## PEER REVIEW HISTORY



## ARTICLE DETAILS

| | |
|---|---|
| **TITLE (PROVISIONAL)** | **Decline of meticillin resistant S. aureus in Oxfordshire hospitals is strain specific and preceded infection control intensification** |
| **AUTHORS** | Wyllie, David; Walker, Ann; Miller, Ruth; Moore, Catrin; Williamson, Susan; Schlackow, Iryna; Finney, John; O'Connor, Ellen; Peto, Tim; Crook, Derrick |

## VERSION 1 - REVIEW

| | |
|---|---|
| **REVIEWER** | ***G Gopal Rao***<br>Consultant Microbiologist<br>North West London Hospitals NHS Trust<br>Northwick Park Hospital<br>Harrow<br>Middlesex<br>UK |
| **REVIEW RETURNED** | 18-May-2011 |

| | |
|---|---|
| **THE STUDY** | "Rates were also calculated according to whether the MRSA strain was resistant or sensitive to<br>erythromycin. We estimated rates of ST22 and ST36 MRSA assuming that (see Results) (i) all MRSA<br>strains were represented by ST22 and ST36 only (ii) all erythromycin-sensitive MRSA strains were<br>ST22, (iii) ST22 consisted of similar proportions of erythromycin-sensitive and -resistant MRSA<br>strains. Sensitivity analyses replaced (iii) with the upper and lower 95% confidence limits for the<br>estimated proportions of Erythromycin-sensitive and -resistant ST22". (Lines24-30)<br>Comment: I have serious doubts about this assumption. A majority of the strains of both ST 22 (EMRSA 15) and ST 36 (EMRSA 16) are resistant to erythromycin. Erythromycin resistance MRSA strains is mainly plasmid mediated and is not a stable phenotypic characteristic. Thus to make an assumption using thisunstable characteristic may lead to incorrect conclusion about the relative decline of the strains. I refer the authors to the table comparing EMRSA 15 and EMRSA 16 in CDR weekly Volume 5;Number 35 dated 1 September 1995 (ISSN 1350-9357) |
| **RESULTS & CONCLUSIONS** | As stated in the section above, I have serious difficulty in agreeing to the assumptions made about erythromycin resistance and its association with EMRSA strains including the proportions. If the authors had limited their conclusions only to state there was decline of MRSA preceding the the 2006 interventions, that would be supported by their data. But they cannot conclude that a specific strain (especially ST 36) has declined based on erythromycin sensitivity data. (lines 39-57)<br>Compatible with the pattern observed in blood, declines were essentially restricted to |

| | |
|---|---|
| | ST36 in isolations from invasive samples and surface swabs (data not shown). The authors state:<br>"Compatible with the pattern observed in blood, declines were essentially restricted to<br>ST36 in isolations from invasive samples and surface swabs (data not shown)." In fact this data will be crucial in supporting their assumptions, if indeed the strains have been characterised as ST 36 and there is significant correlation between erythromycin resistance/sensitivity and ST types. I have checked with the Antibiotic Reference Laboratory, Health Protection Agency, England and I am given to believe that the majority of EMRSA 15 strains continue to be erythromycin resistant. The authors quote the study by Ellington et al as a support for their use of differing proportion of erythromycin resistance as a marker for diffrentiating strains. In fact, Ellington et al paper states "As noted previously, the majority of both EMRSA-15 and EMRSA-16 isolates were resistant to macrolides (74% and 85%, respectively)". However it is concievable that Oxfordshire hospitals may be seeing a different clone of EMRSA 15 strain. While I do agree that EMRSA 16 has declined, I do not think the methods used in this study support the key message " The onset of the decline preceded infection control interventions, and study of antibiotic resistant patterns and typing data suggests that the two dominant MRSA clones followed different trajectories." Lines 24-27 But for this major limitation, I think the paper is well written and will be of considerable interest to the readers. If the authors can retrieve any representative archived strains for the study period, they should type the strains and establish the correlation between erythromycin resistance and EMRSA strain types. |

| | |
|---|---|
| **REVIEWER** | ***Dr Alan Johnson***<br>Consultant Clinical Scientist<br>HPA Centre for Infections<br>London, UK<br><br>No competing interests |
| **REVIEW RETURNED** | 23-May-2011 |

| | |
|---|---|
| **GENERAL COMMENTS** | While the decrease in EMRSA-16 (ST36) in the UK has already been documented among blood cultures isolates of MRSA (reference 8), this paper gives important further insight into the epidemiology of MRSA infection. It demonstrates that the change in clonal structure of MRSA has occurred not only in isolates from blood (which as mentioned above is already known) but also in invasive isolates and in isolates from wounds. This is an important observation as only a minority of clinical isolates of MRSA are from blood. The data presented here also give a more robust picture of the chronology of the change in clonality and, critically, indicate that the change (which is associated with the overall decrease in the incidence of MRSA) occurred before interventions aimed at reducing MRSA bacteraemia in response to government targets were implemented. While many interventions to reduce MRSA that have been implemented in hospitals are intuitively sensible, this paper highlights that the associated evidence base is weaker than is perhaps commonly appreciated. Given the importance of healthcare-associated infections, this paper gives support to the idea that research into the effectiveness of infection control measures should be a priority for research. |

| | |
|---|---|
| | Minor point: In line 21 it is incorrectly stated that EMRSA-15 and -16 were responsible for 95% of MRSA cases from the late 1980s - it was the mid 1990s. |

| | |
|---|---|
| **REVIEWER** | ***Professor Mark H. Wilcox***<br>Consultant / Clinical Director of Microbiology/Pathology<br>(Leeds Teaching Hospitals NHS Trust)<br>Professor of Medical Microbiology<br>(University of Leeds)<br>Health Protection Agency Lead on Clostridium difficile<br><br>Microbiology,<br>Old Medical School,<br>Leeds General Infirmary,<br>Leeds LS1 3EX,<br>W. Yorks., UK.<br><br>I have no competing interests. |
| **REVIEW RETURNED** | 30-May-2011 |

| | |
|---|---|
| **GENERAL COMMENTS** | This is an excellent, clearly written manuscript.<br><br>I have no major comments to make.<br><br>The authors have understandably concentrated on the changes in MRSA epidemiology and possible strain dependent explanations for the observed rate fluctuations. Study of the strains accounting for the considerable burden of MSSA infection is appropriate, not least given the greater preponderance of infection due to these S. aureus isolates. |

## VERSION 1 – AUTHOR RESPONSE

A copy of this response (with formatting and colour) is uploaded as a file.

We thank the reviewers for their careful scrutiny our work. Professor Wilcox advised no changes; we have made the alteration suggested by Dr Johnson. Dr Rao requests that:

If the authors can retrieve any representative archived strains for the study period, they should type the strains and establish the correlation between erythromycin resistance and EMRSA strain types.

We confirm have done exactly this, as was stated in Results:
.. we estimated rates of ST22 and ST36 MRSA by comparing multi-locus sequence types of two archival collections of nosocomial bacteraemia Oxford MRSA strains(17, 20) with erythromycin sensitivities from 1999 and 2007.
Although (as Dr Rao points out) the relationship between strain and macrolide resistance need not be constant over time (cf. Harris SR et al Science. 2010 Jan 22;327(5964):469-74), our data suggest this is likely to be the case, which is compatible with Harris' observation that development of resistance within a clonal complex is a relatively rare event. This comment is already made in the discussion.

The precise mechanism(s) of resistance (e.g. phage vs. Chromosomal) are not relevant to the inference drawn in this paper, which depends only on the relationship between laboratory sensitivity results and MLST type over time. We agree the resistance rates observed for the strains in this study differ somewhat from those reported elsewhere, although the ranking is similar (E-MRSA16/ST36

being more resistant). Laboratory protocols used by the two studies are not completely identical, and since inducible macrolide resistance in S. aureus is well described, precise laboratory testing protocols may well influence in vitro results. Alternatively, as Dr Rao notes, our strains may differ in some way from those prevalent elsewhere. These are interesting possibilities, and we are investigating them; the strains described have been subject to complete genomic sequencing, and results of this will be presented elsewhere.

Dr Rao comments on the section
Compatible with the pattern observed in blood, declines were essentially restricted to ST36 in isolations from invasive samples and surface swabs (data not shown)
We do not have archival collections of non-blood isolates. Although we think it is very unlikely different strains are responsible for blood and non-blood isolates, we cannot prove this is the case. Consequently, we have qualified the above statement, modifying it to read

Compatible with the pattern observed in blood, declines appeared largely restricted to ST36 (based on the relationships derived from analysis of blood culture collections) in isolations from invasive samples and surface swabs (Figure 3).

Dr Rao felt that the pattern in tissues other than blood was important; we agree. In view of this, we have included a figure illustrating the data as Figure 3, with legend.

**VERSION 2 - REVIEW**

| | |
|---|---|
| **REVIEWER** | ***Guduru Gopal Rao*** |
| **REVIEW RETURNED** | 08-Jun-2011 |

| | |
|---|---|
| **GENERAL COMMENTS** | Reviewer completed checklist only. No further comments were made. |