Rapid diagnosis of bloodstream infections

Valorisation of the thesis

Background

Bloodstream infections are the most common form of infections. Mortality is high, between 15 and 35%[1-5]. As a result, bloodstream infections in the top 7 of most common causes of death, and account for approximately 157.000 deaths in Europe annually[6]. The incidence is rising, and as a result, the total number of deaths attributed to bloodstream infections has risen[7,8].

Treatment of bloodstream infections has two pillars: symptomatic treatment, by fluid resuscitation, and treatment of the cause of infection with antibiotics. Empirically one or more broad-spectrum antibiotics are administered, but since no 'one size fits all' antibiotic exists, therapy will likely be too narrow or too broad. In the past years antibiotic resistance has increased substantially in Gram-negative bacteria[9]. Thus, empirical therapy is more likely to be inadequate, which is associated with increased mortality. It is therefore increasingly important to know the antibiotic susceptibility pattern of the causative microorganism. Data on the antibiotic susceptibility of a microorganism currently are not available until at least 2 days after initiation of antibiotic therapy, since a positive blood culture requires more than one working day to grow and results of antibiotic susceptibility testing will not become available until another day later. More rapid identification and antibiotic susceptibility testing potentially enable earlier switching to adequate therapy and reduce the use of broad-spectrum therapy.

Conclusions of this thesis

In this thesis more rapid methods for ID and AST are presented, combining techniques and devices that are already available in the majority of microbiological labs (**Chapters 2**,**3** en **4**). Furthermore, it was shown that by using these methods, results of ID and AST are available significantly earlier than standard-of-care methods (**Chapter 5**). Also, significantly more patients received appropriate therapy on the same working day the blood culture had signalled positive, due to the rapid tests. However, due to lack of implementation of test results this did not result in a reduction of total time too-broad or inadequate therapy was used (**Chapter 5**). In addition it was shown that MALDI-TOF MS can be a useful tool in discerning blood culture contamination from true bloodstream infection, possibly leading to less use of antibiotic therapy (**Chapter 6**).

Target groups

The methods and results presented in this thesis are important for many groups of professionals involved in the healthcare system.

For clinical microbiologists and molecular medical microbiologists, the methods presented in this thesis offer new approaches to the diagnosis of bloodstream infections. It was shown that

more rapid bacterial ID and AST are possible, using techniques that they very likely already have available. The DNA-extraction techniques used in this thesis were very simple to perform, cheap and reliable, showing that PCR-assays on positive blood cultures can be performed without the need of expensive and extensive DNA-extraction kits. Additionally, it was shown that MALDI-TOF MS, which nowadays has become standard equipment in almost every lab, can help in determining whether a blood culture was contaminated or not. Such test results may help clinicians in deciding whether or not to prescribe antibiotic therapy.
For clinical microbiologists, as well as for infectious diseases specialists, the thesis shows how important implementation of test results is, and that focus on successful implementation strategies is essential in gaining full benefit from technical advances.

Ultimately, and most importantly, the results of the thesis are important for the patient. It shows that rapid ID and AST did not result in an overall time reduction of administering inadequate or too broad-spectrum antibiotic therapy. However, for a small group of patients it did result in significantly earlier appropriate antibiotic therapy. When implementation rates are higher, earlier overall appropriate therapy may be achieved with these new techniques. Earlier adequate antibiotic therapy and early recognition of a clinically relevant blood culture can improve the patients' prognosis[10,11]. Narrowing down antibiotic therapy earlier can also be beneficial for the patient, since using less antibiotics means a lower risk of side effects, and more narrow-spectrum antibiotics carry a lower risk of *Clostridium difficile* infections[12,13].
The benefits of narrowing down antibiotic therapy earlier will be more profound on the population level, since it reduces the total use of antibiotics. The more antibiotics are being used, the higher the levels of antibiotic resistance are[14]. In some countries, carbapenem resistant Enterobacteriaceae (CRE) and MRSA have already become endemic, making treatment of infections with these bacteria much more difficult, and sometimes even impossible[15,16]. In the Netherlands, this is not (yet) the case, infections with CRE and MRSA are still rare. However, infections with ESBL-producing Enterobacteriaceae seem to become more and more common[9], and outbreaks of infections caused by CRE have already been described[15]. These data indicate that in the future we may also face more serious resistance problems if no preventive measures are implemented. Reducing the use of antibiotics could result in lower rates of development and selection of resistant microorganisms, and more rapid ID and AST could aid in the reduction of antibiotic use, if properly implemented.

Financial implications

In 2014, expenses on antibiotics in the Netherlands were 35,6 million euros, which was an increase of 0.6 million euros when compared to 2011. The total number of Defined Daily Doses (DDD) of antibiotics had decreased with 6% between 2011 and 2014, and prices of most antibiotics have also decreased over the past years[17]. That the total expenses on antibiotics have risen nevertheless can be attributed to the increased use of more expensive broad-spectrum antibiotics, such as 3rd generation cefalosporins, linezolid, vancomycin and

meropenem[9,17]. Prices for these antibiotics range from 14.82 euros/dose (for vancomycin) to 60.84 euros per dose (linezolid), whereas the more frequently used amoxicillin, amoxicillin-clavulanate and cefuroxime are relatively inexpensive, ranging from 1.37 to 5.04 euros/dose[9,18].
Earlier results of ID and AST can lead to an earlier switch to a more narrow-spectrum antibiotic and thus a reduction of use of these expensive broad-spectrum antibiotics. However, as a result of the relatively low antibiotic resistance rates in the Netherlands, expensive broad-spectrum antibiotics are used relatively infrequent. The most widely used broad-spectrum antibiotics are 3rd generation cefalosporins, with 5 DDD per 100 patient-days, on a total amount of antibiotics used in hospitals of 74.7 DDD per 100 patient-days (6.7%)[9]. But the other expensive very broad-spectrum antibiotics combined represent only a very small fraction of the total of amount of antibiotics used in hospitals, with 3.3 DDD per 100 patient-days (4.4%)[9]. Although antibiotic costs are only a small fraction (<1%) of the total costs of pharmaceuticals in the Netherlands, which are >4 billion euros[17,19], earlier ID and AST can result in a reduction of direct costs of antibiotics.

But earlier adequate therapy results in a better prognosis for the patient[10,11], which results in a reduction in health care costs, for instance by shortening length of hospital stay. And since reducing the amount of antibiotics used could reduce antibiotic resistance development, it would also reduce the additional healthcare costs that are generated by infections with antibiotic resistant microorganisms, which are estimated to be 1,5 billion euros per year in Europe[20] and even 20 billion dollars per year in the USA[21].

Future references

The assays presented in this thesis can easily be adjusted, by adding or replacing microorganisms or antibiotics. Automation of the tests potentially allows for more rapid assays, reduced hands-on time, higher throughput and lower costs. And in the future, more rapid PCR techniques could be used to reduce PCR time.

The assays could be combined with other measures to reduce time-to-results, such as extended laboratory opening hours and improved hospital and laboratory logistics.
Implementation of test results can be improved by the formation of Antibiotic Stewardship Teams. The rapid assays could be of assistance in their goal of earlier adequate therapy and reduction of unnecessary broad-spectrum antibiotic therapy.

Possibly, currently available methods are not sufficient in reducing broad-spectrum antibiotic use and time to appropriate therapy. Development of more rapid techniques is required, especially since studies have shown that the rate of antibiotic switching is inversely related to the time to results[22,23]. Since the incubation of the blood culture bottle is the most time consuming step, this would be the most likely target for improvement. It is possible to reduce

incubation time and still obtain reliable ID results, as was shown by Loonen *et al.* in 2012[24]. Ideally, the blood culture step can be eliminated completely, by using PCR- and sequencing techniques that can be used directly on blood. However, the possibility that, in the near future, these techniques can replace culture based techniques is low, for reasons described in **Chapter 1**. At this moment, most effort is invested in new, more rapid assays and techniques that are performed on positive blood cultures or subcultures on agar.

Additionally, better techniques and tools to discern contaminated blood cultures from those representing true bloodstream infection are needed with the goal to reduce, or, ideally, eliminate blood culture contamination.

It is important to limit the costs of newly developed techniques, because although a more rapid diagnosis can save money and lives, healthcare costs continue to rise and more patients will require diagnosis and treatment of bloodstream infections, since their incidence is rising. Cost effectiveness studies should thus be part of the introduction of new tests.

References

1. Bos MM, Smeets LS, Dumay I, de Jonge E. Bloodstream infections in patients with or without cancer in a large community hospital. *Infection*. 2013;41(5):949-958.
2. Laupland KB, Svenson LW, Gregson DB, Church DL. Long-term mortality associated with community-onset bloodstream infection. *Infection*. 2011;39(5):405-410.
3. Hounsom L, Grayson K, Melzer M. Mortality and associated risk factors in consecutive patients admitted to a UK NHS trust with community acquired bacteraemia. *Postgrad Med J*. 2011;87(1033):757-762.
4. Lillie PJ, Allen J, Hall C, et al. Long-term mortality following bloodstream infection. *Clin Microbiol Infect*. 2013;19(10):955-960.
5. Melzer M, Welch C. Outcomes in UK patients with hospital-acquired bacteraemia and the risk of catheter-associated urinary tract infections. *Postgrad Med J*. 2013;89(1052):329-334.
6. Goto M, Al-Hasan MN. Overall burden of bloodstream infection and nosocomial bloodstream infection in north america and europe. *Clin Microbiol Infect*. 2013;19(6):501-509.
7. Martin GS, Mannino DM, Eaton S, Moss M. The epidemiology of sepsis in the united states from 1979 through 2000. *N Engl J Med*. 2003;348(16):1546-1554.
8. Sogaard M, Norgaard M, Dethlefsen C, Schonheyder HC. Temporal changes in the incidence and 30-day mortality associated with bacteremia in hospitalized patients from 1992 through 2006: A population-based cohort study. *Clin Infect Dis*. 2011;52(1):61-69.
9. The Dutch Foundation of the Working Party on Antibiotic Policy (SWAB), Centre for Infectious disease control (CIb) of the National Institute for Public Health and the Environment of the Netherlands (RIVM). NethMap 2015: Consumption of antimicrobial agents and antimicrobial resistance among medically important bacteria in the netherlands in 2014. 2015.
10. McGregor JC, Rich SE, Harris AD, et al. A systematic review of the methods used to assess the association between appropriate antibiotic therapy and mortality in bacteremic patients. *Clin Infect Dis*. 2007;45(3):329-337.
11. Paul M, Shani V, Muchtar E, Kariv G, Robenshtok E, Leibovici L. Systematic review and meta-analysis of the efficacy of appropriate empiric antibiotic therapy for sepsis. *Antimicrob Agents Chemother*. 2010;54(11):4851-4863.
12. Afshari A, Schrenzel J, Ieven M, Harbarth S. Bench-to-bedside review: Rapid molecular diagnostics for bloodstream infection--a new frontier? *Crit Care*. 2012;16(3):222.
13. Dark PM, Dean P, Warhurst G. Bench-to-bedside review: The promise of rapid infection diagnosis during sepsis using polymerase chain reaction-based pathogen detection. *Crit Care*. 2009;13(4):217.
14. ECDC (European Centre for Disease Prevention and Control), EFSA (European Food Safety Authority) and EMA (European Medicines Agency). ECDC/EFSA/EMA first joint report on the integrated analysis of the consumption of antimicrobial agents and occurrence of antimicrobial resistance in bacteria from humans and food- producing animals. *EFSA Journal*. 2015;13(1):4006-4114.
15. Glasner C, Albiger B, Buist G, et al. Carbapenemase-producing enterobacteriaceae in europe: A survey among national experts from 39 countries, february 2013. *Euro Surveill*. 2013;18(28):20525.
16. European Centre for Disease Prevention and Control. Http://Ecdc.europa.eu/en/healthtopics/antimicrobial_resistance/database/pages/map_reports.aspx. 2015.
17. Medisch Contact. Gebruik antibiotica gedaald. http://medischcontact.artsennet.nl/Actueel/Nieuws/Nieuwsbericht/151670/Gebruik-antibiotica-gedaald.htm. Updated 5 November 2015.
18. Zorginstituut Nederland. http://www.medicijnkosten.nl.
19. Stichting Farmaceutische Kengetallen. *Data en feiten 2015; het jaar 2014 in cijfers.* Den Haag: Stichting Farmaceutische Kengetallen; 2015.

20. ECDC (European Centre for Disease Prevention and Control) and EMA (European Medicines Agency). The bacterial challenge:Time to react; A call to narrow the gap between multidrug-resistant bacteria in the EU and the development of new antibacterial agents. 2009.
21. Centers for Disease Control and Prevention (CDC). Antibiotic resistance threats in the united states, 2013. . 2013.
22. Munson EL, Diekema DJ, Beekmann SE, Chapin KC, Doern GV. Detection and treatment of bloodstream infection: Laboratory reporting and antimicrobial management. *J Clin Microbiol*. 2003;41(1):495-497.
23. Kerremans JJ, Verbrugh HA, Vos MC. Frequency of microbiologically correct antibiotic therapy increased by infectious disease consultations and microbiological results. *J Clin Microbiol*. 2012;50(6):2066-2068.
24. Loonen AJ, Jansz AR, Stalpers J, Wolffs PF, van den Brule AJ. An evaluation of three processing methods and the effect of reduced culture times for faster direct identification of pathogens from BacT/ALERT blood cultures by MALDI-TOF MS. *Eur J Clin Microbiol Infect Dis*. 2012;31(7):1575-1583.