General Discussion

Initially, antimicrobial control policies were initiated mainly to control costs and not to limit the development of resistance [1-3]. Nowadays, the opposite is true, the costs of most antimicrobial agents being extremely low, and the selective pressure of antimicrobial therapy (AMT) resulting in increased resistance concerns the whole medical world. [4,5].

The purpose of the studies presented in this thesis was to gain more insight into the prescription of AMT and the epidemiology of resistant micro-organisms in hospitals. Based onthese findings a quality improvement program was implemented.

Monitoring of antimicrobial use in hospitalised patients

The most common method to monitor the use of AMT is based on consumption data, expressed as Defined Daily Doses (DDD) per patient day [6]. Filius et al. adapted this method and concluded that DDD per 100 admissions was an additional measurement, which was especially useful to compare antimicrobial consumption rates over time or between different hospitals,geographical regions or countries, because it was less sensitive to changes in hospital resource indicators [7]. These methods provide information about the quantitative use of antimicrobials, however no information about the appropriateness of use is provided.

We developed a point prevalence survey on antimicrobial use in which the appropriateness of use of AMT can be judged on a patient level. In addition, demographic and infection-related data can be collected to study potential determinants of inappropriate use of AMT. Determinants of inappropriate use of AMT can subsequently be used to develop targeted interventions. By repeating the prevalence surveys, the effectiveness of interventions can be measured. The repeated prevalence surveys in combination with targeted interventions results in a true quality circle for AMT in hospitals.

There are three major requirements for the successful implementation of a prevalence survey of AMT. First, there have to be clear and explicit antimicrobial prescription guidelines in the hospital. A second requirement is the knowledge and decisiveness of the persons who are judging the therapy; and finally, the availability of and access to medical records (**chapter2.2**). The assessment of appropriateness of AMT on a patient level can be very complicated. It is therefore important that the local guidelines are explicit in their choices. These guidelines should be used strictly when judging the appropriateness.

In **chapter 2.1** it is shown that the prevalence surveys were successfully performed in our hospital, with a very low proportion of cases in which no judgment could be made. However, when this method was implemented in other hospitals, many hospitals had a relatively high percentage of cases in which no judgment could be given (**chapter 2.2**). More intensive trainingis necessary to implement this method in other hospitals.

The prevalence surveys described in this thesis (**chapter 2.1 and 2.2**) were performed in a country with an extremely low use of AMT [8]. Nevertheless, a substantial proportion of the patients on AMT did not require AMT. The conclusion is that there is enough room for improvement of AMT and that prevalence surveys are useful tools to measure the amount and appropriateness of AMT.

Incidence and transmission of Highly Resistant Micro-organisms (HRMO)

The original concept of antimicrobial resistance was that resistance was a result of spontaneous mutation and subsequent selection by antimicrobial use [9]. The widespread development of multi-drug resistance in many species of bacteria led scientists to believe that other mechanisms presumably play a role as well. Horizontal gene transfer is a process whereby genetic material contained in small packets of DNA can be transferred between individual bacteria [10]. Nowadays this is considered the most important mechanism for acquired resistance in clinically relevant species. In general antimicrobial use does not actively induce resistance. It provides a selective advantage of resistant strains by suppressing the normal flora [11,12]. Some antimicrobial agents, e.g. fluoroquinolones may play a more active role by stimulating gene transfer [13,14].

A restrictive antibiotic policy is considered essential to maintain low resistance rates. Another factor essential to limit the rate of transmission is an active infection control program. This consists mainly of good hand hygiene compliance, isolation of patients infected with resistant bacteria, screening high risk patients and contact tracing in case of unexpected findings.

Theoretically the incidence of Highly Resistant Micro-organisms (HRMO) in hospitals is influenced by the use of AMT and by the infection control policy. A high incidence is most likely when the use of AMT is high and infection control is poor. The effects of these measures have hardly been studied in clinical practice.

Surveillance of Highly Resistant Micro-organisms (HRMO)

A clear definition for resistance is required to determine the incidence of HRMO. Most studies mention the percentage of resistant strains against individual antimicrobial agents [15,16].However, multidrug resistance is the true problem that hampers clinical management of patients and this has to be taken into account as well. It is almost impossible to compare incidences of HRMO, because of the variation in definitions used. One study used the criterion of being susceptible to less than two groups of bactericide antibiotics [17] and one defined “pan-resistance (i.e. decreased susceptibility to cefepime, ceftazidime, imipenem, meropenem,piperacillin/tazobactam, ciproflocacin and levofloxacin)” [18].

In the past years the international medical community (CDC, IDSA, ECDC and WHO), has attempted to highlight the antimicrobial-resistance problem, including the need for accurate surveillance. Nevertheless a general agreement about a universal definition of multi-drug resistant has not been obtained [16,19-21]. The definition used in the National Dutch guidelineis based on both single-drug resistance and multidrug resistance,

depending on the impact forindividual patients and clinical decisions [22,23]. This definition covers the entire spectrum of clinically relevant bacteria (excluding Mycobacterium species).

This definition was used in the surveillance studies in **chapter 3**. In a single-centre study, performed in a large teaching hospital (**chapter 3.2**), we determined an incidence density of43 HRMO / 100,000 patient days, this included Highly Resistant – Gram negative Rods (HR-GNR),Penicillin Non-susceptible S. pneumoniae (PNSP) and Methicillin-Resistant *S. aureus* (MRSA).The majority were HR-GNR, with an incidence density of 35/100,000 patient days.

Molecular typing of the strains was used to investigate the genetic relatedness of HRMO. Analyses of epidemiologically linkage within the window period indicated that the rate of transmission for HR-GNR and PNSP was relatively low (Transmission Index of 0.05 and 0.29respectively). Transmission of MRSA was not observed. Transmission of PNSP always occurred when the patient had not been placed in isolation, because laboratory results were not available yet. Once droplet precautions had been installed no further transmission was observed. This illustrates the need for more rapid laboratory detection of resistant isolates.

In HR-GNR, besides the transmission of the bacterium (HR-GNR) itself, the transmission of transmissible plasmids and integrons may also play a role. In the study, **described in 3.3**, the role of integron transmission was investigated. The transmission index of integrons was nearly twice the transmission of the HR-GNR themselves. This shows that, al least in this hospital, the contribution of horizontal gene transfer to the spread of resistance is greater than that of horizontal spread of bacteria. Only a few studies have investigated the role of mobile genetic elements in the population dynamics of bacterial resistance. The results of this study show that more attention should be directed towards the role of mobile genetic elements.

To obtain more insight in the occurrence of HRMO, and especially HR-GNR, and their transmission in Dutch hospitals a multi-centre study was initiated, the TRIANGLe study (**chapter3.4**). The design was based on the single-centre study, described in **chapter 3.2**. All hospitals, university-, teaching- and non-teaching hospitals, used the same set of definitions for HR-GNR, as defined in the Dutch national Guideline [22].

The mean observed incidence density of HR-GNR was 43/100,000 patient days and a large range was observed in the 18 hospitals that participated (range 8 – 123 per 100,000 patientdays; mean 43; SD 38). The only independent determinant associated with the incidence of HR-GNR was staying in a university hospital, which is considered to be a surrogate marker for the complexity of the patient population and the complexity of care provided.

In the general hospitals no transmission of HR-GNR was observed. The adjusted Transmission Index (adjusted for the proportion of available HR-GNR isolates) in the other hospitals ranged from 0.0 through 0.2 (mean 0.06; SD 0.05). No independent determinants associated with a higher transmission were identified.

In both the single- and the multicentre study, described in **chapter 3.2 and 3.4** we proved that nosocomial transmission of HR-GNR was effectively controlled in all hospitals using transmission-based precautions. Although the transmission index varied considerably between the participating hospitals, it was always far below the epidemic level (≥1.0).

The proportion of HR-GNR that was caused by nosocomial transmission of bacteria was only a minority of all HR-GNR found (**chapter 3.2 and 3.4**). Therefore the main source of HR-GNR was likely to be the endogenous flora of the patient already present on admission to the hospital[24,25]. This implicates that there probably are community reservoirs of resistance that have to be identified to remain in control of antimicrobial resistance in the future.

Focussing on fluoroquinolone use

The prevalence survey in **chapter 2.1** identified the use of fluoroquinolones as an independent determinant of inappropriate AMT. In **chapter 3.1**, it is shown that fluoroquinolones are associated with a more rapid development of resistance than beta-lactam antibiotics. Based on these results, it is concluded that fluoroquinolones are more prone to induce resistance in microorganisms than beta-lactams. In 1987, Philips et al. already reported that the use of fluoroquinolones promotes horizontal dissemination of antimicrobial resistance genes by activating an SOS response, which was later confirmed by Beaber et al. [13,14]. This may explain why the use of fluoroquinolones results in an extremely rapid dissemination of resistance genes. This is worrying, considering the increase of fluoroquinolone use and the large proportion of inappropriate use of fluoroquinolones (**chapter 2.1 and 2.2**). In 1999, only 11 years ago, Thomson reported on the global epidemiology of resistance. He concluded that the results of sensitivity testing to ciprofloxacin were encouraging and he challenged the users to minimise the use[24]. Today, the highest amount of fluoroquinolone use in Europe in outpatients is found in Greece (around 1.0 DDD/100 inhabitants/day in 2004). This is about 10 times more than in the Netherlands (0.1 DDD/100 inhabitants/day in 2008) [8,27]. Fluoroquinolone resistance was found in 64% of all strains of Klebsiellapneumoniae isolated from blood cultures in Greece. In the Netherlands only 7% of all K. pneumoniae strains isolated from blood cultures were fluoroquinolone resistant in 2008 [28].

Although the use of fluoroquinolones in The Netherlands is relatively low, we identified fluoroquinolones as the most important determinant of inappropriate use of AMT. The use of fluoroquinolones is increasing and the resistance rates are going up. In the HR-GNR collection, obtained during the TRIANGLe study, more than half of the Enterobacteriaceae were resistantto ciprofloxacin (**chapter 3.4**). Considering these findings an intervention was started to reduce the use of fluoroquinolones.

Improving antimicrobial use by interventions

The success of interventions to improve AMT depends on several parameters. Firstly, the support from the hospital director or management before starting an intervention. The intervention studies in chapter 4 were financially supported by the hospital. A business case was made predicting that the standardisation of the protocol for peri-operative prophylaxis in surgical procedures and the ciprofloxacin SWITCH project would result in cost savings that were sufficient to fund the personnel costs for all projects in this thesis. These assumptions were confirmed afterwards.

A second essential factor for a successful intervention is a dedicated project-coordinator, who coordinates a multidisciplinary team (i.e. Microbiologist, Pharmacist, Physicians) [29,30].

Thirdly, the intervention protocols and the antimicrobial prescription guidelines should be as straight forward as possible. Both the content and the implementation in the clinical setting should be simple. In the study described in **chapter 4.1**, it was likely that the simplicity of the guideline, in combination with personal instruction, were critical success factors for the implementation and the improvement on timing of prophylaxis. In the ciprofloxacin interventions(**chapter 4.2**), again, it was the simplicity of the procedure (SWITCH according to 3 criteria)and the personal instruction (one-on-one education & feedback strategy), which made the intervention successful. Personal approach with feedback is the fourth factor that improves the outcome of an intervention [31,32]. Finally, education is an important aspect and this should be targeted at learning needs and include interactive educational activities. [33]. All these conditions were met in the study described in chapter 4.2.

The effects of the interventions can be measured using a quasi-experimental design (before-after measurement), as performed in the peri-operative antimicrobial prophylaxis intervention (**chapter 4.1**). Use of this methodology might overestimate or underestimate the effect of an intervention because natural trends are not taken into consideration. A study design using interrupted time series with segmented regression allows for both stepwise changes and changes in trends (used in the ciprofloxacin intervention study in **chapter 4.2**) [34-36]. This is a useful method to measure the effect of the interventions on the use of AMT.

The intravenous use of ciprofloxacin was reduced with 71 prescribed daily doses (PDD) per month (95% CI: 47-97) in association with a SWITCH intervention and the total use of ciprofloxacin(iv and oral) was reduced with 107 PDD per month (95% CI: 56-158) using a bundle of interventions. The interventions that were most likely to have had an effect on the overall use were the introduction of a new antimicrobial guideline and an educational program. Although there was a clear effect on the amount of use, the effect on the observed resistance rates is more complex as the relations are probably non-linear or indirect [37]. Nevertheless, the model indicated that the reduced use of fluoroquinolones had a significant effect on the increasing trend of fluoroquinolone resistance.

Before the start of the bundle of interventions the ciprofloxacin resistance rate in *E. coli* was increasing with 4.6% per year. The statistical model revealed a significant stepwise decrease associated with the bundle of interventions on ciprofloxacin use. This is one of the few studies that found an effect of reduced use of AMT on resistance rates. Considering the uncertainty whether reduced use can lower resistance rates once these are established the conclusion must be that it is of utmost importance to prevent the development of resistance at a very early stage.

Conclusions

In conclusion, the prevalence surveys described in this thesis are useful methods to measure the amount and the appropriateness of AMT. Determinants of inappropriate use were identified and served as targets for interventions. The effect of the interventions can be measured by ongoing prevalence surveys.

Surveillance of HRMO in a teaching hospital revealed that the Intensive Care Unit was a main determinant for the presence of HRMO, and that the transmission of HRMO could be largely prevented using transmission precautions. A multi-centre study, based on the same method, was performed to determine the incidence and transmission of HR-GNR and the relation with infection control precautions, type of hospital (university, teaching, non-teaching) and antimicrobial use. It showed that there was considerable variation of the incidence density that was related to the complexity of the patient population and of the care provided. Nosocomial transmission of HR-GNR was effectively controlled in all hospitals and played a minor role. Additional research on the presence of integrons showed that the contribution of horizontal gene transfer to the spread of resistance is larger than that of nosocomial transmission of bacteria.

Finally, a bundle of interventions that was implemented to improve the use of fluoroquinolones resulted in substantial cost-savings as well as a marked reduction in the hospital use of fluoroquinolones. Furthermore, the reduction in fluoroquinolone use was associated with a change in the increasing trend of the observed resistance against ciprofloxacin.

The studies in this thesis provide information on the relation between the use of antimicrobials, infection control and antimicrobial resistance in hospitals. They can be used as a benchmark for others and to monitor future trends. It was shown that even in a setting with a relatively low use of antimicrobial agents, substantial improvements can be achieved.

Considerations for the future

In Europe, the ECDC published a report called “The bacterial challenge: time to react”, in which the yearly burden of infections due to selected antibiotic-resistant bacteria were estimated [16].The estimated number of additional deaths and hospital days, due to bloodstream infections caused by third-generation cephalosporin-resistant *E. coli* and third-generation cephalosporin resistant *Klebsiella spp.*, was 8,000 deaths and 566,000 hospital days (EU Member States, Iceland and Norway in 2007).

During the TRIANGLe study period, 70 highly resistant *E. coli* and *Klebsiella spp*. were recovered from blood cultures. Using the Dutch hospital resource indicators, obtained from the Dutch Hospital Data information office, the TRIANGLe findings can be converted to a national figure for the Netherlands of 605 bloodstream infections with highly resistant *E. coli* and *Klebsiella spp*. [38]. Using the estimates for attributable mortality and costs from the ECDC, this result in 94 additional deaths and 6,674 additional hospital days. Compared to the total additional deaths and hospital days in Europe, the Dutch figure is just over one percent. This figure might be an overestimation because of the large contribution of university hospitals in this study. However, in the Netherlands, the incidence of HR-GNR is increasing rapidly, and so will the hospital costs and the number of deaths due to an infection with a resistant micro-organism [27].

We can conclude that the guidelines for infection control and the restrictive antimicrobial policy in the Netherlands are effective. To maintain this situation, the current policy should be continued but additional measures are needed to remain in control. New research has to be initiated to define sources of resistant micro-organisms outside the hospital, to elucidate the transmission dynamics of mobile genetic resistance elements and to investigate new resistance mechanisms, like metallo carbapenemases. Our results can be used to optimise current control strategies and to monitor the development of resistance in the future.