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ORIGINAL ARTICLES

Surveillance report

ANTIBIOTIC RESISTANCE IN THE SOUTHEASTERN MEDITERRANEAN – PRELIMINARY RESULTS FROM THE ARMED PROJECT

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Sporadic reports from centres in the south and east of the Mediterranean have suggested that the prevalence of antibiotic resistance in this region appears to be considerable, yet pan-regional studies using comparable methodology have been lacking in the past.

Susceptibility test results from invasive isolates of *Staphylococcus aureus, Streptococcus pneumoniae, Escherichia coli, Enterococcus faecium* and *faecalis* routinely recovered from clinical samples of blood and cerebrospinal fluid within participating laboratories situated in Algeria, Cyprus, Egypt, Jordan, Lebanon, Malta, Morocco, Tunisia and Turkey were collected as part of the ARMed project.

Preliminary data from the first two years of the project showed the prevalence of penicillin non-susceptibility in *S. pneumoniae* to range from 0% (Malta) to 36% (Algeria) [median: 29%] whilst methicillin resistance in *Staphylococcus aureus* varied from 10% in Lebanon to 65% in Jordan [median: 43%]. Significant country specific resistance in *E. coli* was also seen, with 72% of isolates from Egyptian hospitals reported to be resistant to third generation cephalosporins and 40% non-susceptible to fluoroquinolones in Turkey. Vancomycin non-susceptibility was only reported in 0.9% of *E. faecalis* isolates from Turkey and in 3.8% of *E. faecium* isolates from Cyprus.

The preliminary results from the ARMed project appear to support previous sporadic reports suggesting high antibiotic resistance in the Mediterranean region. They suggest that this is particularly the case in the eastern Mediterranean region where resistance in *S. aureus* and *E. coli* seems to be higher than that reported in the other countries of the Mediterranean.

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Introduction

Data from the European Antimicrobial Resistance Surveillance System (EARSS) [www.earss.rivm] indicate that the highest levels of antibiotic resistance have been found within the Mediterranean countries participating in the system. On the other hand, information about the prevalence of antimicrobial resistance in the non-European countries of the southern and eastern Mediterranean has, in the past, been sparse. Nevertheless, high levels of resistance have been reported in *Streptococcus pneumoniae* [1,2], *Staphylococcus aureus* [3] as well as within species of the Enterobacteriacae [4,5]. Unfortunately, besides being few in number, these studies have been totally unrelated, using different methodologies and, as a result, are difficult to compare [6].

This deficiency has been addressed by the Antibiotic Resistance Surveillance & Control in the Mediterranean Region (ARMed) project [www.slh.gov.mt/armed] which began in January 2003,7 and is funded by the European Commission under the INCOMED programme of the DG Research Fifth Framework Protocol (ICA3-CT-2002-10015). Over its four year funding period, this study is documenting the prevalence of antibiotic resistance in southern and eastern Mediterranean countries, as well as attempting to investigate potential factors such as antibiotic consumption and infection control. We report on the midway findings of ARMed-EARSS, the resistance epidemiology subcomponent of the project.

Methodology

ARMed-EARSS collects susceptibility test results from invasive isolates of *Staphylococcus aureus*, *Streptococcus pneumoniae*, *Escherichia coli*, *Enterococcus faecium* and *faecalis* routinely isolated from clinical samples of blood and cerebrospinal fluid in the participating laboratories situated in Algeria, Cyprus, Egypt, Jordan, Lebanon, Malta, Morocco, Tunisia and Turkey. These laboratories are asked to send information only about the first isolate of each organism from each patient. ARMed-EARSS uses almost identical protocols to those adopted and validated by the EARSS project, enabling comparison between data from the two projects. The laboratories follow their routine procedures and breakpoints, which in 86.8% of the participants were based on CLSI (formerly NCCLS) guidelines.

S. aureus testing determines oxacillin susceptibility by an oxacillin screen plate (6 mg/l) or alternatively, by an oxacillin disk test (1 µg or 5 µg) and a cefoxitin disk. ARMed laboratories screen invasive S. pneumoniae isolates for oxacillin resistance which, when found to be non-susceptible, is confirmed and determined to intermediate or high-level resistance to penicillin by determination of minimum inhibitory concentration (MIC) using E-test (AB Biodisk, Solna, Sweden). The protocol for *E. coli* susceptibility testing requires disc diffusion testing, amongst others, of a fluoroquinolone (ciprofloxacin and/or ofloxacin) and a third-generation cephalosporin (cefotaxime or ceftriaxone and/or ceftazidime). E-test confirmation is requested in cases of resistance to third generation cephalosporins in *E. coli* and vancomycin in enterococci. ARMed-EARSS protocols are accessible at the ARMed website (http://www.slh.gov.mt/armed/earss.asp). The website also includes an interactive function where maps for specific drug-bug combinations for any of the participating countries can be generated, as specified by the user.

To assess the reliability and comparability of susceptibility test results, two external quality assurance (EQA) exercises were performed in September 2003 and 2004 by all ARMed participating laboratories. These exercises were performed in collaboration with UK NEQAS (United Kingdom National External Quality Assessment Service) and undertaken concurrently with those run by EARSS for their laboratories.

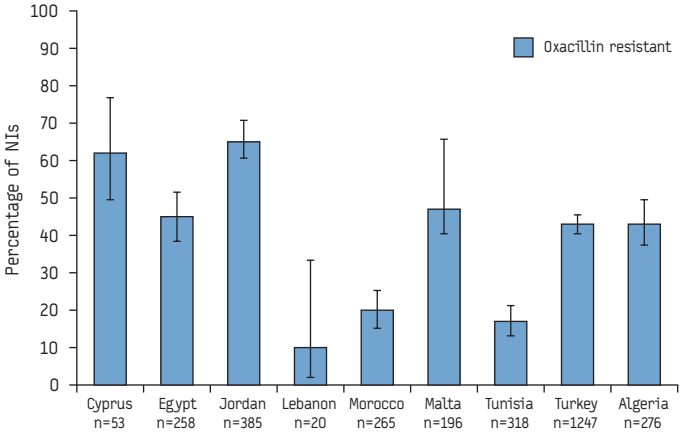
Results

A total of 5883 isolates were investigated, as reported in the first 21 months of the project by the 53 participating ARMed laboratories, which in turn serve 60 hospitals. Of these, 3017 (51.3%) were *S. aureus*, 1567 (26.6%) were *E. coli*, 745 (12.7%) were *S. pneumoniae*, 390 (6.6%) were *E. faecalis*, and 164 (2.8%) were *E. faecium*. Resistance to penicillin in isolates of *S. pneumoniae* was reported from all the collaborating countries except for Malta and Cyprus [FIGURE 1]. When only data from countries reporting at least 10 isolates during the study period were evaluated, the prevalence of penicillin non-susceptibility *S. pneumoniae* ranged from 0% (Malta) to 36% (Algeria) (median: 29% [interquartile range 18% – 33%]). Except for Tunisia, macrolide non-susceptibility in *S. pneumoniae* was generally lower than penicillin non-susceptibility for each country, particularly in Turkey, where the difference was statistically significant (P=0.001).

There was considerably greater variability for resistance within *S. aureus* isolates [FIGURE 2]. Percentages of oxacillin resistance - used as a marker for methicillin resistant *Staphylococcus aureus* (MRSA) - varied from 10% in Lebanon to 65% in Jordan (median: 43% [IQR: 20 – 47%]). The most common resistance pattern showed co-resistance to methicillin, erythromycin and gentamicin. These isolates were particularly evident in Turkey where they accounted for more than a third of all *S. aureus* isolates.

FIGURE 2

Percentage of non-susceptiblity to oxacillin (methicillin) in *Staphylococcus aureus* isolates reported by participating ARMed centres



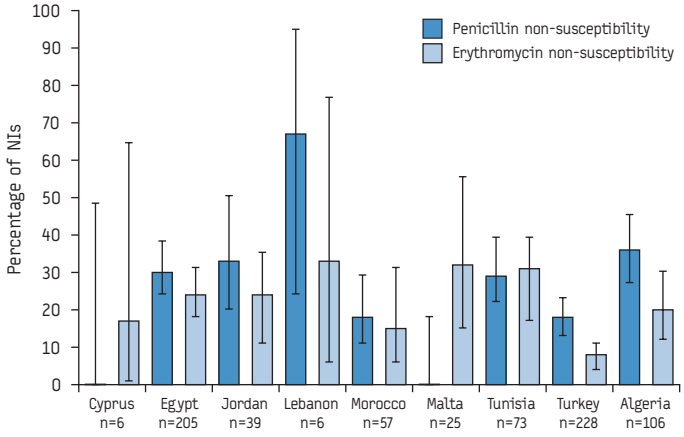
I: 95% confidence interval

Note: ARMed centres are grouped by country and reported number of isolates

Even greater disparity was seen in the resistance patterns for *E. coli*, mainly for fluoroquinolones and third generation cephalosporins [FIGURE 3]. Resistance to third generation cephalosporins varied from 3% in Malta to 72% in Egypt (median 18% [16 – 26%]) and in the fluoroquinolones ranged from 5% in Algeria to 40% in Turkey (median 27% [18 – 33%]). Multiresistant isolates were also particularly evident. In fact, simultaneous resistance to four major antimicrobial classes (aminoglycosides, third generation cephalosporins, aminopenicillins, fluoroquinolones) was the second most common resistance pattern seen and accounted for 13.7% of all isolates reported. Multiresistant *E. coli* isolates were most commonly found in Egypt, comprising more

FIGURE 1

Percentage of non-susceptibility to penicillin and erythromycin in *Streptococcus pneumoniae* isolates reported by participating ARMed centres

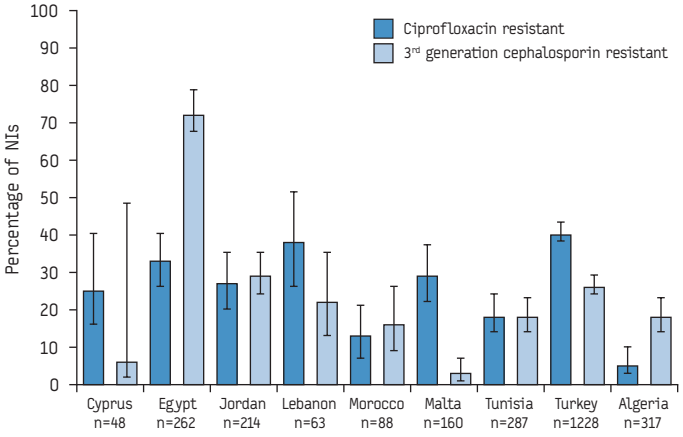


I: 95% confidence interval

Note: ARMed centres are grouped by country and reported number of isolates

FIGURE 3

Percentage of non-susceptibility to fluoroquinolones and third generation cephalosporins in *Escherichia coli* isolates reported by participating ARMed centres



I: 95% confidence interval

Note: ARMed centres are grouped by country and reported number of isolates

than half of all resistant isolates from that country, followed by Turkey and Lebanon where around 30% were similarly multiply resistant to three or more antibiotic groups.

Out of the five countries (Cyprus, Malta, Morocco, Tunisia and Turkey) that provided data on more than 10 isolates of each of the enterococcal species under investigation, only two reported vancomycin non-susceptibility (intermediate or resistant): Turkey, with 0.9% (95% CI 0%–2%) *E. faecalis* and 3.8% (2 – 7 %) *E. faecium*; and Cyprus, with 2% (0%–14%) *E. faecalis*.

Discussion

ARMed results have provided, for the first time, a standardised, comparable snapshot on the prevalence of resistance in important clinically relevant pathogens within hospitals in the southeastern Mediterranean. A median of 43% for methicillin resistance within isolates of *S. aureus* confirms that the Mediterranean region indeed constitutes a high prevalence region for MRSA. It also correlates well with previous sporadic reports from individual countries within the region [8-10]. It appears that MRSA seems to be more widespread in the eastern Mediterranean than in the south. Overall data from the hospitals in Cyprus, Egypt, Jordan and Turkey all showed MRSA proportions in excess of 40%. Results from the EARSS network, using the same methodology, have reported MRSA proportions in excess of 30% from Croatia, France, Greece, Israel and Portugal [11,12]. This would therefore indicate that the whole of the Mediterranean region is a high prevalence region for MRSA.

On the other hand, resistance within *E. coli* to third generation cephalosporins, and by association possibly extended-spectrum beta-lactamase (ESBL) and/or Amp-C enzyme production, appears to be more significant to those previously reported in the region. This seems especially the case in the eastern Mediterranean where the centres in Turkey, Lebanon, Jordan and Egypt reported average proportions in excess of 20%. The figure of 72% resistance to third generation cephalosporins reported from Egypt is one of the highest figures recorded for this resistance trait. Nevertheless indications of high level resistance within Gram negative pathogens in this region are not new. Bouchillon and colleagues, studying isolates from 38 centres in 17 countries, reported the incidence of ESBL production in Enterobacteriacae to be at its highest in their Egyptian centres at 38.5% [13]. El Kholy et al noted that 62% of *E. coli* isolated from blood cultures in three Cairo hospitals were non-susceptible to ceftazidime [14]. The ARMed results seem to confirm these previous reports and indicate that the situation may be even more acute and widespread throughout the Mediterranean region than previously indicated. In addition the presence of co-resistance to other antibiotic groups further compounds the challenge posed by such pathogens.

In contrast, the proportions of penicillin non-susceptibility in *Streptococcus pneumoniae* (PNSP) isolates from the ARMed laboratories are broadly in line with those already reported in centres within other Mediterranean and Middle Eastern countries [15,16]. PNSP levels seem to be uniform throughout the region and the apparent differences seen between eastern and southern Mediterranean centres in *S. aureus* and *E. coli* were not found in pneumococci. It is also interesting to note that, contrary to reports from the European countries of the region where macrolide resistance in pneumococci often exceeds that of penicillin [15, 16], erythromycin non-susceptibility appears to be less prevalent in the southeastern Mediterranean countries. Finally, the presence of vancomycin resistance in enterococci from ARMed laboratories in Turkey supports previous reports from this country [17].

The use of routinely collected clinical laboratory data provides the advantage that epidemiological conclusions mirror the day-to-day situation in the participating institutions. Furthermore, the choice of blood culture isolates minimises sample bias and reflects the clinical situation in the more severe infections. Unlike patients with less critical infections, such as those of the respiratory and urinary tract, most patients with signs of sepsis or meningitis will probably have a microbiological sample taken. In addition, the major strength of the ARMed study resides in the common methodology used throughout the different centres in the nine participating countries. Any such study has the limitation of depending on the accuracy and validity of the individual participating laboratories, which can vary, especially in limited resource countries. It is also hampered by the inability to perform third party reference laboratory verification and do more detailed investigation in results of particular significance. Nevertheless, the concurrent satisfactory quality assurance results would suggest that this potential limitation did not prejudice the conclusions reached from the preliminary data collected by ARMed-EARSS to any significant degree [18].

In addition to the direct repercussions for the countries of the Mediterranean region [19], the epidemiology of resistance in the southern and eastern Mediterranean also has European implications. Human mobility in this region is highly significant, in terms of both recognised travel (particularly tourism) and the results of migration. The importation of multiresistant organisms to European hospitals via patients arriving from countries within the Mediterranean region is well documented [20,21]. Such an occurrence may result in the possibility of subsequent intra-institutional spread with the potential for an outbreak [22,23]. Prior knowledge of the epidemiology of resistance in the Mediterranean region will therefore facilitate the introduction of effective interventions on initial contact with patients originating from this region and who may be potentially colonised or infected with multiresistant organisms. It should also prove beneficial for stakeholders in the countries involved to plan and implement correct antibiotic stewardship programmes to attempt control and possibly reduction of the incidence of antimicrobial resistance in pathogens of critical importance [24].

Conclusion

The preliminary results from the ARMed project have started to shed new light on the incidence of antimicrobial resistance in the south and east of the Mediterranean. They appear to support and accentuate previous sporadic reports suggesting a high prevalence in this region and indicate that this is particularly the case in the eastern region where multiresistance in *S. aureus* and *E. coli* seem to be especially high, and higher than that reported in the other countries of the Mediterranean. This picture will become clearer once the full duration of the study is completed and a more comprehensive isolate database is established.

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ORIGINAL ARTICLES

Surveillance report

HEALTHCARE ASSOCIATED INFECTIONS IN UNIVERSITY HOSPITALS IN LATVIA, LITHUANIA AND SWEDEN: A SIMPLE PROTOCOL FOR QUALITY ASSESSMENT

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Surveillance of healthcare associated infections is an overlooked parameter of good clinical practice in most healthcare institutions, due to the workload demanded in the absence of adequate IT-systems. The aim of the present study was to investigate whether a simple protocol could be used to estimate the burden of healthcare associated infections in three university hospitals in Huddinge in Sweden, Riga in Latvia and Vilnius in Lithuania and form the basis for initiating a long term follow up system.

The medical records of all patients receiving antibiotics were reviewed according to a standardised protocol, focusing on the indications for the drugs and on the frequency of hospital acquired infection (HAI) in a point-prevalence survey. Only comparable specialities were included.

The proportion of patients treated with antibiotics (prophylaxis not included) were 63/280 (22%) in Huddinge, 73/649 (11%) in Riga and 99/682 (15%) in Vilnius. The proportion of admitted patients treated for a HAI were 15%, 3% and 4%, respectively, (both comparisons Huddinge versus other centres P<0.001). Surgical site infections were most common, followed by infections with an onset more than 2 days after admission without any of the other registered risk factors present. Our inexpensive and simple method showed that healthcare associated infections were a significant problem among patients admitted to Huddinge. The figures obtained can be used for further discussion and form a baseline for follow up at the local level. The comparison of figures between centres was far less relevant than the process the study created.

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Key words: Healthcare associated infection, hospital acquired infection, antibiotics, quality assessment

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Introduction

Despite its relevance, the surveillance of healthcare associated infections is overlooked as a parameter of good clinical practice in most healthcare institutions. Apart from purely scientific projects, most of which have time limits, most registration initiatives are hampered by the workload for data collection, administration, feedback and long term sustainability. When long term registration is started, it often relies on a few devoted enthusiasts, rather than a broad acceptance among clinicians. Some of the major obstacles are that the purpose and ambition is ill-defined, protocols and criteria are too extensive and, when computer-based medical records exist, integration with microbiological results is poor.

In order to estimate the burden of healthcare associated infections in Karolinska University Hospital, Huddinge, Sweden (Huddinge), and to initiate a system for follow up over the years, we recently made a survey using a rather simple protocol based on patients receiving antibiotic treatment [1]. The specific aims of the present study were to see whether our protocol was useful for the assessment of the