PDC: 0.54 (SD 0.35) Initiated allopurinol treatment (*n*=10,124): Mean: 0.54 (SD 0.36)	Non-persistence (30-day gap) in 1st year: 56% Non-persistence after 1st prescription: ±25%	
Halpern[29]	(i) Gout patients: *n*=18,243 (84%) (ii) Study population: *n*=10,070	(i) 53.9 (13.5)	Allopurinol	Adherent (MPR ≥0.80): 44% Mean: 0.62 (median 0.71)		Percentage adherent according to sUA <6.0 mg/dL and time (n= 1,473): 45.6% 1) 30-89 days: 49.3% of the adherence users had sUA <6.0 mg/dL *vs* 27.8% of non-adherence users 2) 90-149 days: 51.8% *vs* 22.5% 3) >150 days: 56.8% *vs* 23.8%

Table 2. (continued)

Author	Sample size (male)	Mean age years (SD)	Drugs studied	Medication adherence		
				Adherence	Non-persistence	Medication adherence according to gout related outcomes
Harrold (A)[40]	4,166 (75%)	62 (14)	Allopurinol, probenecid, sulfinpyrazone	Adherent (MPR>0.80) in 1st year: 44% Median MPR: Allopurinol: 0.68 (IQR 0.64) Probenecid: 0.49 (IQR 0.70)		
Harrold (B)[31]					Non-persistence (60-day gap): 70% Days until non-persistence: Median: 120 (IQR 170) Gap in year 1: 75% Gap during study period 1 gap: 54%; 2 gaps: 25% ≥3 gaps: 22% Return to treatment Within 8 months: 50% Within 4 years: 75%	
Briesacher[25]	9,715 (77.5%)	58.7 (0.14)	Allopurinol, uricosurics	Adherent (MPR>0.80): 37% MPR 0.60-0.79: 11% MPR 0.40-0.59: 12% MPR 0.20-0.39: 16% MPR 0.00-0.19: 24% Mean: 0.56		
Sarawate[30]	(i) Gout patients: *n*=5,942 (76%) (ii) Study population, any gout medication: *n*=4,635 (iii) Study population, allopurinol use: *n*=2,405	(i) 57 (14)	Allopurinol, probenecid, sulfinpyrazone	Adherent (MPR>0.80) (iii): 28%	Non-persistence: Any gout medication (ii): 87.9% Allopurinol users (iii): 87% Months to non-persistence: Mean: 8.5 [SD 11.0] Restart therapy: 1,501 of the 2,094 (71.7%)	

Table 2. (continued)

Author	Sample size (male)	Mean age years (SD)	Drugs studied	Medication adherence		
				Adherence	Non-persistence	Medication adherence according to gout related outcomes
Riedel[26]	(i) Gout patients: *n*=9,482 (82%) (ii) Study population: *n*=5,597	51 (11)	Allopurinol	Adherent (taking medication ≥0.80): 56% PDC 0.30-0.80: 29% PDC<0.30: 15% PDC≤0.10: 20% Mean rate: 0.75 (median 0.84)		
Electronic monitoring devices / pill count						
Lee[17]	132 (100%)	51.9 (10.4)	Allopurinol, febuxostat	Adherent (used pills>80%): 71%	Non-persistence (30-day gap): 61%	
De Klerk[16]	(i) Gout patients: *n*=29 (80%) (ii) Study population, ULT user: *N*=17	58 (12)	Allopurinol, benzbromarone	Mean adherence: 0.84		
Patient-reported medication adherence						
Singh[23]	(i) Gout patients: *n*=499 (74%) (ii) Study population: *n*=251	(i) 56.3 (12.6)	Allopurinol, febuxostat	Adherent (>80% medication used): 78.5%		
Van Onna[22]	15 (93%)	63 (12)	ULT, colchicine, NSAIDs	Adherent: 73%		
Martini et al.[20]	(i) Gout patients: *n*=60 (90%) (ii) Study population, ULT users: *N*=56	61 (range 23-93)	Allopurinol, colchicine	Semi-structured interview Adherent: 79% Among those with knowledge on ULT (*n*=25): 88%		
Dalbeth (A)[17]	(i) Gout patients: *n*=273 (ii) Study population: *n*=181	sUA (mmol/L) <0.36: 61 (14) sUA ≥0.36: 58 (16)	Allopurinol, Probenecid			Mean MARS score according to sUA level: sUA level <0.36 mmol/L: 42.7 (SD 2.7) sUA level ≥0.36 mmol/L: 39.0 (SD 7.4)
Dalbeth (B)[19]	(i) Gout patients: *n*=142 (ii) Study population: *n*=105	57 (range: 19-85)	ULT, unspecified	Mean MARS score: 39.8 (SD 6.8)		sUA was inversely correlated with adherence (r -0.33, *P* 0.003)

Table 2. (continued)

Author	Sample size (male)	Mean age years (SD)	Drugs studied	Medication adherence		
				Adherence	Non-persistence	Medication adherence according to gout related outcomes
Silva[21]	34	57.1 (11.8)	Allopurinol, Benzbromarone, colchicine, NSAID, corticosteroid	Adherent: Baseline: Dropouts (*n*=9): 40% On protocol (*n*=25): 17% Final visit (at 1 year FU): Dropouts (*n*=9): 32% On protocol (*n*=25): 60%		37% of the adherent users had urine UA <250 mg/day *vs* 0% of non-complaint users

CI: confidence interval; IQR: interquartile range; MPR: medication possession ratio; NSAIDs: nonsteroidal anti-inflammatory drugs; OR: odds ratio; PDC: proportion days covered: SD: standard deviation; sUA: serum uric acid

Table 3. Determinants associated with better medication persistence or adherence

Category	Specific factor	Positive	Not significant	Negative	Level of evidence
Social – economic	Age (older)	Zandman[24 c] Solomon[27 c] Riedel[26 c] Harrold (A)[40 c] Briesacher[25 a] Silva[21 ^ a] Harrold (B)[31 a] Martini[20 a] Lee[17 a] McGowan[14 a] Rashid (A)[28 c]	Mantarro[13 c] Singh[23 c]	Horsburgh[12 b]	Strong, positive
	Gender (male)	Zandman[24 c] McGowan[14 a]	Solomon[27 c] Briesacher[25 a] Mantarro[13 c] Singh[23 c] Horsburgh[12 b] Harrold (A)[40 c] Rashid (A)[28 c]	Riedel[26 c]	Conflicting
	Race - African –American / minority race *vs* Caucasian - Māori *vs* non-Māori			Solomon[27 c] Rashid (A)[28 c] Horsburgh[12 b]	Moderate, negative
	Socio-economic status (low *vs* high)		Horsburgh[12 b]	Zandman[24 c]	Conflicting
	Marital status (yes *vs* no)	Zandman[23 c]	Lee[17 a]		Conflicting
	Income (low *vs* high)		Lee[17 a]		Limited, no association
Condition – related	Diagnosis (newly *vs* previously)		Lee[17 a]	Sarawate[30 c]	Conflicting
	Gout flares (more *vs* less)	Mantarro[13 c] Sarawate[30 c]		Rashid (B)[c] Silva[21 a]	Conflicting
	Tophaceous gout		Lee[17 a]	Solomon[27 c]	Conflicting
	Hospitalization (more vs less)	Solomon[276 c]	Harrold (A)[40 c] Mantarro[13 c]		Conflicting
	Health care utilization (more vs less)	Harrold (A)[40 c]			Limited, positive
	Alcohol consumption (yes *vs* no)		Lee[17 a]		Limited, no association
	Comorbidity (more vs less)	Solomon[27 c] Harrold (A)[40 c] Harrold (B)[31 a] Briesacher[25 a] Martini[7 a] Lee[17 a] McGowan[14 a]			Strong, positive
Comorbiditie s (yes vs no)	Alcohol-related diseases		Mantarro[13 c]		Limited, no association
	Anaemia		Mantarro[13 c]		Limited, no association
	Arrhythmias		Mantarro[13 c]		Limited, no association
	Cancer history		Zandman[24 c]		Limited, no association
	Cardiovascular disease	Zandman[24 c]	Lee[17 a]		Conflicting
	Chronic kidney disease	Lee[17 a]		Rashid (A)[28 c]	Conflicting

Table 3. (continued)

Category	Specific factor	Positive	Not significant	Negative	Level of evidence
	Diabetes	Zandman[24 c] Riedel[26 c] Rashid (A)[28 c] Mantarro[13 c]	Lee[17 a]		Strong, positive
	Heart failure	Rashid (A)[28 c]	Mantarro[13 c]		Conflicting
	Hypertension	Zandman[24 c] Riedel[26 c] Sarawate[30 c] Lee[17 a] Mantarro[13 c] Rashid (A)[28 c]			Strong, positive
	Dyslipidaemia		Lee[17 a] Mantarro[13 c]		Limited, no association
	Myocardial infarction	Rashid (A)[28 c]			Limited, positive
	Nephrolithiasis		Lee[17 a]		Limited, no association
	Osteoarthritis	Zandman[24 c]	Mantarro[13 c]		Conflicting
	Rheumatoid arthritis		Mantarro[13 c] Zandman[24 c]		Limited, no association
	Use of thiazide / other diuretics	Rashid (A)[28 c]	Mantarro[13]		Conflicting
Therapy – related	ULT type (febuxostat *vs* allopurinol)	McGowan[14 a]	Lee[17 a]		Conflicting
	Drug prescription (specialist *vs* generalist)	Solomon[27 c] Rashid (A)[28 c]			Limited, positive
	Drug doses (higher *vs* lower)	McGowan[14 a]	Lee[17 a]	Riedel[26 c] Rashid (A)[28 c]	Conflicting
	Side effects (yes *vs* no)		De Klerk[16 a]		Limited, no association
	Use of prophylactic (yes *vs* no)		Mantarro[13 c] Lee[17 a]	Harrold (A)[40 c]	Conflicting
Patient – related	Functional capacity, HAQ (greater *vs* poorer)		De Klerk[16 a]		Limited, no association
	Overall health profile (greater *vs* poorer)		De Klerk[16 a]		Limited, no association
	Coping, UCL score (more *vs* less)		De Klerk[16 a]		Limited, no association
	Perceived health status (high *vs* low)			De Klerk[16 a]	Limited, negative
	Previous history of non-persistence to ULT (yes *vs* no)		Lee[17 a]		Limited, no association
	Preference for non-pharmacological treatments (yes *vs* no)		Singh[23 b]		Limited, no association
	Understanding illness (greater *vs* poorer)	Dalbeth (B)[19 a]			Limited, positive
	Body mass index (high *vs* low)	Zandman[24 c]	Lee[17 a] Mantarro[13 c]		Conflicting
	Smoking (current *vs* never)		Lee[17 a] Mantarro[13 c]	Zandman[24 c]	Conflicting

* Positive association indicates better medication adherence; # Associated with adherence, but not with persistence; ^ There was a trend for younger age; a crude; b sex, age, race and socioeconomic status adjusted (if applicable); c multivariable adjusted analyses.

For six[24] out of seven studies a better adherence was associated with lower serum uric acid concentration.[13,18,19,21,24,28,29] One other study showed that time until non-persistence was longer for those patients with a serum uric acid concentration above 6 mg/dL compared to those who had a serum uric acid of less than 6 mg/dL.[15] Finally, one study found that patients with zero gout flares 74% were considered adherent (PDC ≥80%) during 12 months follow-up, while patients with 1–2 or ≥3 gout flares were 39% and 28% adherent, respectively.[32]

DISCUSSION

Our study shows a large variation in medication adherence to ULT among patients with gout. Poor medication adherence was found in studies using prescription/claims data, whereas studies using MEMS/pill count or patient-reported data showed considerable better adherence rates. In a subgroup of studies (n=13) that reported the proportion of patients adherent based on the commonly used threshold of 80%, meta-analysis revealed a pooled adherence of only 46%. The proportion of patients being non-persistent, examined in five studies using prescription/claims data, was even lower and showed rates ranging from 56%[27] to 87%.[30] Consistent strong evidence was found that elderly patients, the presence of hypertension and/or diabetes, and those with more comorbidities were more prone to be adherent. Finally, poor medication adherence was associated with higher number of gout flares and an elevated serum uric acid.

Our findings are overall in concordance with a previous systematic literature review of 16 studies from De Vera et al.[6] We expanded upon that review by adding seven new studies, of which six were conducted outside the USA, and by performing a meta-analysis. Similar results were found across studies conducted inside and outside the USA for which pooled estimate were 45% and 48% respectively. As expected, studies using MPR to calculate adherence showed a higher estimate of the proportion of patients being adherent than studies using the more conservative PDC, 44% and 34%, respectively. Remarkably, one community study conducted in New Zealand and using dispensing records (sample size=732) showed a substantial higher adherence rate of 78% compared to other studies using prescription/claims data.[12] This study differed from the others as only patients with a minimum of 90 days of dispensed allopurinol and a minimal follow-up of six months were included. However, results did not change substantially after eliminating the restriction for inclusion on the minimum days of dispensed allopurinol. Whether this high rate is due to an actual better patient adherence, different study population or methodology, remains unclear.

Adherence found in studies using MEMS/pill count (*n*=2) or patient-reported (*n*=6) was considerably better. It is known that patient-reported approaches highly overestimate adherence,[33] due to social desirability. To avoid this bias partially, the use of a validated questionnaire is recommended. Two out of six studies, both conducted in New Zealand by Dalbeth et al.[18,19] used the MARS questionnaire. Although not specifically validated in

gout, it was developed to assess adherence to long-term medication regimens. The use of MEMS has been considered to be an accurate method for medication adherence, as it observes 'real-time' behaviour.[34] The monitor itself might modify medication taking behaviour, but these effects are considered small.[35]

The difference in adherence between prescription/claims opposed to MEMS/pill count data or patient-reported data might not be solely explained by the difference in methods to assess medication adherence, but could also reflect a difference in study population. The majority of prescription/claims studies included gout patients with no ULT prescription 12 months prior the start of the study, whereas studies using patient-reported data often included patients with previous use of ULT. These patients might have longer disease duration and might have experienced the negative consequences of not being adherent and therefore greater motivation to take their medication as prescribed. This might also apply to the patients treated by a rheumatologist instead of a general practitioner. Two prescription/claims studies showed indeed that patients receiving ULT from a specialist instead of a generalist had better adherence.[27,28] Unfortunately in the present review, only a few studies were conducted among patients exclusively under care of a general practitioner[2,13] or rheumatologist[16,17,21], due to low number of studies and difference in method to assess adherence it was impossible to conclude whether medication adherence differs between patients seen in first and second line.

Non-persistence was assessed in five prescription/claims studies and rates ranged between 56% and 88%.[14,23,27,30,31] This broad range is likely to be explained by the differences in permissible gap length and observation time. A drawback of the studies is the absence of information on reasons to stop or interrupt medication, it could even be provider directed. Thus far, only two studies investigated whether patients return to therapy. Harrold et al. found that among the 70% of patients who had a gap in therapy of at least 60-days, about 50% returned to treatment within 8 months, and 75% within 4 years.[31] This was in line with the study from Sarawate et al., who showed that 72% returned to therapy after experiencing a gap.[30] None of the studies investigated adherence to medication after restarting therapy. This could be valuable, as it can be assumed that in this subgroup the indication to use ULT is likely more pertinent, and the group that was non-adherent due to severe side effects has likely been excluded.

Nine studies related medication adherence to clinical gout related outcomes.[13,15,18,19,21,24,28,29,32] For six out of seven studies poor medication adherence was associated with lower serum uric acid concentration[13,18,19,21,24,28,29] and in one study with fewer gout flares.[32] However, for all studies reverse causality cannot be excluded, i.e. the dispersion of monosodium urate crystals during the initiation phase of ULT increases the risk of having a gout flare,[36] which may lead to poor medication adherence. Compared to the review from De Vera et al, the current review reports for 12 additional studies the relationship between medication adherence and potential influencing factors.[12-14,16,17,19-21,25,28,31,32] Findings strengthened insight into the protective role of older age, presence of hypertension and/or diabetes, and higher number of

comorbidities on medication adherence. In addition, moderate evidence was found for a role of origin in medication adherence. Further confirmation and exploration of reasons for lower adherence among ethnic minorities is needed. Other *patient-* and *therapy-related* factors, such as knowledge and perceptions of the disease and medication, and *health care team* and *system-related* factors [36] have only been investigated in a few small studies.[16,17,19,23]

The current study should be interpreted within the context of its potential limitations. First, all studies assessed medication adherence by using an indirect method; none of them verified if the patients actually consumed their medication. Second, the thresholds to define 'good' and 'poor' adherence were all arbitrary. Most studies used a cut-off value of 80%, whether this is clinically relevant and required for therapeutic benefit is uncertain.[38] Third, the wide range of measures and definitions of adherence and non-persistence complicates the interpretation and summation of the results. In the meta-analysis we therefore only included studies who defined adherent as the proportion of patients who took at least 80% of their medication during observation. To restrict the influence of other factors we performed a stratified meta-analysis on country of investigation and measure of adherence. Notwithstanding, we cannot exclude that other clinical or methodology factors explain the high heterogeneity of the results. A meta-regression analysis would have been more accurate but was impossible, since information on potential variables was often lacking and the total number of studies was too low. Fourth, for two studies the identification of gout patients relied on prescription of ULT only. It is unknown whether the patients had gout or used the medication for other purposes, like tumour lysis syndrome.[12,14]

In conclusion, this study points to the need for more attention to medication adherence to ULT among gout patients. Awareness among clinicians is also important in the view of their role in improving medication adherence.[39] Combining quantitative and qualitative studies to assess medication adherence are needed not only to reveal the dynamics in medication use but also to understand the rationale for interrupting or stopping medication usage.

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APPENDIX 1. SEARCH STRATEGY

Pubmed

(("Gout"[Mesh] OR "Gout" OR "Hyperuricemia"[Mesh] OR "Hyperuricemia" OR "Uric acid"[Mesh] OR "Uric acid" OR "Allopurinol"[Mesh] OR "Allopurinol" OR "Probenecid"[Mesh] OR "Probenecid" OR "Benzbromarone"[Mesh] OR "Benzbromarone" OR "Pegloticase"[Supplementary Concept] OR "Pegloticase" OR "Rasburicase"[Supplementary Concept] OR "Rasburicase") AND ("Patient Compliance"[Mesh] OR "Medication Adherence"[Mesh] OR "Compliance" OR "Adherence" OR "Persistence"))

APPENDIX 2. SENSITIVITY ANALYSES

Table S1. Results meta-analysis, proportion of patients adherent

Study	Sample size	Proportion (%)	95% Confidence interval	Weight (%) Fixed	Random
McGowan, Ireland (2016)	15,908	35.3	34,6 to 36,1	12,60	7,89
Rashid (A), USA (2015)	10,991	42.3	41.4 to 43.3	8.70	7.88
Horshburg, New Zeeland (2014)	732	78.0	74.8 to 81.0	0.58	7.62
Kuo, UK (2014)	49,395	39.7	39.2 to 40.1	39.11	7.90
Zandman-Goddard, Israel (2013)	7,644	17.4	16.6 to 18.3	6.05	7.88
Solomon, USA (2008)	9,823	36.3	35.3 to 37.2	7.78	7.88
Halpern, USA (2009)	10,070	43.6	42.6 to 44.5	7.97	7.88
Harrold (A), USA (2009)	4,166	44.0	42.5 to 45.5	3.30	7.85
Briesacher, USA (2008)	9,175	36.8	35.8 to 37.8	7.27	7.88
Sarawate, USA (2006)	2,405	28.0	26.2 to 29.8	1.90	7.81
Riedel, USA (2004)	5,597	56.0	54.7 to 57.3	4.43	7.86
Lee, Korea (2016)	132	71.2	62.7 to 78.8	0.11	6.54
Singh, USA (2016)	251	78.5	72.9 to 83.4	0.20	7.12
Total (fixed effects)	126,289	38.6	38.4 to 38.9	100.0	100.0
Total (random effects)	126,289	46.6	40.8 to 51.2	100.0	100.0

Table S2. Subgroup meta-analysis, proportion of patients adherent to their uric acid lowering therapy

Study	*N*	Sample size	Range of proportions (%)	Proportion (%)	95% CI	I^2
Overall, random	13	126,289	17.4 to 78.5	46.0	40.8 to 51.2	99.7%
Conducted inside USA	8	52,478	28.0 to 78.5	45.2	39.8 to 50.6	99.3%
Conducted outside USA	5	73,881	17.4 to 78.0	47.5	36.5 to 58.6	99.8%
Outcome measure PDC	4	77,853	17.4 to 42.4	33.5	24.4 to 43.2	99.8%
Outcome measure MPR	6	42,456	28.0 to 78.0	44.1	37.6 to 50.7	99.4%

7

Knowledge, illness perceptions and stated clinical practice behaviour in management of gout: a mixed methods study in general practice

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ABSTRACT

Objective

The objective of the present study is to explore knowledge, illness perceptions and stated practice behaviour in relation to gout in primary care.

Methods

This is a mixed methods study among 32 general practitioners (GPs). The quantitative assessment included the Gout Knowledge Questionnaire (GKQ; range 0–10; better) and Brief Illness Perceptions Questionnaire (BIPQ; nine items, range 0-10; stronger). Structured individual interviews obtained further qualitative insight into knowledge and perceptions, in the context of daily practice.

Results

Among 32 GPs, 18 (56.3 %) were male, mean age 44.4 years (SD 9.6) and mean working experience 17.1 years (SD 9.7). Median score [interquartile ranges (IQR)] on the GKQ was 7.8 [6.7-8.9] and 9.0 [8.0-10.0], when presented as open or multiple-choice questions, respectively. The BIPQ (median; [IQR]) revealed that gout was seen as a chronic disease (8.0; [7.0-9.0]), affecting life and emotions moderately (6.5; [5.0-7.0]), having many severe symptoms (8.0; [7.0-9.0]) and in which treatment could be very helpful (8.0; [7.0-9.0]). Further interviews revealed large variation in specific aspects of knowledge and about gaps concerning indications for uric acid-lowering therapy (UALT), duration of UALT, target serum uric acid (sUA) level or duration of prophylactic treatment. Finally, patients' adherence was not checked systematically.

Conclusion

Specific knowledge gaps and discrepancies between perceptions and stated practice behaviour were identified, which might hamper effective management of this well treatable disease. Improving evidence on the rationale and effectiveness of treatment targets and adherence interventions, tailoring guidelines to general practice and intensification of implementation of guidelines in primary health care seem to be needed.

INTRODUCTION

Gout is a chronic rheumatic disease with a reported prevalence of 2.5% in the UK and 3.9% in the USA, making it the most common inflammatory joint disease.[1-3] Despite being a well-treatable disease, it is recognized that the management of gout is suboptimal in both primary[4] and secondary care.[5] In a primary care study among patients with gout, low levels of allopurinol prescribing (57%), serum uric acid (sUA) level testing (55%) and achievement of target sUA level (<0.36 mmol/L) (22.4%) during a 5-year study period were shown.[6] In secondary care, adherence to American College of Rheumatology (ACR) guideline recommendations by rheumatologists could be highly improved, shown by a mean adherence score of 5.8 out of 8 ACR guideline recommendations. Low adherence on first-line uric acid-lowering therapy (UALT) dosage, acute prophylaxis dosage and length of prophylaxis was shown.[5] Common barriers for effective management can be distinguished into patient and physician barriers. Important barriers among patients were not only misperception of the severity and chronicity of gout, but also inadequate patient education resources, resulting in poor adherence to treatment.[7–9] Among physicians, underestimation of long-term complications and insufficient knowledge about the indications for UALT and about adequate dosing of UALT have already been suggested.[10,11]

On the same line, some qualitative studies explored barriers to effective gout management among patients and physicians.[12–16] Despite general practitioners (GPs) being the most relevant (in most countries) health care professionals when it comes to diagnosing and treating the disease, the studies in current literature included only a low number of GPs. Therefore, broad insight into how gout is managed by GPs is missing. Notwithstanding, linking knowledge, perceptions and stated practice behaviour is essential when planning to improve gout management for patients with gout.

The current study uses a quantitatively and qualitatively approach to understand knowledge, illness perceptions and stated clinical behaviour of GPs when managing gout with specific attention to the role of UALT, sUA level and prophylactic treatment.

MATERIALS AND METHODS

Study design and data collection

A mixed methods approach was used to investigate and understand specific knowledge gaps in pathophysiology and management of gout, illness perceptions about the disease and clinical stated practice behaviours in management. The Good Reporting of A Mixed Methods Study (GRAMMS) guidelines as provided by the Enhancing the QUAlity and Transparency Of health Research (EQUATOR) network were followed.[17,18]

The study was conducted in the southern part of The Netherlands. During a period of 2 weeks, GPs were asked to participate in the study. After agreeing, questionnaires on

demographics, gout knowledge and gout perceptions were administered, followed by a structured interview to explore in depth understanding and to relate both issues to practice behaviour. The structured interviews were audiotaped and transcribed verbatim. The GPs consented to quote part of the interviews in anonymized form.

Demographics

General questions about age, sex, years working experience as GP, hours involved in patient care, estimated number of patients with gout per year, practice type, recent education on gout (within the last year, yes/no) and familiarity with gout (on a 0-10 scale, 0 being not familiar and 10 being extremely familiar) were recorded.

Gout knowledge questionnaire

The Gout Knowledge Questionnaire (GKQ) aims to assess knowledge of patients or physicians and addresses ten multiple-choice questions related to the pathogenesis, treatment of acute attacks and also management of chronic gout.[19,20] GPs were first asked to answer the questions while blind for the answer options. In case that they hesitated (or did not use one of the questionnaire answer options), the interviewer showed the original GKQ multiple-choice answers. The GKQ was previously translated into Dutch according to the International Society for Pharmacoeconomics and Outcome Research (ISPOR) principles of good practice,[21] which is consistent with the approach proposed as best practice in rheumatology by Beaton.[22] As each correct answer provides a score of one, the total score ranges from 0 to 10, with higher scores indicating better knowledge.

Brief illness perceptions questionnaire

When completing the validated Dutch version of the Brief Illness Perceptions Questionnaire (BIPQ),[23,24] the GPs were asked to rate their personal perceptions, while imagining that they would suffer from gout with an "average" disease course. The BIPQ is a nine-item questionnaire that assesses cognitive and emotional perceptions of a disease, within nine domains (Q1: consequences, Q2: timeline, Q3: personal control, Q4: treatment control, Q5: identity, Q6: coherence, Q7: emotional representation, Q8: concern, Q9: cause). Q1 to Q8 are scored on an 11-point numeric rating scale (0 to 10), with higher scores representing a more threatening view per domain. Q9 additionally permits to list up to three items that play a causative role in the disease.

Structured interviews

After completing the questionnaires, the GPs were interviewed to gain more in-depth insight into knowledge and perceptions and link these to clinical practice behaviour,

specifically with regard to the role of sUA in diagnosis and follow-up, appropriate usage of UALT and the role of adherence in relation to management of the disease.

Data analysis

Descriptive statistics were used to present the demographics and results of questionnaires, and means with standard deviation (SD) or medians with interquartile ranges [IQR] were used depending on skewness of data.
For qualitative analysis, the verbatim transcripts were read repeatedly and independently by two readers. Using the grounded theory approach, a coding system with categories that were identified in the previous step was developed as well as a taxonomy of the data.[25] The two readers met regularly to discuss coding and interpretation of data. Wherever necessary, consensus was reached after discussing specific passages or a third reader acted as referee. Representative quotes were collected during data analysis and reported based on the frequency of the particular (and similar) quotes.

RESULTS

Demographic characteristics

Thirty-two GPs were interviewed. Eighteen (56.3%) were male; the mean age was 44.4 years (S.D. 9.6 years); the mean number of years of working experience as GP is 17.1 years (S.D. 9.7 years). The GPs were 34.1 h (S.D. 11.0) per week involved in patient care, and only four (12.9%) had followed an educational event on gout in the past year. The estimated number (mean) of new patients with gout in their practice was 8.9 (S.D. 7.0) per year, and familiarity with the disease was scored as 7.0 out of 10.0 (S.D. 1.1) (Table 1).

Table 1. Baseline characteristics for general practitioners (GP) (*n*=32)

Age (years), mean ± S.D.	44.4 ± 9.6
Male sex; *n* (%)	18 (56.3)
Practice type, *n* (%)	
Group practice	15 (46.9)
Private practice	4 (12.5)
Self-employed substitute	10 (31.2)
Other	3 (9.4)
Years' experience as GP, mean ± S.D.	17.1 ± 9.7
Hours involved in patient care, mean ± S.D. [range]	34.1 ± 10.9 [8-55]
Estimated new patients with gout per year, mean, (median), [IQR]	8.9 (7.0) [4.3-11.5]
Recent (<1 year) education in gout, n (%)	5 (15.6)
Self-reported Gout familiarity (score 0-10), mean ± S.D. [range]	7.0 ± 1.1 [5-9]
Gout Knowledge Questionnaire (score 0-10), mean, (median), [IQR]	
Open answers	7.4 (7.8) [6.7-8.9]
Multiple-choice answers	9.1 (9.0) [8.0-10.0]

Gout knowledge

The mean scores (number of correct answers) for the GKQ were 7.4 (median 7.8) [IQR 6.7-8.9] and 9.1 (9.0) [8.0-10.0] when answering an open or multiple-choice question with the original answer options, respectively. The numbers (%) of GPs with correct answers for each item are summarized in Table 2. Questions on the cause of gout (Q1, Q3), signs indicating an acute attack (Q2), treatment of an acute attack (Q4) and recognition of allopurinol being UALT (Q5) were correctly answered by 88 to 100% of the GPs in the open questioning part, respectively. Questions on flare prevention (Q9) and comorbidity (Q10) were answered correctly by 72 and 50%, respectively, but improved to 97 and 100% when presenting the original answer options. On the other hand, the question on the target value ("ideal value") (Q6) was answered correctly by 12%, in the open question, but it increased to 84 % when presenting the original answer options. Finally, the questions about non-pharmacological interventions (Q7) and duration of UALT (Q8) improved to only 75 and 69 % correct answers after seeing the answer options.

Illness perceptions about gout

The results of the perceptions of GPs about gout are summarized in Table 3. GPs considered gout to be a chronic disease (Q2: median 8.0), with (a considerable number of) severe symptoms (Q5: median 8.0), but with moderate impact on life and emotions (Q1 and Q8: median 6.5), and for which treatment is very helpful (Q4: median 8.0). They believed that gout is not strongly influenced by personal actions (Q3: median 4.0). A large variation was observed in perceptions of the amount of concerns gout can raise (Q6: median 5.0, IQR 3.3-6.8) and the level of understanding of the disease (median 6.0, IQR 3.3-7.0). Finally, 16 of 32 (50%) reported diet (alcohol, obesity) to be a major contributing cause of gout (Q9).

Qualitative analysis on knowledge, beliefs and practice behaviour

Table 4 shows the most frequent quotes per topic that were collected during the data analysis.

Assessment of serum uric acid and (use) of uric acid lowering therapy

First, divergent opinions about the usefulness of sUA to diagnose gout were observed. Ten GPs believe that sUA is necessary to diagnose gout, as gout cannot be diagnosed in the absence of hyperuricaemia. Twelve GPs indicated that sUA levels are required in some specific situations, namely the following: (1) "to differentiate gout from other diagnoses in atypical cases" (six GPs) and (2) "to strengthen the diagnosis of gout, which will lead to better treatment" (six GPs). The remaining ten GPs felt that sUA is not necessary and not even useful to diagnose gout. Reasons were (1) "gout is a clinical

diagnosis, preferably confirmed by joint aspiration" (four GPs) and 2) "a low sUA level does not exclude gout" (three GPs) and "sUA may be low, in particular when patients have an acute gouty arthritis" (three GPs).

Table 2. Gout knowledge level of general practitioners per question (*n*=32)

Question		Open question, correct answered, *n* (%)	Multiple-choice, correct answered, *n* (%)
1)	Q: What causes gout? Answer options: a) too little calcium, b) too much uric acid, c) an infection, d) diabetes	31 (96.9)	32 (100)
2)	Q: How do you know if you have an acute attack of gout? Answer options: a) you have a painful swollen joint, b) you have a change in blood tests, c) your skin gets red and itchy, d) you have a lump on your ear	30 (93.8)	32 (100)
3)	Q: What inside the joint causes attacks of gout? Answer options: a) bacteria, b) viruses, c) crystals, d) calcium	30 (93.8)	32 (100)
4)	Q: Which of these is a good treatment during a sudden painful attack of gout in someone with no other medical condition? Answer options: a) exercise, b) allopurinol, c) NSAIDs like ibuprofen, naproxen or indomethacin, d) benzbromarone	32 (100)	32 (100)
5)	Q: Lowering your blood uric acid can help prevent future attacks of gout. Which of these drugs can lower your blood uric acid? Answer options: a) allopurinol, b) prednisone, c) NSAIDs like ibuprofen, naproxen or indomethacin, d) colchicine	28 (87.5)	32 (100)
6)	Q: What is the ideal blood uric acid level to aim for after treatment of gout? Answer options: a) lower than 0.59 mmol/L, b) lower than 0.48 mmol/L, c) lower than 0.36 mmol/L, d) lower than 0.12 mmol/L	4 (12.5)	27 (84.4)
7)	Q: In order to reduce the serum uric acid, what can you do in addition to medications? Answer options: a) drink more beer, b) eat more seafood, c) eat more red meat, d) lose weight if you are overweight	Not applicable	24 (75.0)
8)	Q: If you are taking a drug to lower your blood uric acid levels, how long do you need to take this drug? Answer options: a) one month, b) one year, c) two years, d) forever	20 (62.5)	22 (68.8)
9)	Q: When taking a drug to lower your blood uric acid levels, there can be a temporary increase in gouty attacks. How can you prevent such attacks? Answer options: a) skip doses of the drug and restart, b) drink less water, c) drink alcohol every day, d) take daily colchicine	23 (71.9)	31 (96.9)
10)	Q: Which is a medical condition that is common in patients with gout? Answer options: a) high blood pressure, b) cancer, c) AIDS, d) asthma	16 (50.0)	30 (93.8)
Total correct score mean (median) [IQR]		7.4 (7.8) [6.7-8.9]	9.1 (9) [8-10]

Table 3. Results of the BIPQ in general practitioners

	Mean, (median) [IQR]	*n* (%)
Q1 Consequences (10 = severely affects life)	6.2 (6.5) [5.0-7.0]	
Q2 Timeline (10 = continues forever)	7.5 (8.0) [7.0-9.0]	
Q3 Personal control (10 = extreme amount)	4.3 (4.0) [3.0-5.0]	
Q4 Treatment control (10 = extremely helpful)	7.8 (8.0) [7.0-9.0]	
Q5 Identity score (10 = many severe symptoms)	7.7 (8.0) [7.0-9.0]	
Q6 Illness concern (10 = extremely concerned)	5.0 (5.0) [3.3-6.8]	
Q7 Coherence (10 = understands very clearly)	5.7 (6.0) [3.3-7.0]	
Q8 Emotional representation (10 = extremely affected emotionally)	6.2 (6.5) [5.0-8.0]	
Q9: Top listed causes:		
1. Diet (alcohol, obesity)		16(50.0)
2. Hereditary		13 (40.6)
3. Medication (i.e. diuretics)		12 (37.5)

Second, reasons to start treatment with UALT were very diverse. The most important reasons were the number of gout attacks per year: "The main reason to start with UALT is when patients have more than 3 gout attacks per year", severity of symptoms: "If patients have fewer attacks (e.g. <3), but the complaints during the attack are severe, then this is a reason to start UALT" and hyperuricaemia in case of a gout attack. Only six GPs mentioned tophi as reason to start with UALT, and three of these GPs determined the effectiveness of UALT, based on the resolution of (if present) tophi.

Third, with regard to duration of UALT, 12 GPs did not prescribe lifelong UALT, for one or more different reasons. Seven of these GPs tried to stop the UALT after 1 year: "If patients have no gout attacks for a longer period of time (e.g. 1 year), I try to reduce and eventually stop UALT"; five GPs suggested that UALT could be stopped after adjustment of lifestyle: "Allopurinol is prescribed lifelong, unless patients change their lives in such a way, you do not expect them to get gout attacks anymore (after weight reduction or stopping diuretics)"; six GPs terminated UALT in the occurrence of renal impairment. One GP thought that allopurinol could be used to treat an acute gouty arthritis.

Fourth, when initiating UALT, nine did not add prophylactic treatment to prevent flares. These GPs advised changing medication/lifestyle (three GPs), prescribed higher doses (or a combination) of UALT in case of flares during the drug start-up phase (three GPs) or waited until the patient was attack-free for a longer period before starting UALT (three GPs). Of the 23 GPs starting colchicine or a non-steroidal anti-inflammatory drug (NSAID) during UALT start-up, none prescribed prophylactic treatment for longer than 2 months: "I combine allopurinol and colchicine to prevent acute gout flares, for a period of 2-4 weeks". (14 GPs).

Finally, to determine effectiveness of UALT, 26 GPs determined sUA, of which six only in case patients continue to have attacks. Seventeen GPs explicitly stated that they did not strive for the target level of 0.36 mmol/L but based effectiveness of UALT on the absence of new gout attacks and stated that higher sUA levels were acceptable: "The target level of 0.36 mmol/L is not a strict treatment goal. I accept higher serum uric acid

levels if the number of acute attacks is decreased". Six GPs never determine sUA to monitor treatment.

Table 4. Themes from qualitative analysis with representative quotes

Number / Themes	Quotes
1 Knowledge	"I don't know the target level of serum uric acid; I always look in the lab form for the reference values." *(which are 0.20-0.42 mmol/L)*
2 Illness perceptions	"Gout is a chronic devastating systemic disease, leading to functional disability." "The associated kidney disease or heart failure are very serious conditions, but the acute attacks are the worst for patients."
3 Necessity of uric acid	"Gout cannot be diagnosed without the presence of hyperuricemia." "Serum uric acid is not useful, because it will be low in patients with an acute attack"
4 Treatment with UALT	"The main reason to start with UALT is when patients have more than 3 gout attacks per year." "If patients have fewer attacks (e.g. <3), but the complaints are very severe, then this is a reason to start UALT."
5 Duration of treatment with UALT	"Allopurinol is prescribed lifelong, unless patients change their lives in such a way, you do not expect them to get gout attacks anymore (after weight reduction or stopping diuretics)." "If patients have no gout attacks for a longer period of time (e.g. 1 year), I try to reduce and thereafter stop the UALT."
6 Flare prophylaxis	"I combine allopurinol and colchicine to prevent acute gout flares, for a period of 2-4 weeks." "I never prescribe allopurinol after an acute flare, first I prescribe colchicine (or a NSAID) and after 4 weeks I stop it and start allopurinol." "I do not prescribe prophylactic treatment, I advise patients to drink more and sometimes stop diuretics."
7 Target level serum uric acid	"The target level of 0.36 mmol/L is not a strict treatment goal. I accept higher serum uric acid levels if the number of acute attacks is decreased." "If patients have gout, I try to reduce the serum uric acid level below 0.36 mmol/L in order to reduce the hyperuricemia-associated risk of cardiovascular events. Furthermore, I will check and if necessary adjust cholesterol, blood pressure and glucose."
8 Adherence	"Adherence to UALT is not a problem in patients with gout, since they are well aware of the fact they will get new gout attacks if they do not take their medication." "I think patients with gout take their medication (UALT) very well in the beginning, but in the course of time become less adherent. Then these patients will return with a gout flare." "I have too little time to check whether patients with gout are adherent."
9 Lifestyle advices	"I refer my patients to a website (www.thuisarts.nl)* where al truths and untruths about gout are presented. If I am correct, there is no evidence for all these dietary advices" "I give the same lifestyle advices as I give patients in cardiovascular risk management" "I warn patients for the possible danger of alcohol and organ meats. Also, I try to motivate them to lose some weight"

* a Dutch website with the most essential information in plain language, understandable by patients, about diseases treated by GPs, an initiative from the Dutch College of GPs.

Adherence to drug therapy

Nineteen GPs believed that patients with gout are adherent to their drug treatment. "Patients are well aware of the fact new gout attacks will occur if they don't take their medication". Of the 13 GPs that assumed that patients were not adherent to therapy, nine GPs believed that they were adherent in the beginning but stop UALT over time: "I think patients with gout take their medication (UALT) very well in the beginning, but in over the course of time become less adherent". All GPs assumed that these patients would restart therapy themselves in case of a new attack Only eight GPs actively monitored patient adherence by planning appointments at a regular interval (varying from 1 month in the start-up phase to once a year), during which two determined sUA to assess adherence. Seven GPs check adherence when patients had an appointment for any reason. Furthermore, when specifically inquired, 12 confirmed that they checked regularly whether patients pickup their repeat prescriptions, but only electronically and no contact with the nonadherent patients would follow. If non-adherence was recognized (in any way), only 12 GPs would discuss the effects and complications of being non-adherent. Ten GPs admitted to spend insufficient effort in the follow-up of adherence. Main reasons are lack of time or beliefs that patients are adherent anyhow.

Lifestyle advice in patients with gout

Sixteen GPs believed that diet and drinking habits were main contributing causes of gout (BIPQ (Q9)), and all of these mentioned that adjustment of these factors (weight loss, less alcohol, no organ meats, drink more water) would lower sUA in addition to medication. It was therefore surprising to see that only four GPs gave any lifestyle advice(s) to patients with gout. Seven GPs explicitly mentioned that adjustment of diet was outdated.

DISCUSSION

Our study adds fuel to the ongoing debate about why gout, a treatable disease, is often insufficiently controlled.[8,26] The strength of this study is that it is the first to address, at the same time, knowledge, illness perceptions and stated clinical practice behaviour in GPs, the medical professionals that commonly diagnose and treat gout. Moreover, the use of a mixed quantitative and qualitative approach allowed to gain in-depth insight into the consequences of gout knowledge and (inadequate) perceptions on gout in general practice, while, at the same time, providing an overall quantification. In Figure 1, we summarized several potential barriers identified in our study and illustrated graphically how these barriers might eventually effect quality of care in patients with gout.

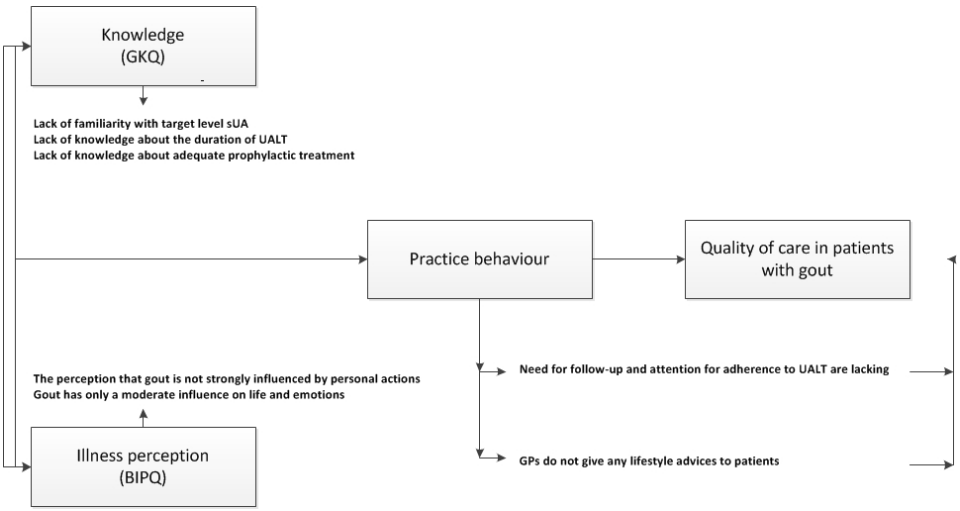

Figure 1. Identified barriers to optimal management in patients with gout treated by general practitioners

The GPs' knowledge, as measured with the GKQ, on pathophysiology, signs and symptoms and treatment of an acute gout attack was mostly excellent, although only half of them indicated that dietary factors play a causative role in gout. However, GKQ-data combined with interviews on knowledge and practice behaviour learned that there is a large variation in the long-term management of gout, specifically in reasons to start UALT, the duration of UALT prescribing and prophylactic treatment at initiation of UALT. The latter finding is in line with one other study already showing inappropriate use of prophylactic colchicine among 74% of the patients under care of a primary care physician.[27] Furthermore, although GPs have a pragmatic and realistic view on the evaluation of effectiveness of UALT, it was interesting that most GPs were not aware of the sUA target level of 0.36 mmol/L as recommended by guidelines and stated to use (if any) the upper limit of laboratory normal ranges (0.42 mmol/L). Finally, half of the GPs indicated that dietary factors play a causative role in gout, but only few would give lifestyle advices to improve and eliminate causative factors, although it might be attributed partly due to lack of high-quality evidence for specific dietary interventions (avoidance of alcohol, weight loss).[28] Moreover, lifestyle interventions could have a role in management of gout-associated comorbidities (e.g. cardiovascular diseases, renal disease).

It is well known that also perception of the burden of disease influences the dedication of professionals to a disease and its management. It was therefore reassuring that the GPs perceived gout as a chronic disease with severe symptoms and important impact, in which treatment is very helpful. The GPs' illness perceptions are in accordance with those of 142 patients with gout, which showed that patients also viewed gout as a

chronic condition responsive to therapy, but not influenced by personal actions.[29] Nevertheless, there was a striking unawareness among GPs with respect to need for follow-up and/or attention for adherence, as most GPs sincerely believed that patients were adherent to treatment. Other reasons to not make follow-up appointments or check adherence regularly were lack of time or believe that patients who were non-adherent would present themselves automatically when having a new gout flare, actually referring to the patients' personal responsibility. So, even when adherence was checked by the GP, actions to improve inadequate adherence were rare.

We realize that the interpretation of the results of our study might be difficult, since we did not actually evaluated quality of care by auditing GPs' adherence to treatment guidelines or quality indicators (QI). Nevertheless, using the ACR and European League Against Rheumatism (EULAR) guidelines as external standard,[30-32] we implicitly took a large number of the formulated QI by Mikuls et al.[33] into account. A first example would be the QI about the role of follow-up of sUA level when prescribing UALT: "IF a gout patient is given a prescription for a xanthine oxidase inhibitor, THEN a serum urate level should be checked at least once during the first 6 months of continued use, BECAUSE periodic serum urate measurements are required for appropriate dose adjustments of xanthine oxidase inhibitors (escalations or reductions)". A second example would be the QI about behavioural modifications: "IF a patient is diagnosed with gout and has either (1) obesity (defined as a body mass index ≥28 kg/m^2) or (2) frequent alcohol use (≥1 alcoholic beverage per day), THEN as part of their overall therapy, patients should be advised on the importance of weight loss and/or decreased alcohol use, BECAUSE weight loss and reduction of alcohol intake may be beneficial components of gout therapy". On this line, it is important to realize that the guideline on "arthritis", including recommendations how to diagnose and manage gout of the Dutch College of General Practitioners (NHG), currently does not mention a specific sUA level as a treatment target, does not recommend prophylactic treatment when initiating UALT and does not provide specific advises on behavioural modifications, follow-up or monitoring of adherence for patients with gout.[34] On the other hand, while the GPs' standard mentioned the presence of tophi as indication to initiate UALT, 26 GPs do not mention tophi as a reason to start UALT. Although guidelines are a good starting point to improve quality of care, it is well known that recommendations do not guarantee ubiquitous agreement or compliance with them. Harrold et al. reported among a random sample of US PCPs (including 444 GPs) that only 9.6% of the GPs were aware of the guidelines and adhered to recommended treatment for acute, intercritical and tophaceous gout in only 47, 3.4 and 12.5% of the cases, respectively.[35] In addition to (non)awareness, physicians (including GPs) are experts with strong opinions whom might not always agree with recommendations in guidelines and might question the evidence.

Although already a large amount of evidence is available and summarized in the 2006 EULAR and 2012 ACR guidelines, it should be recognized that the strength of evidence for several recommendations, such as the role and value using sUA as a target for treatment, still needs improvement. As such, we believe that QI that are part of GP's

audit might be more effective. Nevertheless, these QI still require strong evidence and a costly organization for monitoring and auditing.

Last, the differences in views between GP and international guidelines might be explained by the heterogeneity of the disease itself and important differences in disease spectra between primary and secondary care will be present. Undoubtedly, GPs treat the milder cases. Therefore, one of the outstanding issues is to collect high-quality registry data in primary care and identify factors that might predict poor prognosis.

This study has other limitations that need to be addressed. First, GPs were recruited from one region in The Netherlands. This might limit the generalizability of these results to all GPs in (and outside) The Netherlands. Nevertheless, we included a broad spectrum of GP that was also representative for the Dutch situation, with regard to years of working experience, sex distribution and age, as this is necessary for qualitative studies (in The Netherlands, 56% of the GPs are male with a mean age of 48.8 years and of which 46% have a fulltime employment). As such, the current study represents to date the largest qualitative study in gout. The number of 32 GPs is acceptable from a quantitative view, as for the qualitative part of the study, the theoretical saturation points of information were reached. Second, the GKQ was developed as a multiple-choice questionnaire with some of the multiple-choice answers being too obvious in our opinion. Therefore, the questionnaire was presented first with open-ended items (i.e. hiding the answer options), thereby eliciting quotes and thus supporting the qualitative analyses. Finally, in our study, as in any study with qualitative analyses, it might be possible that the interviewer, the questionnaires (that were completed before the interview) or the semi-structured character of the interview itself unintentionally influenced the GPs' answers.

In conclusion, among a sizable proportion of GPs, we have identified specific knowledge gaps and discrepancies between illness perceptions and stated clinical practice behaviour of GPs that might imply risks for shortcoming patient management in primary health care. Improvement of knowledge of evidence-based treatment targets, implementing adherence interventions and tailoring up-to-date guidelines to general practice are needed to ultimately improve the care of all patients with gout.

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8

Summary and general discussion

SUMMARY AND GENERAL DISCUSSION

Gout and its main risk factor hyperuricemia have been associated with several comorbidities, including hypertension. However, the underlying mechanism remains uncertain. Considerable research efforts have been made to understand the role of uric acid as an independent risk factor for the development of hypertension. Thus far, studies focused mostly on the role of total serum uric acid and have ignored the distinction between uric acid *concentration* and its *production*. Since there biological evidence that the production of uric acid may contribute, independent of uric acid concentration, to the pathogenesis of hypertension, the *production* should be investigated as well.[1,2] In *Part I* of the thesis we therefore investigated the role of uric acid production in the association between uric acid and blood pressure.

Gout is a well-manageable disease. In patients with recurrent gout flares or tophi, it is recommended to start long-term uric acid lowering therapy to reduce the number of gout flares and resolve tophi.[3] Notwithstanding, a substantial proportion of patients with gout continue to experience multiple gout flares, signifying insufficient control of the disease.[4] To improve the management of gout, studies exploring the barriers for effective gout management among both patients and physicians are needed.[5] In *Part II* of this thesis we explored first medication adherence in patients with gout requiring uric acid lowering therapy and next the knowledge, illness perceptions and stated clinical practice behaviour of general practitioners when managing gout.

This chapter summarizes the main findings of the studies in this thesis and discusses the most important methodological considerations. Finally, the clinical implications are presented and some directions for future research are proposed.

MAIN FINDINGS

Part I Uric acid and blood pressure: The role of uric acid production

In each of the **Chapters 2-4**, we tested the hypothesis that the production of uric acid is associated with blood pressure and hypertension. Three different proxies for uric acid production were investigated.

In **Chapter 2,** genetic variation in xanthine oxidoreductase (*XOR*) as a proxy for uric acid production was used to investigate whether uric acid production was associated with secular trends in mean arterial pressure, pulse pressure and the development of hypertension. Longitudinal data (median follow-up of 8.8 years) from 2769 participants of the Flemish Study on Environment, Genes, and Health Outcomes (FLEMENGHO) and European Project on Genes in Hypertension (EPOGH) study was used. Three of 25 tagging *XOR* single nucleotide polymorphisms (SNPs) were associated with the increase in pulse pressure (rs11904439), or mean arterial pressure (rs2043013), or the risk of

hypertension (rs148756340 and rs11904439). In normotensive participants (*n*=2050) the risk of hypertension was 30-70% higher in minor allele carriers of rs148756340 and rs11904439, respectively. With a false discovery rate set at 0.25, the aforementioned associations retained significance. Overall, these findings suggest that variation in uric acid production might be associated with secular trends in the steady and pulsatile blood pressure components and the risk of hypertension. It is of note, serum levels of uric acid at baseline (*n*=1949) were not associated with variation in *XOR* (*P*-value ≥0.05). Overall, these findings suggest that variation in uric acid production might be associated with secular trends in the steady and pulsatile blood pressure components and the risk of hypertension.

In **Chapter 3**, ratios of the different purine metabolites (precursors of uric acid) as proxies for XOR activity were used to examine the association between uric acid production and blood pressure in school-age children. An additional aim was to extend existing evidence on the relation between plasma uric acid concentration and blood pressure to this young population. Cross-sectional data from 246 children from the KOALA Birth Cohort Study was used. Higher ratios of uric acid/xanthine and xanthine/hypoxanthine, indicating higher XOR activity, were associated with higher diastolic blood pressure. Furthermore, higher plasma uric acid concentration was associated with higher diastolic blood pressure. However, no association with systolic blood pressure was found. Overall, these findings suggest that uric acid production was associated with blood pressure. In addition, plasma uric acid concentration was significantly associated with blood pressure already in school-age children.

Finally, in **Chapter 4** 24-h uric acid excretion in urine as a proxy for uric acid production was used to investigate whether uric acid production was associated with the steady and pulsatile blood pressure components and the prevalence of hypertension. In addition, we investigated whether serum uric acid was associated with one of these outcomes. Cross-sectional data from 2555 participants of The Maastricht Study was used. After adjustment for traditional hypertension risk factors, serum uric acid and 24-h urinary uric acid excretion were both associated with mean arterial pressure and hypertension. The association of both serum and urinary uric acid with mean arterial pressure remained significant after further adjustment for urinary or serum uric acid, respectively. These findings suggest that both serum uric acid and its production were, independent of each other, associated with mean arterial pressure and hypertension.

Taken together, the findings from **Chapters 2-4** suggest that uric acid production might be associated with elevated blood pressure, in particular with the steady component of blood pressure and hypertension. Regardless of the difference in studied proxies and study populations, our results point towards the same direction.

Part II Gout management by the patient and general practitioner

In **Chapter 5**, medication adherence among gout patients was investigated. A retrospective cohort study was conducted using the UK Clinical Practice Research Datalink (CPRD). Among 48,280 newly diagnosed gout patients initiated allopurinol the vast majority had poor medication adherence. Only 38% of the patients were considered adherent, defined as the proportion of days covered (PDC) of at least 80% during the entire observation time (mean of 5.5 years). Non-persistence, defined as the occurrence of a first gap of at least 90 days, increased from 39% to 57% after 1 and 5 years of initiation, respectively. After the occurrence of a first medication gap, 43% restarted therapy within 1 year, yet 52% experienced again a 90-day gap in the first year after re-initiation. In particular females and current smokers were at increased risk for non-persistence and non-adherence, while those being overweight, taking antihypertensive medication, and suffering from comorbidities such as dementia, diabetes, dyslipidaemia, and depression were more likely to be persistent and adherent.

In **Chapter 6**, we conducted a systematic literature review on medication adherence among gout patients treated with uric acid lowering therapy. Of the 23 included studies, medication adherence was much higher when assessed by electronic monitoring device (*n*=1), pill count (*n*=1), or patient-reported (*n*=6) data than when assessed by prescription/claims data (*n*=15). Of the 23 studies considered, 13 met the criteria of defining adherence at a threshold of 80% and were included in the meta-analysis. The pooled proportion of adherent patients among those studies was 46%, with very high heterogeneity (I^2>99%). Subgroup analyses were performed to determine if this heterogeneity could be explained by the country of investigation or measure of adherence. The pooled proportion of patients adherent was similar across studies conducted in the USA (*n*=8; 45%) and other countries (*n*=5; 48%). The pooled proportion of patients adherent was higher in studies using medication possession ratio (MPR) (*n*=6; 44%) compared to studies using proportion days covered (PDC) (*n*=4; 34%) to measure adherence. Non-persistence, occurrence of an interruption in therapy, was reported in five studies, but the heterogeneity in defining non-persistence (30-, 60- or 90-day interruption) made comparisons difficult. When considering the five studies and all possible gap lengths, more than 50% of the patients became non-persistent in the first year after starting uric acid lowering therapy. Poor medication adherence was associated with elevated serum uric acid as well as frequent gout flares. Factors associated with better medication adherence were older age, higher number of comorbidities, and the presence of diabetes or hypertension.

In **Chapter 7**, we explored the knowledge, illness perception, and stated practice behaviour in relation to gout among 32 general practitioners. By using a mixed quantitative and qualitative approach, we showed that knowledge on pathophysiology, signs and symptoms, and treatment of an acute gout attack was mostly excellent among general practitioners. However, there was a large variation in the stated long-term

management of gout, specifically concerning lifestyle advice and the use of uric acid lowering therapy. General practitioners perceived gout as a chronic disease with severe symptoms and important impact, in which treatment is very helpful. Nonetheless, there was a striking unawareness with respect to need for follow-up visits and/or attention for medication adherence.

Overall, from **Chapters 5-7** we can conclude that gout management was far from optimal. We showed poor medication adherence, in particular among younger gout patients, those with less comorbidities and not using other medications on a regular basis. Moreover, among general practitioners knowledge and illness perceptions of gout and its acute treatment were adequate and mostly excellent, whereas long-term management, including accurate prescription of uric acid lowering therapy, lifestyle advice, and adherence support were insufficient.

URIC ACID PRODUCTION AND BLOOD PRESSURE

While the strengths and weaknesses of the individual studies were described in the discussions of the respective chapters, it is worthwhile to add general and overarching reflections on (i) the challenges to measure uric acid production, (ii) influence of uric acid production on the different blood pressure components and (iii) and directions for future research.

Challenges to measure uric acid production

In the absence of a feasible approach to directly measure the production of uric acid three different proxies were investigated: variation in the *XOR* gene, ratios of different purine metabolites (hypoxanthine, xanthine, and uric acid), and 24-h urinary uric acid excretion. For each proxy the advantages and disadvantages are summarized in Table 1.

Table 1. The pros and cons for measures explored as proxies for uric acid product in this thesis

Proxy	Identification of overproducers	Drawbacks	Reliable	Participants burden
SNPs in the *XOR* gene	Partly, only those with increased *XOR* activity	- Linkage disequilibrium - Pleiotropic effect	Yes: the genotype is constant during life	*Low*: one sample is sufficient, e.g. from a buccal swab
Ratios of purine metabolites	Partly, only those with increased *XOR* activity	Feedback mechanisms, e.g. increased xanthine excretion via the kidneys	Time dependent: might be influenced by diet, physical activity, and medication use	*Moderate*: collection of one or multiple blood samples
Uric acid excretion in 24-h urine	Yes, those with increased *XOR* activity AND increased purine supply	Feedback mechanisms, e.g. increased uric acid excretion via the intestine	Time dependent: might be influenced by diet, physical activity, and medication use	*Moderate - high*: collection of one or multiple 24-h urine collections

First, variation in the *XOR* gene was used as a proxy for the enzyme activity of XOR. A genetic variant can function as a proxy for the exposure with the advantage that it reduces the potential for confounding and eliminates reverse causation.[6,7] This approach, called Mendelian randomization, can be considered as a natural randomized controlled trial.[6,7] Such an approach is useful in order to test our hypothesis. Furthermore, it is relatively easy and inexpensive to obtain DNA and to genotype the individual. Via a single buccal swab DNA can be extracted, this is inexpensive, not invasive and can easily be obtained at home, with the advantage that this is not time-consuming and burdensome for the participant.[8,9]

Although it is well established that the *XOR* gene encodes the enzyme XOR, the functionality of the investigated SNPs is unknown at present. Therefore, we do not know if these SNPs alter enzyme activity and subsequently increase the production of uric acid. In addition, a potential limitation by using genetic variation as a proxy for the exposure is that there might be linkage disequilibrium between variants of one gene to polymorphisms in other genes. Confounding will result if both the variant of interest and that with which it is in linkage disequilibrium are both associated with elevated blood pressure.[7] Another potential limitation is the chance of a pleiotropic effect. Pleiotropy is the phenomenon in genetics whereby a variant in the gene influences multiple phenotypes.[10] Interpretation of associations between variants in *XOR* and blood pressure might not be straightforward if the variant has pleiotropic effects.[7] It is unlikely that pleiotropy plays a significant role in our study, since *XOR* mainly catalyses the oxidation of hypoxanthine and xanthine. Another disadvantage, patients with increased purine supply will not be identified, limiting the discovery of overproduction of uric acid to a subgroup of individuals with increased XOR activity.

Second, ratios of the purine metabolites hypoxanthine, xanthine and uric acid were used as a proxy for increased XOR activity. As described in the introduction section, in animal studies and studies examining patients with XOR deficiency a decreased concentration of upstream metabolites was found.[11-14] We assumed an opposite effect in case of increased XOR activity, so a relatively lower hypoxanthine compared to xanthine and lower xanthine compared to uric acid concentration, but whether this opposite effect actually occurs requires further investigation. It is important to take into account that the activity of XOR also relies on regulatory feedback systems as well as concentrations and activities of other enzymes involved in the pathway.[15] For example, an accumulation of the purine metabolites might be compensated by alterations in urinary excretion of xanthine or hypoxanthine,[16] or the degradation of hypoxanthine to inosine monophosphate (IMP) and xanthine to xanthosine monophosphate (XMP) by hypoxanthine-guanine phosphoribosyltransferase (HGPRT).[14] Another disadvantage is that only people with increased XOR activity will be identified.

The advantage of using plasma purine metabolites as proxy is that blood samples are easily obtained, the collection is slightly burdensome for the participants, and the metabolites can even be determined in stored samples. The collection of a blood sample is slightly burdensome for the participant.

Third, 24-h urinary uric acid excretion was used as a proxy for increased uric acid production. Previous studies showed that an increase in uric acid production, from either endogenous or exogenous sources, increases uric acid excretion.[17,18] Since uric acid is predominantly excreted by the kidneys, we assumed to find an increase urinary excretion. However, whether the relative contribution of renal and extra-renal excretion is comparable in case of increased uric acid production or disorders in one of the eliminations pathways needs further investigation.[19-22] An advantage of urinary uric acid excretion as a means to identify uric acid overproducers is that overproducers, either because of increased XOR activity or increased purine intake, can be identified. A disadvantage is the collection of 24-urine samples, which is time-consuming and burdensome for the patient. The collection itself might therefore influence daily life regarding diet and physical activity and influences uric acid production and its subsequent excretion.

With regard to all laboratory measurements of uric acid and purine metabolites, it is of note that they were determined in a single sample. For study purposes the variability in concentrations was minimized by measuring all individuals under the same condition; in the KOALA study children were asked not to eat or drink 1.5 hours prior to and during the home visit, resulting in a mean fast time of 2.2 hours (standard deviation of 1.2 hours). In The Maastricht Study, blood values from the participants were measured after an overnight fast and urinary uric acid was collected over a 24-h time period. Despite the standardized protocol, biological variability due to diet, physical activity, diurnal cycles and seasonal rhythms might have influenced the concentration and cannot be excluded.[23-27] While repeated measurements will improve reliability, they are time-consuming and more burdensome for the participant.

From none of the proxies studied we have information on the form of the enzyme responsible for the oxidation. The original form xanthine dehydrogenase (XDH), preferentially reduces nicotinamide adenine dinucleotide (NAD^+) to NADH, whereas xanthine oxidase (XO), particularly present in the circulation, reduces molecular oxygen. Only for the latter form of the enzyme applies that during oxidation the reactive oxygen species (ROS) superoxide and hydrogen peroxide are generated as by products.[28-34] Because of the presence of two form of the XOR enzyme, the clinical impact of increased uric acid production is likely to depend on this. Several factors can influence and induce the conversion of XDH to XO, for example hypoxia and ischaemia.[35] The presence of these conditions might indicate that the oxidation is more often carried out by XO then when there is absence of these conditions.

In conclusion, among the proxies investigated in this thesis 24-h urinary uric acid excretion would be the most feasible and comprehensive proxy to identify uric acid overproducers. In this case, those with increased XOR activity and those with increased uric acid production can be identified.

Blood pressure as outcome

A second aspect worthwhile discussing when studying the relation between uric acid and blood pressure is the measurement of blood pressure itself. Different approaches can be liable to different types of measurement errors. In addition, the association of uric acid production with the steady and pulsatile blood pressure component should be discussed, as these might reflect different pathophysiological states.

Measurement of blood pressure

In the FLEMENGHO and EPOGH study, the average of five conventional blood pressure measurements was used to report blood pressure of individuals. A limitation of conventional blood pressure measurement is that the presence of masked hypertension (normal office blood pressure but ambulatory or home blood pressure readings are in the hypertensive range) or white coat hypertension (elevated office blood pressure but ambulatory or home blood pressure readings are in the normal range) cannot be excluded. A previous study showed that among untreated persons, by using 24-h ambulatory blood pressure measurement as the gold standard, the prevalence was as high as 20% for masked and 10-15% for white-coat hypertension.[37,38] In The Maastricht Study we used 24-h ambulatory blood pressure measurement, in this case patients with masked hypertension will be identified and those patients with white-coat hypertension will not be identified as having hypertension. In the KOALA Birth Cohort Study, the child's blood pressure was measured by a trained nurse and by the parents. In the discussion section of Chapter 3 the possible advantages and possible measurement errors by using nurse and parent measured blood pressure have already been extensively discussed.

Blood pressure components

As indicated in the introduction, the association of uric acid production with the specific blood pressure components should be discussed as these might reflect different pathophysiological states. The three proxies for uric acid production were associated with the steady blood pressure component and in one study with the pulsatile component, represented by mean arterial pressure or diastolic blood pressure and represented by pulse pressure, respectively (Table 2).

Table 2. Summary of the results between the studied proxies and the different blood pressure components

	Age of the population	Steady BP	Pulsatile BP	Hypertension
Variants in the *XOR* gene	All ages	**MAP**	**PP**	**Yes**
Ratios of purine metabolites	School-age, 7 years	**DBP**	SBP (P >0.50)	NA
Uric acid excretion in 24-h urine	40 – 75 years	**MAP**	PP (P = 0.18)	**Yes**

BP is blood pressure; DBP: diastolic blood pressure; MAP: mean arterial pressure; NA: not applicable; PP: pulse pressure; SBP: systolic blood pressure. Blood pressure component in bold indicate a significant association with the investigated proxy.

In the KOALA Birth Cohort study among school-aged children we found an association between two ratios of the purine metabolites with diastolic but not with systolic blood pressure (all *P*-values >0.50). This is in line with the idea that an increase in uric acid production increases vascular resistance but would initially lead only to an elevation in the steady blood pressure component, known as first phase.[1,28,39-41] After prolonged exposure, this might lead to embedding of the vessel structure and eventually an increase in the pulsatile blood pressure component, considered as the irreversible second phase.[42,43] Because of the young study population it was not possible to investigate the effects of prolonged exposure. In the older study population of The Maastricht Study, which consisted of individuals between 40-75 years of age, increased urinary uric acid excretion was associated with mean arterial pressure. The association with pulse pressure (*P*=0.15) was closer to significance compared to the KOALA Birth Cohort Study, but still insignificant.

In the FLEMENGHO and EPOGH study, there was no restriction on age and the study population consisted of individuals from ten years of age and older. Three SNPs of the *XOR* gene were associated with mean arterial pressure, and we also found an association with pulse pressure, in contrast with the KOALA Birth Cohort Study and The Maastricht Study. Of interest would be to investigate whether elevated pulse pressure results from prolonged exposure to increased uric acid production. Stratification on the duration of increased uric acid production would be a good approach, since polymorphisms are age independent a stratification by age could be used. However, only 159 out of 2050 individuals were minor allele carriers for rs11904439, and stratifying the group according to age was not possible because of insufficient statistical power.

Implications for clinical practice and future directions

As yet, the findings from our studies on the role of uric acid production in blood-pressure and hypertension have no clinical implications. Several steps are still required before it is possible to identify overproducers of uric acid and to collect evidence that overproduction leads to hypertension and subsequent cardiovascular disease. First, the studied proxies, or other proxies reflecting increased uric acid production, need to be validated. Second, after validation the current findings need to be confirmed. Third, it needs to be established at which thresholds proxies confer a sufficient risk for high blood pressure or hypertension. Finally, evidence needs to be collected whether a reduction in uric acid production, for example by the administration of an XO inhibitor, reduces the risk of high blood pressure and hypertension.

From a biological point of view, the relation between uric acid overproduction and blood pressure is complex and in this thesis we have taken a simplified view on this. Uric acid production is a process involving various factors that regulate the flux through the purine pathway. It depends besides on the activity of XOR and purine supply on the concentration and activities of all enzymes involved in the pathway, and the activity of feedback and other regulatory control systems.[15] A system biology approach can be of

additional value to study the entire flux of uric acid production, and eventually its association with blood pressure.
If future studies are able to convincingly demonstrate a relation between uric acid production and hypertension, it might have potential clinical relevance. Even if the effect is small, given the burden of chronic hypertension and the fact that only 37% of the patients obtain a well-managed blood pressure, controlling uric acid production might have clinical benefit.[44] Moreover, identifying overproducers of uric acid may have implications beyond patients with hyperuricemia. Since overproduction can also be a problem in individuals with normal uric acid concentrations. If their excretion capacity is adequate, they will not develop hyperuricemia, but may still suffer from the adverse consequences of increased uric acid production.

GOUT MANAGEMENT

Chapters in *Part II* revealed large variation in medication adherence in gout patients and showed a paradox in general practitioner knowledge on the long-term management of gout and their stated clinical behaviour. In addition to considerations raised in the discussion of each chapter, we will further reflect on (i) the methodology used to assess medication adherence, (ii) the reason for poor medication adherence, (iii) the role of the guidelines for optimal gout management, and (iv) implications for future research and further directions.

Assessing medication adherence

Our analyses in CPRD combined with the systematic literature review revealed large differences in medication adherence in studies using prescription/claims data compared to those using Medication Event Monitoring System (MEMS), pill count, or using patient-reported data. Several factors might explain this discrepancy, among which the method of assessing adherence and selection of the study population seem the most important. The advantages and disadvantages of the different methods to assess medication adherence are summarized in Table 3.
Overall, studies based on prescription/claims data (*n*=15) showed poor adherence to medication, independent of country of investigation and measure used to assess adherence. The main advantage of using prescription/claims data is that large populations can be studied and that any distortion caused by patient recall or desire to give socially accepted answers is eliminated.[46] A disadvantage, which also applies for our study conducted in CPRD, is that over-the-counter medication use, prescriptions in secondary care, and prescriptions filled are often not available. Subsequently, the number of gout patients considered to have poor medication adherence may therefore be overestimated. To minimize this overestimation in defining non-persistence a gap length of at least 30-days is considered. In this case an interruption of a few days or

weeks due to hospitalization, emergency supply, or holiday will not automatically lead to misclassification.

Table 3. Methods for measuring medication adherence.[45]

Test	Advantages	Disadvantages
Direct methods		
Directly observed therapy	Most accurate	Patients can hide pills in the mouth and discard them; impractical for routine use
Measurement of the level of medicine or metabolite in blood	Objective	Variations in metabolisms and "white coat" adherence can give a false impression of adherence; expensive
Measurement of the biologic marker in blood	Objective in clinical trials, can also be used to measure placebo	Requires expensive quantitative essays and collection of bodily fluids
Indirect methods		
Patient questionnaires, patient self-reports	Simple; inexpensive; the most useful methods in the clinical setting	Susceptible to error with increases in time between visits; results are easily distorted by the patient
Pill counts	Objective; quantifiable; and easy to perform	Data easily altered by the patient (e.g. pill dumping)
Rates of prescription refills	Objective; easy to obtain data	A prescription refill is not equivalent to the ingestion of medications; requires a closed pharmacy system.
Assessment of the patient`s clinical response	Simple; generally easy to perform	Factors other than medication adherence can affect clinical response
Electronic medication monitors	Precise; results are easily quantified; tracks patterns of taking medication	Expensive; requires return visits and downloading data from medication vials
Measurement of physiologic markers (e.g. heart rate in patients taking beta-blockers)	Often easy to perform	Marker may be absent for other reasons (e.g. increased metabolism, poor absorption, lack of response)
Patient diaries	Help to correct for poor recall	Easily altered by the patient
When the patient is a child, questionnaire for caregiver of teacher	Simple; objective	Susceptible to distortion

Contrary to prescription/claims studies, studies using self-reported adherence (n=6) showed considerably better adherence rates. Whether this is attributable to actual better adherence, the use of self-reported data, or selection bias could not be disentangled. Although depending on the type of questionnaire and study population, overall self-reported approaches overestimate adherence.[47] The validity is limited by social desirability bias and by subjective interpretations of responses by the interviewer.[48] Since only two studies used a validated questionnaire,[49,50] it is likely that, at least for the other four studies, adherence is somewhat overestimated. In this respect, objective methods would be more valid. Thus far only two small studies used MEMS (n=17)[51] or pill count (n=132)[52] data and both showed a high level of medication adherence.

The discrepancy in medication adherence between population studies (claims/prescriptions) and studies among patients treated in secondary care raises the suggestion that patients under care of a general practitioner behave differently compared to those under care of a rheumatologist. The patients under care by a rheumatologist are likely to have more severe gout, more comorbidities, and might have experienced the negative consequences of not being adherent, all this might lead to better adherence compared to those patients seen by the general practitioner. This idea is supported by two studies who found that patients receiving uric acid lowering therapy from a rheumatologist instead of a non-rheumatologist had better adherence.[53,54] It should be noted, due to the small sample sizes and difference in method to assess adherence, we cannot formally conclude that patients in secondary care have better adherence.[51,52]

Measuring persistence or adherence

The overall concept "medication adherence" consists of multiple components, including *implementation*, *non-persistence* and overall *adherence*. In this thesis mainly patient adherence and non-persistence (**Chapter 5 and 6**) and the initiation of uric acid lowering therapy by the general practitioner has been examined (**Chapter 7**). Which concept of medication adherence is clinically most relevant may depend on the specific disease, the prescribed medication and the clinical outcome of interest. With regard to uric acid lowering therapy in patients with gout, the therapy is usually considered to be lifelong after initiation. When uric acid lowering therapy is initiated because of the frequency of flares, but with absence of subcutaneous or intra-articular tophi, *persistence* with uric acid lowering therapy might be of primary importance. In patients in which uric acid lowering therapy is started in the presence of tophi, the velocity of tophi reduction is linearly related to serum uric acid concentration.[55] Thereupon, *adherence* to uric acid lowering therapy might be equally relevant as *persistence,* so serum uric acid concentration lower than the saturation point, e.g. less than 4 (238 µmol/L) or 5 mg/dL (300 µmol/L) can be achieved.

Factors affecting medication adherence

Thus far, the results from previous studies and our CPRD study gave a good overview of demographic as well as clinical patient characteristics which were associated with poor medication adherence, although the exact rationale for interrupting or stopping medication usage was seldom studied. Medication adherence is a multidimensional phenomenon determined by the interplay of five sets of factors, all of these *"dimensions"* play a significant role in patient medication adherence (Figure 1).[56]

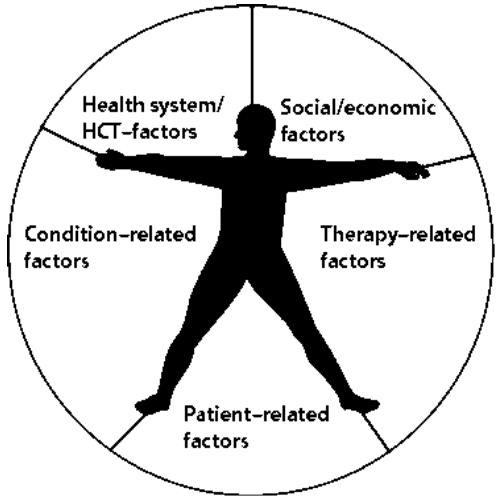

Figure 1. The five dimensions of medication adherence.[56]

One of these dimensions are *patient related factors* representing the resources, knowledge, attitudes, beliefs, perceptions and expectations of the gout patients and the use of medication.[56] For example, concerns about using medication might either be of practical nature, such as taking the medicine on a daily basis (forgetfulness), or perceptual nature, fears for actual short and long-term side effects or perceived efficacy of the medication, can influence patients' behaviour. These factors have not been frequently studied[50,52,57,58] and therefore no conclusion regarding their role in medication adherence can be made.

Other important aspects are related to *Health care team* and *system-related factors*. In The Netherlands, as in most other countries, the general practitioner is mainly responsible for the management of gout. The general practitioners in our study indicated that they had a lack of time and had a practical approach concerning adherence to medication and relied on the patient presenting themselves after experiencing a gout flare; the general practitioners themselves showed little concern for monitoring adherence directly. Our CPRD study confirmed that almost 50% of patients returned to therapy after an initial gap. However, assuming they all had a correct indication to start and to continue long-term uric acid lowering therapy, half of the patients did not return (Chapter 5). Even after restarting therapy, less than 50% were persistent in the following year. Restarting therapy thus seems not to guarantee better adherence to medication. Other reasons for the limited attention of the general practitioner to adherence could be an overestimation of their own communication skills or the patient's ability to understand the importance of appropriate medication use and lifestyle choices.[59] Finally, general practitioners may feel that their role in lifestyle and adherence support is of secondary

concern compared to their role in diagnosis and providing treatment. Thus, the common belief that solely the patient is responsible for taking the medication is misleading and obsolete.

Guidelines for the management of gout

Our study among general practitioners revealed a large variation in the long-term management of gout, mainly with regard to initiation and duration of uric acid lowering therapy, and initiation of prophylactic treatment. When trying to understand these differences, remarkable contradictions between the recommendations for the long-term management of gout developed by rheumatology societies or the American College of Physicians (ACP)[60] and Dutch general practitioner[61] societies were noticed (Table 4).

For example, treatment recommendations for gout developed by rheumatology societies promote a treat to target strategy, which is a serum uric acid concentration below the physiologic threshold of urate crystallization (<6.8 mg/dL) and below 5 mg/dL when tophi is present. Whereas, general practitioner and ACP societies promote a treat to symptom (gout flares) strategy. Another discrepancy is present for the initiation of uric acid lowering therapy; rheumatology societies promote the initiation of uric acid lowering therapy, while general practitioner and ACP are more reserved in the initiation of uric acid lowering therapy.

The discrepancy in recommendations has created much debate between both societies.[62-64] The developers of the ACP[60] guidelines argue their recommendations are based on verifiable systematic review of published evidence, whereas American Congress of Rheumatology (ACR)[65] and European League Against Rheumatism (EULAR)[66] rely, in case of absence of evidence, on expert panel opinions. This latter approach was supported by the argument that published literature may not be adequate in providing sufficient evidence for day-to-day clinical practice.

It is beyond the scope of this thesis to discuss details of the evidence and decision process that resulted in the different recommendations. However, it is of uttermost importance to draft a common agenda with a strategy how to solve the contradicting long-term management advises.

One unresolved issue, not specifically addressed in the current discussion on the different guidelines, is whether gout patients in first and secondary care differ in phenotype and therefore need different care and treatment. Possibly, experiences of general practitioners and other physicians truly differ compared to the experience of the rheumatologist. Moreover, most evidence on management of gout is based on studies conducted in the secondary or even tertiary care. Studies addressing the possible differences in patients that need to be referred to secondary care are needed to resolve this issue.

Table 4. Simplified overview of recommendations for the long-term treatment of gout, according to the different guidelines and rheumatology societies

Topics	Dutch College of General Practitioners (NHG) - 2009[61]	Dutch Society for Rheumatology (NVR) - 2013[67]	American College of Physician (ACP) - 2016[60]	European League Against Rheumatology (EULAR) - 2016[66]	American Congress of Rheumatology (ACR) - 2012[65,68]
Initiation uric acid lowering therapy	- >3 flares a year - Tophi	- According to number of flares a patient finds unacceptable (> 3 flares a year is considered as the standard). - Tophi - Urolithiasis	- Recommends against initiating in patients after a first gout attack or with infrequent attacks. - Patients with frequent gout attacks, shared decision while discussing the benefits, harms, costs, and individual preferences.	- Should be considered and discussed with every patient from the first presentation. - Patients with recurrent flares, tophi, urate arthropathy, or renal stones. - Recommended in patients aged <40 years, serum uric acid >8.0 mg/dL, or comorbidities.	- Frequent attacks (≥2 attacks per year) - Tophi - Chronic kidney disease stage 2 or worse. - Past urolithiasis.
Target	- Treat to avoid symptom: acceptable number of gout flares and removal of tophi. - Should be accompanied with lower uric acid concentration. - Preferably, lifelong treatment.	- Treat to target: serum uric acid <5.0 or 6.0 mg/dL, depending on the severity of the disease, so with or without tophi, respectively.	- Treat to avoid symptoms, based on what is acceptable for the patient.	- Treat to target: serum uric acid <5.0 or 6.0 mg/dL, depending on the severity of the disease, so with or without tophi, respectively. - Preferably, lifelong treatment.	- Treat to target: serum uric acid <5.0 or 6.0 mg/dL, depending on the severity of the disease, so with or without tophi, respectively.
Prophylaxis treatment when starting uric acid lowering therapy	- No recommendations	- Preferably six months of colchicine.	- In case of initiation of uric acid lowering therapy, the prophylactic treatment should be discussed as well.	- Explained and discussed with the patient, recommended during the first 6 months of uric acid lowering therapy.	- Yes, continued until there is no gout disease activity and/or the serum uric acid target has not been achieved.
Monitoring of serum uric acid	- Before initiation of uric acid lowering therapy, afterward every 4 weeks until target is achieved.	-	- The intensity of monitoring should be based on discussions with the patient, given the uncertainties of different treatment strategies	- Yes, so the target can be maintained. No information on the frequency.	- Regularly monitor serum uric acid (every 2 – 5 weeks) during titration of uric acid lowering therapy and every 6 months after achieving the target.

Implications for clinical practice and future directions

Even in the absence of agreement on when to start uric acid lowering therapy in patients with gout, it cannot be ignored that poor medication adherence is common and seems to have clinical relevance. Therefore, interventions to improve medication adherence would be welcomed. Recently, excellent long-term persistence and adherence on uric acid lowering therapy was achieved when gout patients received individualized education by the nurse.[69] In parallel to such an intervention, insight into the dynamics and reasons for poor medication adherence are needed to improve adherence and thus the care of gout. Recognizing that there is no consensus yet on the initiation of uric acid lowering therapy, a personalized approach and shared decision-making, might be a path to improve medication adherence.[70,71]

Next, action should be taken to solve the remarkable discrepancies between general practitioners and rheumatologists in the long-term management of gout. Next to tailored education for the provider, evidence based recommendation are required to resolve the ongoing debate on the best strategy in the long-term treatment of gout. The only way to overcome this would be a long-term trial which investigates the outcomes of the two different approaches: treat-to-target and treat-to-avoid symptoms. Such a study should be stratified according to setting, either general practitioner or rheumatologist, to ensure that the outcome is valid for the entire gout population, and not only those seen and treated in secondary care.

OVERALL CONCLUSION

Our findings suggest that overproduction of uric acid, reflected by studying three different proxies, might be involved in the pathogenesis of hypertension. Further research on the validation of the proxies and confirmation of the hypothesis is required before the clinical relevance can be considered. Furthermore, we showed suboptimal long-term management of gout in population studies. Poor medication adherence in gout patients as well as large variation in general practitioners' long-term gout management contribute to this problem. To ultimately improve the care of gout patients the implementation of adherence interventions and creating clear guidelines are necessary. Important to take into account, patients seen in first or second line might actually need different care; which might result into different recommendation for general practitioners and rheumatologists.

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ADDENDUM

Valorisation

Samenvatting

Acknowledgments

Curriculum vitae

List of publications

VALORISATION

An important aspect of research is to ensure its results will have, besides scientific merit, also social and economic value. This addendum describes how society may benefit from the work conducted in this thesis.

Gout is the most prevalent form of inflammatory rheumatic disease and affects 1 – 2% of the population worldwide.[1] The prevalence is particularly high among men, and approximately 7% of men above 65 are affected.[2] Hyperuricemia, the most important risk factor for gout, is with a prevalence of 20% far more prevalent than gout. Recent studies showed that both gout and asymptomatic hyperuricemia have been associated with an increased risk for cardiovascular disease, cardiovascular mortality and all-cause mortality.[3-5] Moreover, patients with gout or asymptomatic hyperuricemia have higher all-cause health care utilization and cost compared with those without a diagnosis of gout or without hyperuricemia.[6-8] For example, the estimated all-cause total direct costs was $16,925 per elderly gout patient and $10,590 for a non-gout patient.[9] Overall, gout and asymptomatic hyperuricemia impose a significant economic and social burden for society.[10]

While elevated serum uric acid is a clear risk marker for gout, the role of uric acid in the development of high blood pressure (hypertension) and cardiovascular disease remains uncertain. Since there is biological evidence that the production of uric acid might lead to vascular dysfunction and elevated blood pressure it is of interest to investigate this hypothesis.[11,12] In *Part I* of this thesis we therefore investigated the role of uric acid production, as an underlying mechanism, in the association between uric acid and blood pressure. Our findings suggest that overproduction of uric acid might be involved in the pathogenesis of hypertension. Although the study outcomes from *Part I* of this thesis may not directly lead to societal benefit at first, they can increase insight into the association of uric acid with hypertension and cardiovascular disease and may lead to further research initiatives.

Overall the studied proxies need to be validated in biological experiments to establish if they reflect increased uric acid production. Furthermore, the present findings need to be confirmed in other existing datasets. If confirmed, defining the threshold for uric acid overproducers at risk for developing hypertension is required. Finally, randomized controlled trials are needed to examine whether in uric acid overproducers inhibitors of uric acid production reduce (high) blood pressure or prevent the development of hypertension.

If our hypothesis will be confirmed, the identification of uric acid overproducers might be valuable to identify people who are at high risk for developing hypertension and cardiovascular disease. Today only 37% of the hypertensive patients obtain a well-

controlled blood pressure.[13] Even this estimate is conservative; the average expenditure for a hypertensive patient in the United States is about $1320 per year.[14] In other words, uncontrolled blood pressure is a high health burden for the patient and economic burden for society. Therefore, a better understanding of the underlying pathophysiology, the identification of persons at risk and novel pharmacological treatment is of main importance.

In the management of chronic gout, a substantial group of patients need long-term management with uric acid lowering therapy. From the findings of *Part II* of this thesis it appears that poor medication adherence among gout patients is common. We found that less than 40% of the patients in whom allopurinol treatment is started are adherent to their medication. Moreover, although patients who have stopped therapy are likely to return, the changes of a backlash are as likely.

Assuming that uric acid lowering therapy is initiated for the right reasons, non-adherence may not only lead to a higher health-burden for the patient, but may also lead to an economic burden due to additional health care expenditures in those with more advanced and possibly complicated gout. In order to improve medication adherence insights into factors associated with poor medication adherence should be known. The results of this thesis gave a good overview of demographic as well as clinical patient characteristics which were associated with poor medication adherence. Health care providers should provide extra support to those patients who are at high risk for poor medication adherence. Target groups are younger and 'healthier' patients, including patients with a normal weight, who are not on antihypertensive medication, and do not suffer from comorbidities like diabetes or hypertension. Although we identified patients at high risk, the exact rationale for interrupting or stopping medication usage was seldom studied. More insight into patients' understanding of the disease and medication and involved concerns is needed.

As gout is mainly treated in primary care and as poor medication adherence can be either a patient behaviour or provider directed, we explored the knowledge, illness perceptions and stated clinical practice behaviour of general practitioners when managing gout. The attitude about adherence was surprising since almost all general practitioners prefer to wait for recurrent attacks instead of actively screening for adherence. Moreover, if the problem of non-adherence is not addressed, they might draw the false conclusion that the medication is not effective and change to other, often more expensive, medications. Therefore, general practitioners should be more aware of the problem of non-adherence and take responsibility therein.

Changing clinical practice behaviour is a challenge, next to an educational program for general practitioners on the problem and importance of medication adherence, it is of utmost important to elucidate the general practitioners' opinion on the long-term

management of gout. Moreover, as discussed in the *Discussion* section of this thesis, further research should focus more on patients seen in primary care instead of those patients seen in secondary care. Eventually, this teaches us what the best strategy and target is for the long-term management of gout and would provide evidence-based recommendations for the treatment of gout in primary care.

Since non-adherence has consequences for the patient's health, health care provider, and society, medication adherence should be seen as a shared responsibility. It is a reasonable, although not yet proven assumption, that successful long-term management should improve individual's health and reduce the financial burden of gout. Thus far, little is known on the consequences of poor medication adherence, we therefore recommend further studies to investigate the impact of poor medication adherence on the patient's health, quality of life, health care utilization and related costs for society. These data can be the starting point for cost-effectiveness studies into adherence improvement programs.

In the end, the results of this thesis contribute to a better understanding of the role of uric acid in the development of hypertension. However, only after confirmation and when causality is established the societal utilization of the present findings may be feasible. Even if this only accounts for a small proportion of the total risk for hypertension and cardiovascular disease, addressing this may lead to a personalized approach and thereby a better public health. In addition, the results presented in this thesis emphasize the need for more awareness among health care providers for medication adherence. Whereas rheumatologists play an important role in treating the more severe cases of gout, general practitioners are the key player for most gout patients. This should be translated into intervention programs for improving medication adherence in a representative, thus mainly patients treated in primary care, gout population.

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SAMENVATTING

Jicht is de meest voorkomende reumatologische aandoening en ontstaat door het neerslaan van urinezuur kristallen in en rondom de gewrichten. Dit zorgt voor een pijnlijke ontstekingsreactie in het gewricht of de omringende weefsels. De belangrijkste risicofactor voor jicht is een verhoogde urinezuurconcentratie in het bloed (hyperurikemie), hetgeen wordt veroorzaakt door een verstoring in urinezuurproductie en/of –excretie. Daarnaast gaan jicht en asymptomatische hyperurikemie vaak gepaard met ongunstige cardio-metabole uitkomsten, zoals overgewicht en hoge bloeddruk (hypertensie). Jicht en de bijbehorende nevenaandoeningen vormen een grote last voor zowel de patiënt als voor de gezondheidszorg. Het is daarom van belang meer inzicht te krijgen in de samenhang van jicht en andere aandoeningen en de zorgverlening omtrent jicht.

Het doel van dit proefschrift was tweeledig:

I. Het onderzoeken van de rol van productie van urinezuur in de relatie tussen urinezuur en bloeddruk
II. Het in kaart brengen van therapietrouw aan urinezuur verlagende medicatie bij patiënten met jicht en in de zorg omtrent jicht door huisartsen.

Het onderzoek in dit proefschrift werd uitgevoerd in verschillende observationele studies. Voor het beantwoorden van doelstelling II werd daarnaast de literatuur omtrent medicatie inname systematisch bestudeerd en werden huisartsen ondervraagd.

DEEL I: URINEZUUR EN BLOEDDRUK: DE ROL VAN URINEZUURPRODUCTIE

De laatste decennia is er veel belangstelling voor de rol van urinezuur bij de ontwikkeling van hoge bloedruk. De oorzakelijkheid van het verband tussen urinezuur en bloeddruk is omstreden en de mogelijke onderliggende mechanismen zijn onduidelijk. Tot dusver is alleen het verband tussen urinezuurconcentraties en bloeddruk bestudeerd, en ontbrak onderzoek dat onderscheid maakte tussen de oorzaak van een verhoogd urinezuur, namelijk verhoogde productie of verminderde excretie. Er zijn aanwijzingen dat niet alleen het urinezuur zelf, maar ook de oxidatieve stress die ontstaat in de laatste fase van de productie van urinezuur verantwoordelijk is voor de nadelige invloed op bloeddruk. De oxidatieve stress die vrijkomt, kan mogelijk de perifere weerstand in de bloedvaten vergoten wat kan leiden tot een verhoogde bloeddruk.

De hypothese die getoetst wordt in deel I van dit proefschrift is dan ook dat een verhoogd productie van urinezuur, en niet alleen het urinezuur zelf kan leiden tot een hogere bloeddruk. Aangezien het vaststellen van urinezuurproductie niet uitvoerbaar is in grotere studies, hebben we drie verschillende maten als proxy voor urinezuur-productie gebruikt. Hieronder wordt een overzicht gegeven van de belangrijkste bevindingen en conclusies.

Hoofdstuk 2: variatie in het xanthine oxidoreductase gen

Binnen de Vlaamse FLEMENGHO en de Europese EPOGH studie onderzochten wij de associatie tussen varianten van het xanthine oxidoreductase (*XOR*) gen en bloeddruk. Het *XOR* gen codeert voor het enzym xanthine oxidoreductase (XOR) dat verantwoordelijk is voor het aanmaken van urinezuur. Daarbij keken wij of variaties van het *XOR* gen, als proxy voor 'urinezuur productie', geassocieerd waren met het risico op hypertensie en bloeddrukverandering over de tijd. De resultaten laten zien dat drie van de 25 onderzochte varianten in het gen samenhingen met een grotere toename in bloeddruk en een groter risico op hypertensie. Urinezuurconcentraties in het bloed hingen niet samen met variaties van het *XOR* gen.

Hoofdstuk 3: ratio's van verschillende purine metabolieten

Vervolgens onderzochten we de associatie tussen urinezuurconcentratie, urinezuurproductie en bloeddruk bij kinderen van de KOALA studie. In het bloed van deze 7-jarige kinderen hebben we de concentratie van urinezuur en de purines xanthine en hypoxanthine gemeten, en gekeken of deze samenhingen met een hogere systolische en diastolische bloeddruk. Ditmaal dienden ratio's van de verschillende metabolieten als proxy voor activiteit van het XOR enzym. Het hebben van een hogere urinezuur/xanthine en xanthine/hypoxanthine ratio, wijzend op een hogere XOR enzym activiteit, hing samen met een hogere diastolische bloeddruk. Daarnaast hing een hogere concentratie van urinezuur samen met een hogere diastolische bloeddruk. Dit suggereert dat al op jonge leeftijd zowel een verhoogde XOR activiteit *als* een hogere urinezuurconcentratie samenhangt met een hogere diastolische bloeddruk.

Hoofdstuk 4: urinezuur excretie in urine

Binnen De Maastricht Studie keken wij wederom naar de relatie tussen urinezuurproductie en bloeddruk. Ditmaal werd "overproductie" van urinezuur gedefinieerd op basis van de mate van urinezuurexcretie in urine. De aanwezigheid van een hogere concentratie urinezuur, bleek inderdaad samen te hangen met een hoger gemiddelde arteriële bloeddruk en de aanwezigheid van hypertensie.

Concluderend, de bevindingen van **hoofdstuk 2 – 4** suggereren dat een hoge urinezuurproductie samenhangt met een hogere bloeddruk. Het effect was vooral zichtbaar in de bloeddrukcomponenten die de perifere weerstand reflecteren, oftewel diastolische bloeddruk en de gemiddelde arteriële bloeddruk. Of een langdurig verhoogde urinezuurproductie uiteindelijk zal lijden tot vaatstijfheid dient verder te worden onderzocht. De resultaten van deze studies geven aanleiding tot vervolgonderzoek dat zich richt op de validatie van de bestudeerde proxies en het verder bevestigen van de hypothese, gevolgd door het bepalen van de klinische relevantie. Bijvoorbeeld welke afkapwaardes voor productie dienen te worden

gehanteerd om te bepalen of iemand een verhoogd risico heeft op een hypertensie en cardiovasculaire ziektes en of het zinvol is XOR remmers voor te schrijven om urinezuurproductie te verminderen bij patiënten die geen jicht hebben.

DEEL II: THERAPIETROUW BIJ PATIËNTEN MET JICHT EN JICHTMANAGEMENT DOOR DE HUISARTS

Jicht wordt beschouwd als een goed behandelbare ziekte. Een acute jichtaanval wordt behandeld met onstekingsremmers en in geval van chronische jicht is het verlagen van urinezuurconcentraties aanbevolen. Jicht wordt als chronisch beschouwd indien het recidiverend is of bij aanwezigheid van jichtknobbels, ook wel tofi genoemd. Ondanks de goede behandelmogelijkheden lijden veel patiënten aan recidiverende jicht, waaruit blijkt dat de behandeling niet optimaal is. Om de zorg omtrent jicht te verbeteren is het relevant de barrières van zowel de patiënt als behandelaar in kaart te brengen.
Bij veel chronische aandoeningen is het niet trouw gebruiken van medicatie een veelvoorkomend probleem. Therapie-ontrouw kan zorgen voor suboptimale controle van de ziekte, met alle nadelige gevolgen van dien. Eerder onderzoek wees uit dat therapie-ontrouw vaak voorkomt bij patiënten met jicht. Jongere patiënten en patiënten die niet ook bekend zijn met andere chronische aandoening zoals hypertensie of diabetes hebben een verhoogd risico om therapie-ontrouw te worden. Echter, het merendeel van deze studies is uitgevoerd in de Verenigende Staten en maak gebruik van verzekeringsdata. Of deze gegevens representatief zijn voor de jichtpatiënt in andere wereldregio's dient nader te worden onderzocht. Tevens ontbreekt er een overzicht van factoren die van invloed zijn op therapie-ontrouw.
Naast de patiënt speelt ook de huisarts een belangrijke rol bij de behandeling van jicht. Het handelen van de huisarts is afhankelijk van (i) de kennis over de pathofysiologie, oorzaken, en symptomen van jicht en (ii) de persoonlijk ideeën die de huisarts heeft over jicht (ziekteperceptie). De complexe interactie tussen kennis, ziekteperceptie en het daadwerkelijk handelen van de huisarts vraagt erom om gelijktijdig onderzocht te worden. Echter, dit ontbreekt nog.

Deel II van dit proefschrift richt zich daarom op het onderzoeken van therapietrouw bij jichtpatiënten die urinezuurverlagende medicatie voorgeschreven kregen. We keken naar de verschillende fases van therapietrouw: het starten, de uitvoering, en het stoppen of onderbreken van medicatie-inname. Daarnaast onderzochten we of de bevindingen van therapietrouw verschillend waren bij het gebruik van verschillende meetmethoden, bijvoorbeeld wanneer therapietrouw bepaald was aan de hand van het afhaalbewijs van de medicatie bij de apotheek, zelf-rapportage, of het elektronisch monitoren van het openen en sluiten van medicatiepotjes etc. Tot slot onderzochten we de kennis, ziekteperceptie en het daadwerkelijk handelen van de huisarts omtrent de zorg van jicht. Hieronder wordt een overzicht gegeven van de belangrijkste bevindingen en conclusies.

Hoofdstuk 5: therapietrouw bij jichtpatiënten

Binnen de huisartsendatabase 'Clinical Practice Research Datalink' (CPRD) uit het Verenigd Koninkrijk bestudeerden we therapie-ontrouw bij jichtpatiënten die allopurinol voorgeschreven kregen, de meest voorgeschreven urinezuurverlagende medicatie. Meer dan 50% van de patiënten was therapie-ontrouw. Slechts één-derde van de patiënten gebruikte meer dan 80% van de voorgeschreven medicatie gedurende de tijd dat patiëntgegevens aanwezig waren in het databestand (gemiddeld 5,5 jaar). Daarnaast onderbrak 40% van de patiënten hun medicatiegebruik met minimaal 90 dagen in het eerste jaar, oplopend tot bijna 60% in de eerste 5 jaar. Van deze patiënten herstartte bijna de helft, maar ondanks dit stoptte meer dan de helft wederom in het eerste jaar na herstart. Vrouwen en rokers hadden een verhoogd risico om therapie-ontrouw te worden, terwijl patiënten die antihypertensiva voorgeschreven kregen of leden aan overgewicht, dementie, diabetes, hoog cholesterol, en/of depressie vaker therapietrouw waren.

Hoofdstuk 6: systematische literatuur weergave van therapietrouw bij jichtpatiënten

In dit hoofdstuk wordt een systematisch literatuuronderzoek en meta-analyse naar therapietrouw bij jichtpatiënten gepresenteerd. In totaal werden 23 observationele studies geïdentificeerd. Verschillende meetmethodes van therapietrouw en kenmerken van studiepopulaties werden bestudeerd om te achterhalen of zij een deel van de therapietrouw konden verklaren. Wij zagen dat de mate van therapietrouw sterk samenhing met de gebruikte meetmethode. De waargenomen therapietrouw was velen malen hoger bij het gebruik van elektronisch monitoren van medicatiepotjes, het tellen van pillen door de behandelaar of zelf-rapportage, dan wanneer voorschrijf gegevens van de apotheek / huisarts werd gebruikt. Of dit enkel komt door de andere meetmethode of doordat andere meetmethodes ook vaak gepaard gingen met andere patiëntenpopulaties, zoals jichtpatiënten onder behandeling bij de reumatoloog in plaats van bij een andere behandelaar, kon niet worden afgeleid. De 13 studies die therapietrouw definieerden als "ten minste 80% van de observatietijd was gedekt met medicatie" werden samengenomen en lieten zien dat gemiddeld genomen nog geen 50% van de patiënten therapietrouw was. De mate van therapie-ontrouw in studies uitgevoerd in de Verenigde Staten waren vergelijkbaar met de resultaten van studies uit andere wereldregio's. Vijf studies onderzochten of patiënten hun medicatiegebruik onderbraken, hieruit bleek dat meer dan 50% van de patiënten hun therapie onderbraken met ten minste 30 dagen.
Therapietrouw hing samen met lagere urinezuurconcentraties. Daarnaast was er overtuigend bewijs dat patiënten die ouder zijn, lijden aan meerdere chronische aandoeningen, of lijden aan diabetes en/of hypertensie beter therapietrouw zijn. De reden voor therapie-ontrouw was slechts in enkele studies bestudeerd en hier konden geen conclusies aan worden verbonden.

Hoofdstuk 7: management van jicht door de huisarts

Tot slot werden zowel het kennisniveau als de percepties van huisartsen over de ziekte jicht bestudeerd. Hierbij maakten wij gebruik van bestaande vragenlijsten en aanvullende verdiepende vragen. De kennis over de pathofysiologie, symptomen en de behandeling van acute jicht was nagenoeg uitstekend onder huisartsen. Echter, wij vonden een grote variatie betreft de zorg van chronische jicht. Met name op het gebied van leefstijladvies, het voorschrijven van urinezuurverlagende medicatie en de bijkomende controle van therapietrouw en urinezuurbepaling in het bloed. Een gebrek aan tijd en het neerleggen van de verantwoordelijkheid bij de patiënt waren hiervoor voornaamste reden.

Concluderend, de resultaten van **hoofdstuk 5 – 7** laten zien dat management van jicht nog veel te wensen over laat. Een meerderheid van de patiënten was therapie-ontrouw in het nemen van urinezuurverlagende medicatie. Echter, we vonden grote verschillen in de mate van therapietrouw bij de verschillende meetmethodes. Of dit kwam door de gebruikte methode, studieduur of –populatie konden we niet achterhalen. Op basis van onze bevindingen adviseren wij dat toekomstige studies kwantitatief en kwalitatief onderzoek combineren. Zo kan meer inzicht in de dynamiek van medicatiegebruik worden verkregen en kan de reden achter stoppen en herstarten worden achterhaald.
Ten slotte zagen we dat ondanks de goede kennis van de huisarts over de pathofysiologie, oorzaken en symptomen van jicht de lange termijn zorg vaak onvoldoende. Duidelijke richtlijnen voor de lange-termijn zorg van jicht zijn hierbij belangrijk. Bij het bewerkstelligen van dit doel is het belangrijk te realiseren dat de gegeven zorg van de huisarts en reumatoloog van elkaar kunnen verschillen. De patiënt onder behandeling bij de huisarts heeft mogelijk een ander ziekteverloop en daarom andere zorg nodig dan de patiënt onder behandeling bij een reumatoloog. De richtlijnen voor de huisarts zouden dan ook gebaseerd moeten worden op wetenschappelijk onderzoek bij eerstelijns jichtpatiënten en niet, wat nu meestal het geval is, op patiënten uit de tweedelijns zorg.

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"Are you sisters?"
"Uuuh.... Nee"
"But you look like sisters"
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CURRICULUM VITAE

Lieke Scheepers was born on June 20th 1988 in Roosendaal, The Netherlands. In 2005, she graduated from secondary school at the Norbertuscollege in Roosendaal, which was followed by a one-year exchange program to Australia. Afterwards, she studied Human Kinetic Technology at The Hague University of Applied Sciences and obtained her Bachelor of Engineering degree in 2010. During her Bachelor Program, she performed an internship at the orthopedic workshop in Ghana (2008).

After travelling and cycling across Asia and Eastern-Europe for a year, Lieke started her Master in Public Health – Epidemiology at Maastricht University in 2011. During the Master Program, she did an internship at the department of Epidemiology to investigate the association between the intestinal microbiota composition and weight development in children under supervision of prof. Ilja Arts and dr. John Penders.

In January 2013, Lieke started her PhD project at the department of Rheumatology at Maastricht University Medical Center and at the Care and Public Health Research Institute (CAPHRI) at Maastricht University. The research described in this thesis was conducted under supervision of prof. Annelies Boonen, prof. Ilja Arts and prof. Coen Stehouwer. During her PhD project, she also worked at the research center of The Maastricht Study where she participated in data collection.

Currently, she is appointed as a postdoctoral research fellow at the department of Rheumatology at Gothenburg University in Sweden. Under supervision of prof. Lennart Jacobsson and dr. Mats Dehlin she will continue her research on gout.

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Not part of the thesis

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