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Secular trends in nosocomial bloodstream infections: antibiotic-resistant bacteria increase the total burden of infection

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Running title: Trends in nosocomial bloodstream infections

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40-word summary: In this international study we demonstrate that increasing rates of nosocomial bloodstream infections caused by antibiotic-resistant bacteria do not replace

infections caused by antibiotic-susceptible bacteria, but come on top of these infections, thereby increasing the total burden of disease.

ABSTRACT

Background It is unknown whether rising incidence rates of nosocomial bloodstream infections [BSIs] caused by antibiotic-resistant bacteria [ARB] replace nosocomial BSIs caused by antibiotic-susceptible bacteria [ASB], leaving the total BSI incidence rate unaffected.

Methods We investigated temporal trends in annual incidence densities [events/100 000 patient-days] of nosocomial BSIs caused by methicillin-resistant *Staphylococcus aureus* [MRSA], ARB other than MRSA [non-MRSA ARB], and ASB in 7 ARB-endemic and 7 ARB-non-endemic hospitals between 1998 and 2007.

Results 33 140 nosocomial BSIs (14% caused by ARB) yielded 36 679 microorganisms. From 1998 to 2007, the MRSA incidence density increased from 0.2 to 0.7 (annual increase 22%) in ARB-non-endemic hospitals, and from 3.1 to 11.7 (annual increase 10%) in ARB-endemic hospitals (p=0.2), increasing the incidence density-difference between ARB-endemic and ARB-non-endemic hospitals from 2.9 to 11.0. The non-MRSA ARB incidence density increased from 2.8 to 4.1 (annual increase 5%) in ARB-non-endemic hospitals, and from 1.5 to 17.4 (annual increase 22%) in ARB-endemic hospitals (p<0.001), changing the incidence density-difference from -1.3 to 13.3. Trends in incidence densities of ASB were similar in both groups (p=0.7). With annual increases of 3.8% and 5.4% of all nosocomial BSIs in ARB-non-endemic and ARB-endemic hospitals, respectively (p<0.001), the overall incidence density-difference of 3.8 increased to 24.4.

Conclusion Increasing nosocomial BSI rates due to ARB occur in addition to infections caused by ASB, increasing the total burden of disease. Hospitals with high ARB-infection rates in 2005 had an excess burden of BSI of 20.6/100 000 patient-days in a 10-year period, mainly caused by infections with ARB.

INTRODUCTION

Rates of nosocomial bloodstream infections [BSI] caused by antibiotic-resistant bacteria [ARB] are increasing worldwide.[1] Yet, associated changes in total burden of disease and the dynamical interaction between ARB and antibiotic-susceptible bacteria [ASB] have not been quantified accurately.[2] For instance, one could hypothesize that the frequent use of antibiotics in hospitalised patients creates an ecological niche for ARB that replace ASB, without increasing the total burden of disease. Alternatively, ARB and ASB may not compete, and, thus, increasing infection rates caused by ARB incur an additive burden. Such information is critical for quantifying the health-economic effects of antimicrobial resistance and demonstrating the benefits of infection control.[3] Most longitudinal studies on the epidemiology of ARB addressed a single pathogen, frequently with comparison to its antibiotic-susceptible variant, but failed to include all other pathogens.[2] To assess whether incidences remain stable or increase, one should adjust for changes in duration of hospital stay over time. Hence, incidence density (i.e., incidence rate; number of events per number of patient-days) will quantify the problem of antimicrobial resistance more accurately than crude numbers of events.[4]

We quantified temporal trends in the microbiologic aetiology of nosocomial BSI due to ARB and ASB in 14 hospitals in Europe, North and South America over a 10-year period. We categorized hospitals as those with and without endemic ARB. We aimed to determine if ARB replace ASB, or if (and to what extent) they are additive to the total burden of nosocomial BSIs.

METHODS

Ethics statement

Institutional review boards in most of the participating hospitals did not require a formal protocol review because this study was retrospective, thus, did not affect patient care, and data were deidentified. Oxford data came from the Infection in Oxfordshire Research Database, approved by the Oxford Research Ethics Committee (09/H0606/85) and the UK National Information Governance Board (5-07(a)/2009).

Study design, study setting and study population

We performed a cohort study of patients with microbiologically confirmed nosocomial BSI by linking deidentified databases from the hospital information systems and the microbiology laboratories of the participating hospitals. Hospitals were eligible if they could provide (1) data on positive blood cultures, including susceptibility profiles, for at least seven consecutive years, (2) numbers of hospital admissions and lengths of stay for the same period, and (3) the hospital day during which blood cultures were obtained. Patients admitted to ambulatory care and psychiatric units were excluded. Species identification and susceptibility testing were performed according to local guidelines and procedures.

We initially used the 2005 figures of MRSA reported by existing surveillance programs to categorize hospitals based on their countries proportion of MRSA among *S. aureus* invasive infections, being more or less than 0.10, as having high or low infection rates of ARB.[5-7] After data collection we quantified the proportion of MRSA and ARB isolates and the incidence densities of nosocomial MRSA BSI and ARB BSI in 2005.

Data collection and variables of interest

The hospitals' microbiological databases were linked to patient-administrative systems, thereby providing a database with all patient admissions. The database included microbiological results of all positive blood cultures obtained, and data on gender, age,

department of admission, and length of stay before nosocomial BSI acquisition. Hospital departments were categorized as surgery, medicine, and mixed departments and patient care units were categorized as intensive care units or regular wards.

Definitions

Bloodstream infection: Isolation of bacteria or fungi from at least one blood culture set. Microorganisms typically considered to be skin flora (coagulase-negative staphylococci, *Micrococcus* species, *Bacillus* species, diphteroids (corynebacteria or propionibacteria) were considered to be probable contaminants and were excluded.

Nosocomial BSI: BSI occurring more than 48 hours after hospital admission and in patients who did not have documented BSI with the same microorganism during the first 48 hours after admission.

Polymicrobial BSI: BSI with more than one microorganism in a single set of blood cultures or in different blood culture sets obtained within 48 hours of the first positive blood culture.

New episode of BSI: A BSI caused by a different microorganism more than 48 hours after the previous nosocomial BSI or by the same microorganism more than 30 days after the previous BSI.

Antibiotic-resistant bacteria [ARB]: definitions of ARB were based on a Dutch guideline (resistance criteria for isolation of patients; Supplementary Table 1);[8] these bacteria were subdivided in MRSA and non-MRSA ARB.

Antibiotic-susceptible bacteria [ASB]: Bacteria that did not meet the definition of ARB according to the Dutch guideline.

Statistical analysis

Data were analysed from each hospital independently, and from aggregate data from two groups, ARB-endemic and ARB-non-endemic hospitals. While the participating hospitals submitted data from somewhat different time periods, we assessed trends in the rates of nosocomial BSI by pooling results from the 10-year period 1998 to 2007. We did a sensitivity

analysis by repeating calculations from the period 2000 to 2005 for which all participating hospitals submitted data (Supplementary Figure 1). In a second sensitivity analysis, we excluded hospitals with the most extreme nosocomial BSI incidence densities (Porto Alegre and Utrecht for the ARB-endemic and ARB-non-endemic hospitals, respectively).

We assessed changes in the incidence densities of nosocomial BSI to describe changes in the overall burden of disease over time, and we assessed changes in the incidence densities of cultured microorganisms (including each microorganism in polymicrobial nosocomial BSI) to describe changes in burden of nosocomial BSI infections caused by MRSA, ARB, and ASB. Incidence densities were calculated as the number of events per 100 000 patient-days. We modelled temporal trends of incidence densities using Poisson regression, presenting yearly change in incidence density as a rate ratio [RR] with a 95% confidence interval. To determine whether differences between RRs from ARB-endemic and ARB-non-endemic hospitals were statistically significant, we calculated the p-value for heterogeneity.

We repeated calculations with number of events per 10 000 admissions (cumulative incidence), to allow for the possibility of non-parametric changes in length of stay after the onset of nosocomial BSI compared to overall length of stay over the study period, leading to an overestimation of increased burden of disease when expressed as incidence densities (Supplementary Table 2).

We used X^2 tests for dichotomous variables, univariable logistic regression for categorical variables, and Mann Whitney U tests for continuous, non-normally distributed variables to analyze relations between patients and ARB endemicity. The data were analyzed using SPSS version 15.0 (SPSS, Chicago, Ill., USA) and R version 2.6.0.

RESULTS

Hospital characteristics

Fourteen hospitals from nine countries participated: seven hospitals in countries with low proportions of MRSA among *S. aureus* BSI in 2005;[5] the Netherlands (two university hospitals, two general hospitals), Norway (one university hospital), and Sweden (one university hospital, one general hospital), and seven in countries with high proportions of MRSA among *S. aureus* BSI in 2005;[5-7] Germany (two university hospitals), Switzerland (one university hospital), United Kingdom (one university hospital), Republic of Ireland (one university hospital), United States (one university hospital), and Brazil (one university hospital). The observed proportions of MRSA among *S. aureus* nosocomial BSI in 2005 ranged from 0.00 to 0.05 among hospitals in countries with low prevalence of MRSA, and from 0.22 to 0.66 among hospitals in countries with high prevalence of MRSA. The observed incidence densities of nosocomial MRSA BSI in 2005 ranged from 0.0 to 1.1/100 000 patient-days among hospitals in countries with low MRSA rates and from 4.2 to 58/100 000 patient-days among hospitals in countries with high MRSA rates. The observed incidence densities of nosocomial ARB BSI in 2005 ranged from 1.3 to 4.8/100 000 patient-days among hospitals in countries with low MRSA rates (ARB-non-endemic hospitals) and from 9.9 to 91/100 000 patient-days among hospitals in countries with high MRSA rates (ARB-endemic hospitals), which implies that the proportion of MRSA -in these hospitals- is a reliable proxy for ARB BSI.

During the study period, 4 992 357 patients were admitted for a total of 36 391 175 patient-days. The annual number of patient-days increased by 0.3% among ARB-non-endemic hospitals and increased by 0.5% among ARB-endemic hospitals (Supplementary Table 3).

Microbiology

Over the study period, 202 523 positive blood cultures were obtained (not including probable contaminants) during 64 417 BSI episodes (Figure 1), of which 33 140 (51.4%) were

hospital-acquired, yielding 36 679 microorganisms: 9655 (25.9%) grew Enterobacteriaceae, 7367 (22.6%) *S. aureus*, 3673 (11.4%) *Enterococcus* species, 2824 (8.0%) *Streptococcus* species, 508 (1.5%) other Gram-positive species, 1670 (5.3%) *Pseudomonas aeruginosa*, 640 (2.0%) *Acinetobacter* species, 1006 (3.3%) other Gram-negative species, 1666 (4.9%) fungi, 1104 (3.3%) anaerobes, and 3017 (8.3%) episodes were polymicrobial. Of the nosocomial BSIs, 30 178 (91.1%) were a patient's first nosocomial BSI following hospital admission, and the remaining 2962 were patients' second to fifth episodes during the same hospital stay. Nearly fourteen percent (4484/33 140; 13.5%) of nosocomial BSIs were caused by ARB: 18.8% (4040/21 452) in ARB-endemic hospitals compared with 3.8% (444/11 688) in ARB-non-endemic hospitals ($p<0.001$). Nineteen percent (574/3017) of polymicrobial nosocomial BSIs included at least one ARB.

Of nosocomial BSIs, 29 879 (90.1%) occurred between 1998 and 2007 (Table 1). Thus, data from this period were analysed to assess differences in trends between ARB-endemic and ARB-non-endemic hospitals. The sensitivity analysis included 20 272 (61.2%) nosocomial BSIs that occurred from 2000 through 2005 (Supplementary Figure 1).

Patient characteristics

Patients with nosocomial BSI had a median age of 61 years (interquartile range [IQR], 42-73), 59.1% were male, and patients were hospitalized for a median of 13 days (IQR 7-27) when their first positive blood culture was obtained. Thirty-six percent of nosocomial BSI were acquired in a surgical ward, 49.3% in a medical ward, and 14.4% in a mixed surgical and medical ward. Twenty percent of nosocomial BSI were acquired in an intensive care unit. Compared with bacteraemic patients hospitalized in ARB-non-endemic hospitals, patients in ARB-endemic hospitals were younger (59 years [IQR, 40-72] versus 63 years [IQR, 47-74]), had longer lengths of stay before BSI acquisition (14 days [IQR, 7-28] versus 13 days [IQR, 7-25], were more likely to be admitted to a medical ward (51.5% versus 45.0%), and less likely to be admitted to an intensive care unit (18.1% versus 24.1%; all comparisons $p<0.001$).

Incidence rates of nosocomial BSI

Between 1998 and 2007, the average incidence density of nosocomial BSI per hospital ranged from 62.3 to 185.5/100 000 patient-days. The 10-years trend of annual incidence densities increased in twelve hospitals, decreased by 1% in one hospital, and did not change significantly in another one. The increase in incidence densities was mainly due to increased rates of *Enterococcus* spp., anaerobes, and *Candida* spp. in ARB-non-endemic hospitals and to *Enterococcus* spp., Enterobacteriaceae, *Acinetobacter* spp., and *Candida* spp. in ARB-endemic hospitals (Table 1). The incidence density trends for nosocomial BSI caused by ASB, MRSA, and non-MRSA ARB, in the participating hospitals are shown in Figure 2.

From 1998 to 2007, the incidence density of nosocomial BSI caused by MRSA increased from 0.2 to 0.7/100 000 patient-days in ARB-non-endemic hospitals, an annual increase of 22% (95% CI, 6-40%). During the same period, the incidence density of MRSA increased from 3.1 to 11.7/100 000 patient-days in ARB-endemic hospitals, corresponding to an annual increase of 10% (95% CI, 9-12%; Table 2). While the relative rates of increase did not differ significantly between ARB-non-endemic and ARB-endemic hospitals (p=0.2), the MRSA incidence density-difference increased from 2.9 in 1998 to 11.0/100 000 patient-days in 2007. The incidence density of nosocomial BSI caused by non-MRSA ARB increased from 2.8 to 4.1/100 000 patient-days in ARB-non-endemic hospitals, corresponding to an annual increase of 5% (95% CI, 1-9%). In the same period, the incidence density of nosocomial BSI caused by non-MRSA ARB increased from 1.5 to 17.4/100 000 patient-days in ARB-endemic hospitals, an annual increase of 22% (95% CI, 20-25%) (p<0.001; Table 2; Supplementary Figure 2). As a result, the incidence density-difference between ARB-endemic and ARB-non-endemic hospitals for nosocomial BSI caused by non-MRSA ARB increased from -1.3 in 1998 to 13.3/100 000 patient days in 2007.

Trends in incidence densities of nosocomial ASB BSI were similar, with annual increases of 4.5% (95% CI, 4-5%) and 4.2% (95% CI, 4-5%) in ARB-non-endemic and ARB-endemic hospitals, respectively (p=0.7). The overall incidence density-difference between ARB-

endemic and ARB-non-endemic hospitals increased from 3.8 in 1998 (78.1 versus 74.3/100 000 patient-days, respectively) to 24.4/100 000 patient-days in 2007 (130.1 versus 105.7/100 000 patient-days, respectively), fully attributable to infections caused by ARB ($p<0.001$).

Sensitivity analyses evaluating data from 2000 to 2005 and evaluating data that excluded data from the hospitals with the highest rates yielded similar results (data not shown). Moreover, trends in cumulative incidences were comparable to trends in incidence densities (Supplementary Table 2).

DISCUSSION

Based on detailed longitudinal data from 14 hospitals in three continents we have demonstrated that an increasing incidence of nosocomial BSI caused by ARB adds to the total burden of disease without replacing BSI caused by more susceptible bacteria. While the total burden of nosocomial BSI in both cohorts was similar in 1998, the excess increase in incidence rates of nosocomial BSI in ARB-endemic hospitals was 20.6/100 000 patient days in 2007 and almost fully attributable to increased rates of infections caused by ARB.

To the best of our knowledge, this study is the first to conduct integrated trend analyses of all relevant nosocomial pathogens on such a large multicentre dataset. This dataset allowed us to quantify the overall burden of nosocomial BSI caused by ARB and ASB. Although longitudinal changes in incidences and proportions of pathogens causing nosocomial infections have been reported previously, in most studies reported changes in the burden of disease due to ARB reflected the epidemiology of a single pathogen.[2,9] By comparing longitudinal data from hospitals with high and lower rates of nosocomial ARB BSI, we took advantage of a natural experiment that allowed us to observe long-term effects of successful and less successful control of nosocomial spread of ARB. On a global level, this information is critical for assessing benefits of infection prevention and control strategies. In the past years, guidelines have focused specifically on prevention of ARB transmission in hospitals, in particular MRSA.[10-13] Our results stress the importance of successful prevention of all ARB.

Although our study was observational, we feel that the more pronounced increase of BSIs in the ARB-endemic hospitals reflects either differences in infection prevention practices or antibiotic prescription patterns, or both, of the hospitals and their home countries.[14,15] Our findings suggest that hospitals (and their home countries) which effectively prevented emergence of MRSA were also more successful in controlling the more recent emergence of resistance among Gram-negatives.

There are three alternative explanations of our findings that need to be addressed. The first one is an imbalanced change in patient case-mix that may have occurred during the study

period. Such a change in case-mix was not discernible from observed changes in age and length of stay. Moreover, annual changes in incidence rates of ASB BSI were similar in both hospital groups. The second alternative explanation would be that ARB may be more virulent than their susceptible counterparts, but this would contradict the widely accepted view that resistance is associated with reduced fitness, and published studies do not convincingly prove that antimicrobial resistance confers increased virulence on pathogenic bacteria.[16]

Finally, non-parametric changes in length of stay after nosocomial BSI compared with overall length of stay could have caused us to overestimate the burden of disease when expressed as incidence density. However, the cumulative incidence, which is less sensitive to changes in the number of patient-days over time, revealed similar trends.

Our study has several potential limitations. Hospitals were included if they had the availability of an appropriate database, which could have selected hospitals with better surveillance systems and infection control policies. Naturally, results of the various participating hospitals were heterogeneous, which reflects differences in patient populations, local infection prevention measures, hospital organisation, and antibiotic prescribing practices. Nevertheless, Figure 2 demonstrates broadly similar results across our set of hospitals, and does not suggest important ecological biases.

In addition, incidence density analysis in general might be obscured when potential competing events (e.g., in-hospital death and hospital discharge) are not taken into account.[17] However, the incidence density of nosocomial BSIs, as determined in our study, was conditional on patients being alive and hospitalized. Thus, the analysis of incidence densities corresponds to an analysis of the hazard for nosocomial BSI before in-hospital death or hospital discharge.

In theory, hospitals that obtained more blood cultures than others might have higher rates of nosocomial BSI. Since information on negative blood cultures was not part of this study, changes in blood culture practices over time could not be determined.. However, we are unaware of such changes in the participating hospitals. Moreover, we think it is unlikely that such changes differed between hospitals with high and lower rates of nosocomial ARB BSI.

Misclassification may have occurred for some positive blood cultures in patients who were discharged and then readmitted soon thereafter with a BSI, erroneously classified as community-acquired. Furthermore, we may have misclassified some nosocomial BSIs as non-infectious by excluding cultures that grew possible skin contaminants. Although some might have represented true episodes of nosocomial infection, we were unable to identify them as information of clinical signs and symptoms was lacking.[18] However, misclassification would have affected rates of nosocomial BSI in a non-differential manner.

In conclusion, we demonstrated that nosocomial BSI caused by ARB do not replace infections caused by more susceptible bacteria, but rather these infections increase the total burden of disease. This implies that successful control of antibiotic resistance improves patient outcome not only because of lower mortality from better treatable infections, but also because of a reduction, or at least a lower increase, in number of infections.

NOTES

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CONFLICT OF INTEREST

H.S.M. Ammerlaan has received speaking fees from Novartis. S. Harbarth has received consulting fees from 3M and Roche, is a member of the speakers' bureau for Biomerieux, and is a member of the advisory board of Destiny Pharma. The institution of D.W. Crook received per-case funding from Optimer Pharmaceuticals to support trial patient expenses and D.W. Crook also received honoraria from Optimer Pharmaceuticals for participation in additional trial-related meetings. L. Herwaldt has received a research grant from 3M. J.A.J.W. Kluytmans has received speaking fees from 3M, Cepheid and Biomerieux, and is a member of the advisory board of Destiny Pharma, Phico Therapeutics, Pfizer and 3M. E. Lingaas is a member of the advisory board of 3M and has received speakers' honorarium

from Moelnlycke Healthcare. H. Seifert has received research grants from Astellas, Basilea, Bayer, Novartis and Pfizer, has received speaking fees from Astellas, Bayer, Gilead, MSD, Novartis, Oxoid, Pfizer, and Wyeth, and served as a member of the advisory board of 3M, Astellas, AstraZeneca, Baxter, Novartis, and SIRS-Lab. H. Wisplinghoff has received speaking fees from Siemens, BioMerieux, and Bruker. M.J.M. Bonten has received research funding from Novartis and 3M, is a member of the speakers' bureau for Pfizer, and member of the advisory board of 3M and Novartis. All other co-authors have no competing interests to declare.

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FIGURE LEGENDS

Figure 1. Flow diagram

Note: BSI – bloodstream infection; ARB – antibiotic-resistant bacteria; ASB – antibiotic-susceptible bacteria

Figure 2. Trends in incidence densities of microorganisms

Note: lines show trends in incidence densities over the study period for each hospital (ARB endemic versus non-endemic hospitals) contributing data to the analysis. ARB – antibiotic-resistant bacteria; ASB – antibiotic-susceptible bacteria; MRSA – methicillin-resistant *S. aureus*. Light grey dashed line – ARB-endemic hospitals; Dark grey solid line – ARB-non-endemic hospitals. Different y-axis scales were used in each panel.

a. incidence densities of BSI caused by ASB; b. incidence densities of BSI caused by MRSA; c. incidence densities of BSI caused by non-MRSA ARB.

Figure 1. Flow diagram

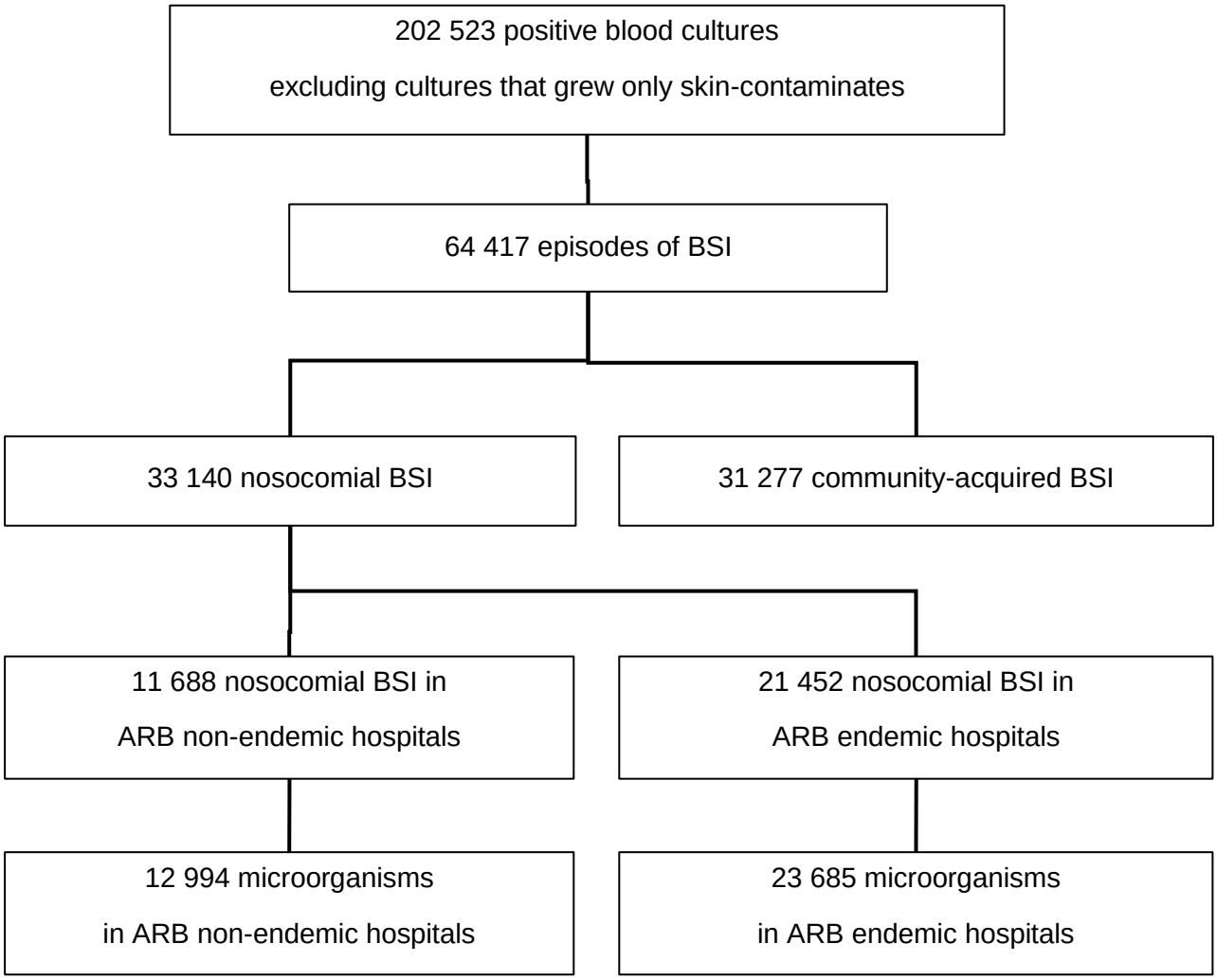


Figure 2. Trends in incidence densities of microorganisms

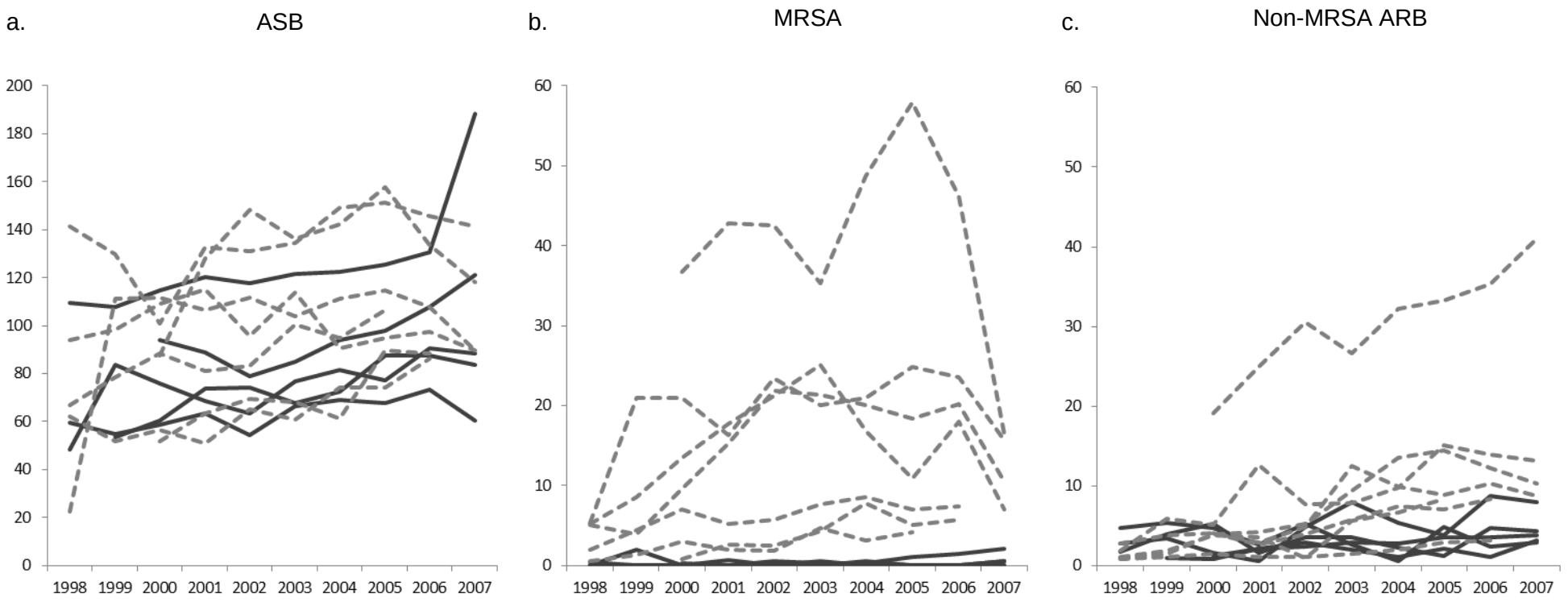


Table 1. Nosocomial bloodstream infections caused by specific microorganisms during the study period 1998-2007 by ARB non-endemic hospitals and endemic hospitals

Microorganism	***ARB non-endemic hospitals***			***ARB endemic hospitals***			***Heterogeneity[1]***
	Total	**ID**	**Trend (ID)** RR (95% CI)	**Total**	**ID**	**Trend (ID)** RR (95% CI)	p-value
Staphylococcus aureus	2216	16.2	1.01 (0.99-1.03)	5113	27.0	1.05 (1.04-1.06)	< 0.001
Enterococcus spp.	1352	9.9	1.07 (1.05-1.09)	3037	16.0	1.06 (1.05-1.08)	0.9
Streptococcus spp.	1100	8.0	1.02 (1.00-1.04)	1417	7.5	1.01 (0.99-1.03)	0.4
Other Gram-positives	198	1.4	1.08 (1.02-1.14)	281	1.5	1.15 (1.10-1.21)	0.06
Enterobacteriaceae	4205	30.7	1.05 (1.04-1.06)	6036	31.9	1.07 (1.06-1.08)	0.005
Acinetobacter spp.	135	1.0	0.94 (0.88-1.00)	626	3.3	1.10 (1.07-1.14)	< 0.001
Pseudomonas aeruginosa	643	4.7	1.05 (1.02-1.08)	1229	6.5	1.05 (1.02-1.07)	0.9
Other Gram-negatives	280	2.0	1.02 (0.99-1.08)	646	3.4	1.05 (1.02-1.07)	0.6
Anaerobes	413	3.0	1.06 (1.02-1.10)	573	3.0	0.99 (0.96-1.03)	0.008
Candida spp.	522	3.8	1.06 (1.03-1.10)	1002	5.3	1.08 (1.05-1.10)	0.5
Polymicrobial	997	7.3	1.05 (1.03-1.08)	1680	8.9	1.04 (1.03-1.06)	0.6

Note: ID – incidence density; Trend (ID) – Secular changes in incidence density (on average, per year) of nosocomial bloodstream infections (number of infections per 100 000 patient-days) from 1998 to 2007, stratified by pathogen; RR – rate ratio of incidence density; CI – confidence interval; spp – species. [1] The statistical significance of the difference between RRs from ARB-endemic and ARB-non-endemic hospitals was assessed by calculating the p-value for the interaction term (calendar year-hospital type). P-values smaller than 0.05 indicate that the trends for ARB endemic and non-endemic hospitals are significantly different.

Table 2. The incidence densities of microorganisms, MRSA, MSSA, non-MRSA ARB and non-MSSA ASB at baseline and end of study

	ARB non-endemic hospitals ID (microorganisms per 100 000 patient-days)				**ARB endemic hospitals** ID (microorganisms per 100 000 patient-days)				**Heterogeneity**[1]
	1998	*2007*	*Increase 1998-2007*	*Trend RR (95% CI)*	*1998*	*2007*	*Increase 1998-2007*	*Trend RR (95% CI)*	*p-value*
Summary of all microorganisms	**74.3**	**105.7**	**31.4**	**1.04 (1.03-1.04)**	**78.1**	**130.1**	**52.0**	**1.05 (1.05-1.06)**	**< 0.001**
ARB	3.0	4.7	1.7	1.06 (1.02-1.10)	4.6	29.1	24.5	1.15 (1.13-1.16)	< 0.001
ASB	71.4	101.0	29.6	1.04 (1.03-1.04)	73.5	101.0	27.5	1.04 (1.03-1.04)	0.7
Staphylococcus aureus	**15.7**	**16.3**	**0.6**	**1.01 (0.99-1.03)**	**14.9**	**26.6**	**11.7**	**1.05 (1.04-1.06)**	**< 0.001**
MRSA	0.2	0.7	0.5	1.22 (1.06-1.40)	3.1	11.7	8.6	1.10 (1.09-1.12)	0.2
MSSA	15.6	15.6	0.06	1.01 (0.99-1.02)	11.8	14.9	3.1	1.00 (0.99-1.02)	0.8
Microorganisms non *S. aureus*	**58.6**	**89.4**	**30.8**	**1.05 (1.04-1.05)**	**63.2**	**103.5**	**40.3**	**1.06 (1.05-1.06)**	**0.03**
Non MRSA-ARB	2.8	4.1	1.3	1.05 (1.01-1.09)	1.5	17.4	15.9	1.22 (1.20-1.25)	< 0.001
- *Enterococcus* spp.	0.0	0.1	0.1	1.33 (1.02-1.84)	0.6	3.1	2.5	1.22 (1.17-1.27)	0.6
- Enterobacteriaceae	2.3	3.3	1.0	1.04 (1.00-1.09)	0.6	10.2	9.6	1.21 (1.18-1.25)	< 0.001
- *Acinetobacter* spp.	0.3	0.1	-0.2	0.81 (0.67-0.96)	0.1	1.9	1.8	1.29 (1.20-1.39)	< 0.001
- *P. aeruginosa*	0.0	0.4	0.4	1.06 (0.96-1.18)	0.1	1.3	1.2	1.19 (1.11-1.27)	0.08
Non MSSA-ASB	55.8	85.4	29.6	1.05 (1.04-1.05)	61.7	86.1	24.4	1.04 (1.04-1.05)	0.5
- *Enterococcus* spp.	8.4	14.2	5.8	1.07 (1.05-1.09)	8.8	11.5	2.7	1.05 (1.03-1.06)	0.09
- Enterobacteriaceae	23.4	37.4	14.0	1.05 (1.04-1.06)	22.0	35.7	13.7	1.05 (1.04-1.06)	0.9
- *Acinetobacter* spp.	0.6	0.7	0.1	0.96 (0.90-1.03)	1.3	4.0	2.7	1.07 (1.03-1.10)	0.007

-	*P. aeruginosa*	3.3	5.2	1.9	1.05 (1.02-1.08)	5.4	6.7	1.3	1.03 (1.01-1.05)	0.3

Note: ID – incidence density; Trend – Secular changes in incidence density (on average, per year) of nosocomial bloodstream infections (number of infections per 100 000 patient-days) from 1998 to 2007, stratified by pathogen; RR – rate ratio of incidence density; CI – confidence interval; MRSA – methicillin-resistant *S. aureus*; MSSA – methicillin-susceptible *S. aureus*; ARB – antibiotic-resistant bacteria; ASB – antibiotic-susceptible bacteria; spp. – species; *P. aeruginosa* – *Pseudomonas aeruginosa*; spp – species.[1] The statistical significance between RRs from ARB-endemic and ARB-non-endemic hospitals was assessed by calculating the p-value for the interaction term (calendar year-hospital type). P-values smaller than 0.05 indicate that the trends for ARB endemic and non-endemic hospitals are significantly different.

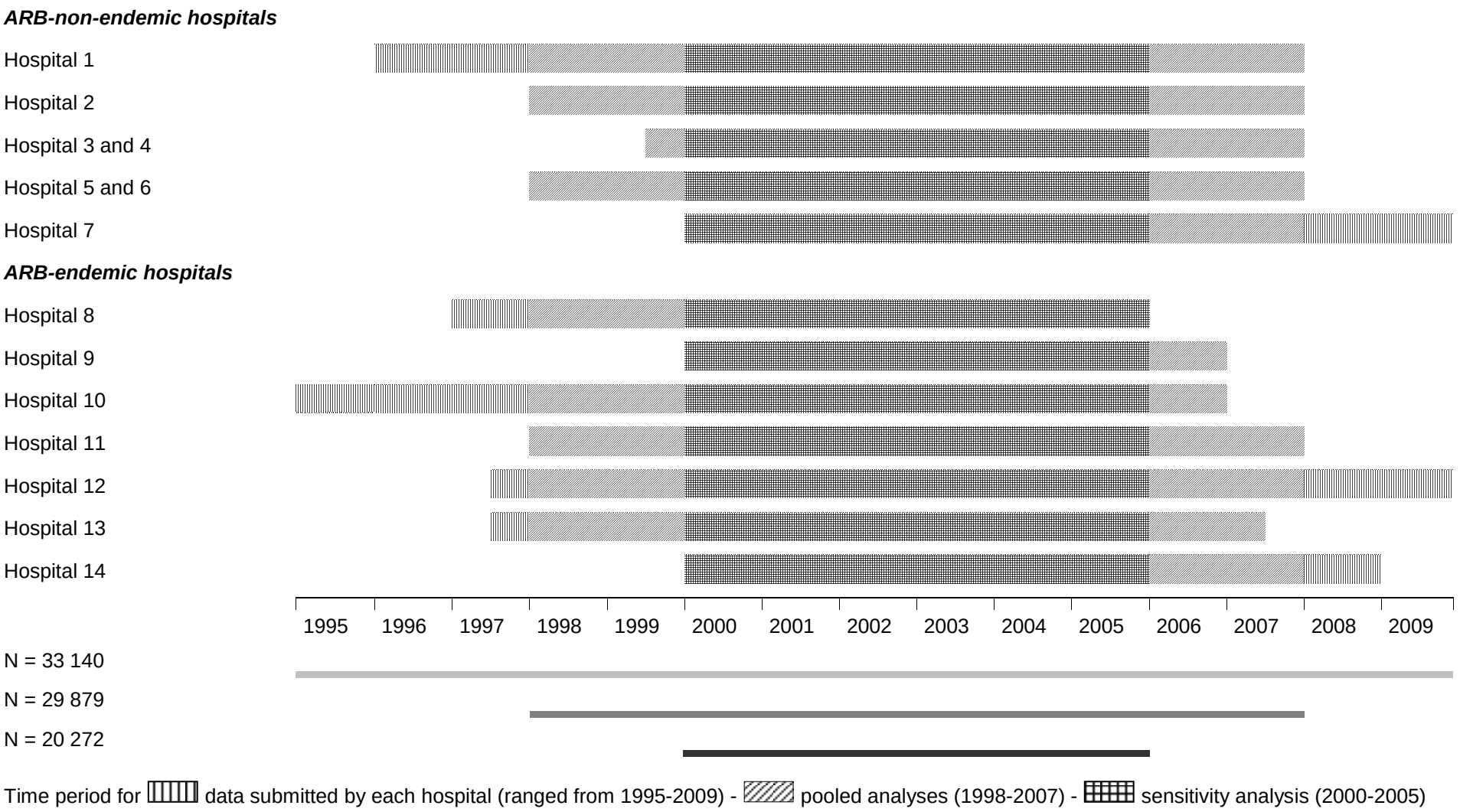
Supplementary Figure 1. Time periods for which hospitals submitted data and time periods considered in different analyses

Note: N – number of nosocomial bloodstream infections included, by time period. [1] Four hospitals provided pooled data (Hospitals 3 and 4, and hospitals 5 and 6).

Supplementary Figure 2. Trends in incidence densities of separate microorganisms

Note: lines show trends in incidence densities over the study period for each hospital (ARB endemic versus non-endemic hospitals). ARB – antibiotic-resistant bacteria; ASB – antibiotic-susceptible bacteria. Grey dashed line – ARB-endemic hospitals; red solid line – ARB-non-endemic hospitals. Incidence densities of BSI caused by: a. methicillin-susceptible *S. aureus*; b. methicillin-resistant *S. aureus*; c. vancomycin-susceptible *Enterococcus* spp; d. vancomycin-resistant *Enterococcus* spp; e. antibiotic-susceptible Enterobacteriaceae; f. antibiotic-resistant Enterobacteriaceae; g. antibiotic-susceptible *Acinetobacter* spp; h. antibiotic-resistant *Acinetobacter* spp; i. antibiotic-susceptible *Pseudomonas aeruginosa*; j. antibiotic-resistant *Pseudomonas aeruginosa.*

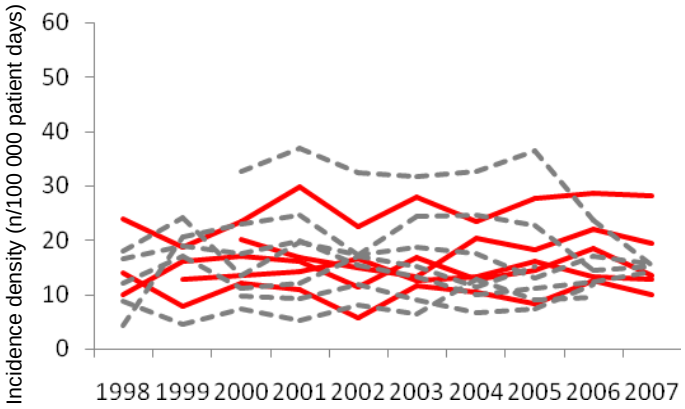
Supplementary Figure 1. Time periods for which hospitals submitted data and time periods considered in different analyses



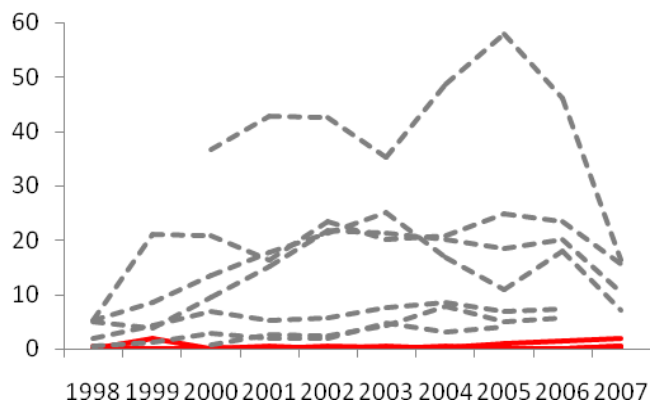
Supplementary Figure 2.

ASB **ARB**

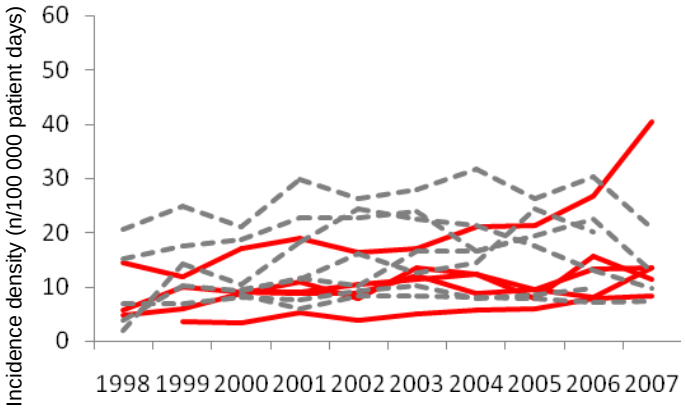
a. methicillin-susceptible *S. aureus*



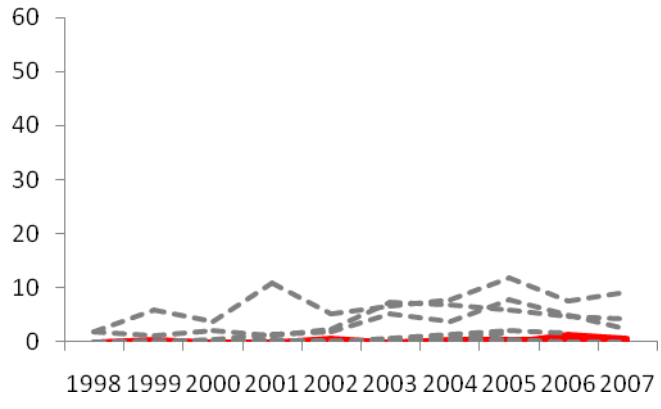
b. methicillin-resistant *S. aureus*



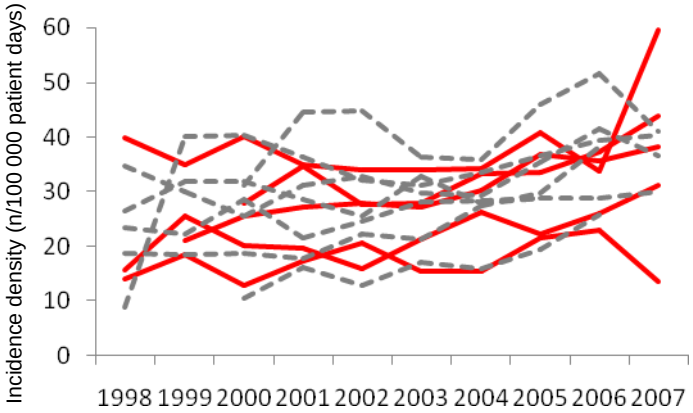
c. vancomycin-susceptible *Enterococcus* spp



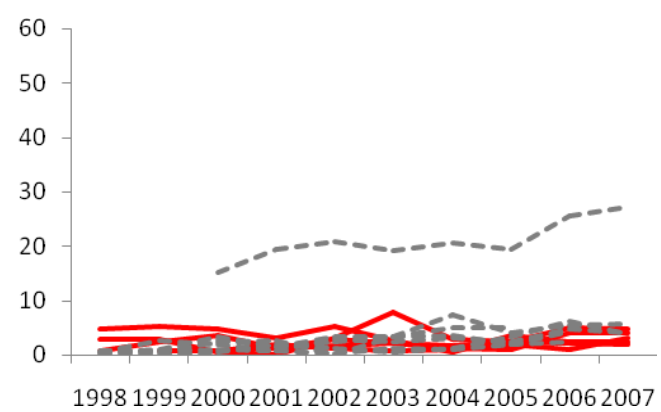
d. vancomycin-resistant *Enterococcus* spp



e. antibiotic-susceptible Enterobacteriaceae

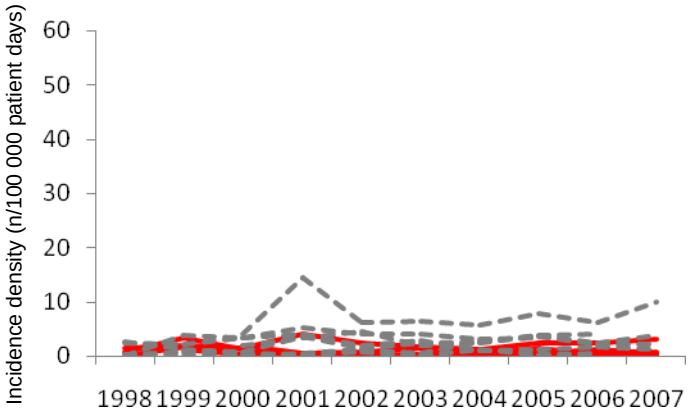


f. antibiotic-resistant Enterobacteriaceae

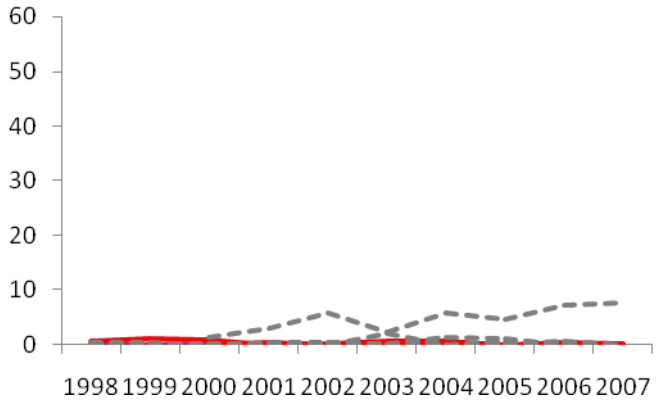


ASB **ARB**

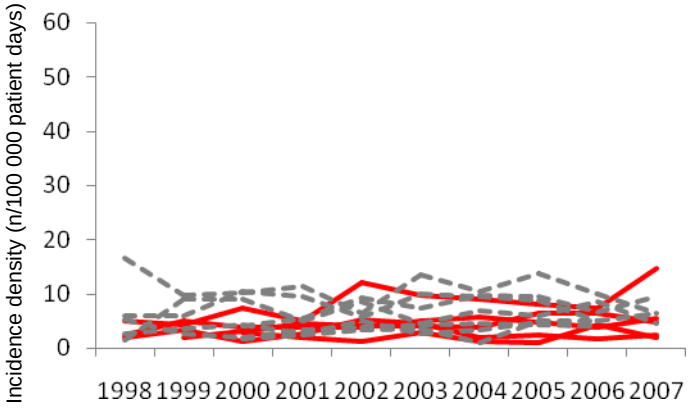
g. antibiotic-susceptible *Acinetobacter* spp



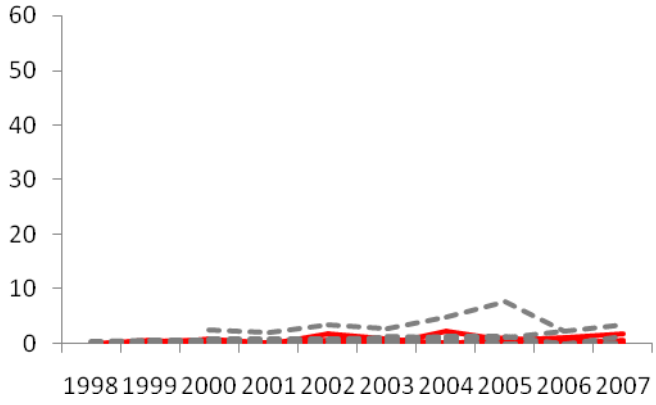
h. antibiotic-resistant *Acinetobacter* spp



i. antibiotic-susceptible *Pseudomonas aeruginosa*



j. antibiotic-resistant *Pseudomonas aeruginosa*



Supplementary Table 1. Definition of antibiotic-resistant bacteria [ARB][1]

Pathogen	***OXA***	***GLY***	***PEN***	***3rdCEP***	***SXT***	***AMG***	***QUI***	***CPM***	***PIP***	***CAZ***
Staphylococcus aureus	A									
Streptococcus pneumoniae		A	A							
Enterococcus faecium		B	B							
Enterobacteriaceae				A[2]	B[3]	B	B	A		
Acinetobacter spp.						B	B	A		B
Pseudomonas aeruginosa						C	C	C	C	C
Stenotrophomonas maltophilia					A					

Note: OXA – oxacillin, GLY – glycopeptides (vancomycin, teicoplanin), PEN – penicillins (benzylpenicillin, aminopenicillin), 3rdCEP – 3rd generation cephalosporins, SXT – trimethoprim/sulfamethoxazole, AMG – aminoglycosides (tobramycin, amikacin, gentamicin), QUI – quinolones (ciprofloxacin, levofloxacin, moxifloxacin), CPM – carbapenems (meropenem, imipenem), PIP – piperacillin, CAZ – ceftazidime. A: resistance to an antibacterial agent from one of the groups in this category is sufficient to define the microorganism as highly resistant; B: resistance to antibacterial agents from at least two of the groups in this category is required to define the microorganism as highly resistant; C: resistance to antibacterial agents from at least three of the groups in this category is required to define the microorganism as highly resistant. [1] Modified according to Kluytmans-VandenBergh.[8] [2] Among Enterobacteriaceae, in-vitro resistance to 3rd generation cephalosporins is a proxy measure for production of extended-spectrum beta-lactamases [ESBL]. [3] Not applicable for *Escherichia coli* and *Klebsiella* spp.

Supplementary Table 2. The cumulative incidences of microorganisms, MRSA, MSSA, non-MRSA ARB and non-MSSA ASB at baseline and end of study

	ARB non-endemic hospitals Microorganisms per 10 000 admissions				**ARB endemic hospitals** Microorganisms per 10 000 admissions			
	1998	*2007*	*Increase 1998-2007*	*Trend RR (95% CI)*	*1998*	*2007*	*Increase 1998-2007*	*Trend RR (95% CI)*
Summary of all microorganisms	59.8	65.4	5.6	1.01 (1.00-1.01)	62.7	91.5	28.8	1.04 (1.04-1.05)
ARB	2.4	2.9	0.5	1.03 (0.99-1.06)	3.7	20.5	16.8	1.13 (1.12-1.14)
ASB	57.4	62.4	5.0	1.01 (1.00-1.01)	59.0	71.0	12.0	1.02 (1.02-1.03)
Staphylococcus aureus	12.7	10.1	-2.6	0.98 (0.96-0.99)	11.9	18.7	6.8	1.04 (1.03-1.05)
MRSA	0.1	0.4	0.3	1.18 (1.03-1.36)	2.5	8.2	5.7	1.09 (1.07-1.11)
MSSA	12.5	9.7	-2.8	0.98 (0.96-0.99)	9.4	10.5	1.1	0.99 (0.98-1.01)
Microorganisms non *S. aureus*	47.1	55.3	8.2	1.01 (1.01-1.02)	50.7	72.8	22.1	1.04 (1.04-1.05)
Non MRSA-ARB	2.3	2.5	0.2	1.02 (0.98-1.05)	1.2	12.3	11.1	1.20 (1.18-1.23)
Non MSSA-ASB	44.9	52.8	7.9	1.01 (1.01-1.02)	49.6	60.5	10.9	1.03 (1.02-1.04)

Note: Trend – Secular changes in cumulative incidence (on average, per year) of nosocomial bloodstream infections (number of infections per 10 000 admissions) from 1998 to 2007, stratified by pathogen; RR – rate ratio of incidence density; CI – confidence interval; MRSA –

methicillin-resistant *S. aureus*; MSSA – methicillin-susceptible *S. aureus*; ARB – antibiotic-resistant bacteria; ASB – antibiotic-susceptible bacteria;

Supplementary Table 3. Overall number of patient-days and the relative change per year during the study period

Hospital	**Study period**	**Patient-days**		**Trend (Patient-days)**	
		Total[1]	Mean[2]	RR (95% CI)	p-value
ARB-non-endemic hospitals	***1998-2007***	***13 683 116***	***1 368 342***	***1.003 (1.00-1.00)***	***< 0.001***
Hospital 1	1996-2007	2 214 335	184 528	0.96 (0.96-0.96)	< 0.001
Hospital 2	1998-2007	3 108 140	310 814	1.01 (1.01-1.01)	< 0.001
Hospital 3 and 4[3]	1999/7-2007	4 752 074	528 008	1.02 (1.02-1.02)	< 0.001
Hospital 5 and 6[3]	2000-2009	3 042 533	304 253	0.97 (0.97-0.97)	< 0.001
Hospital 7	1998-2007	1 574 237	157 424	1.02 (1.02-1.02)	< 0.001
ARB-endemic hospitals	***1998-2007***	***18 943 405***	***1 894 341***	***1.005 (1.01-1.01)***	***< 0.001***
Hospital 8	1997-2005	3 288 943	365 438	0.99 (0.99-0.99)	< 0.001
Hospital 9	2000-2006	2 479 159	354 166	1.01 (1.01-1.01)	< 0.001
Hospital 10	1996-2006	5 223 346	474 850	1.01 (1.01-1.01)	< 0.001
Hospital 11	1998-2007	2 132 070	213 207	1.01 (1.01-1.01)	< 0.001
Hospital 12[4]	1997/4-2003	2 382 692	352 991	1.00 (1.00-1.00)	0.001
	2004-2009	2 621 420	436 903	1.00 (1.00-1.00)	< 0.001
Hospital 13	1997/7-2007/6	1 551 571	155 157	1.00 (1.00-1.00)	0.06
Hospital 14	2000-2008	2 020 655	224 517	1.01 (1.01-1.01)	< 0.001

Note: RR – rate ratio; CI – confidence interval. [1] Total number of patient-days during the study period; [2] Mean yearly number of patient-days during the study period; [3] Data from four hospitals were pooled (Hospital 3 and 4, and hospital 5 and 6); [4] Hospital 12 merged 2 microbiology laboratories in 2004. Data from only 1 laboratory (and its associated patient-days) were available before 2004 and data were available from both laboratories from 2004-2009. Because the total number of patient-days increased in 2004, and data are presented separately for each period.