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What do Molecular Epidemiological Studies on CAMRSA tell us?

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Community MRSA (CA-MRSA) have arisen relatively recently in comparison with hospital associated MRSA (HA-MRSA). There are many apparent differences between the evolution and spread of these two entities. HA-MRSA appears to have arisen from a limited number of MSSA clones and then disseminated widely such that there are only a small number of successful clones worldwide. Clones such as ST5 MRSA II, ST239 MRSA III, and ST22 MRSA IV are commonly found in the Asia Pacific region.

CA-MRSA was first described in the remote indigenous population of Western Australia and subsequently reported in many regions of the world. Unlike HA-MRSA, CA-MRSA appears to have evolved locally from MSSA strains with definite clones initially limited to specific geographical regions. There has been subsequent spread of some of the more successful clones such as ST8-MRSA- IV (USA300), ST80-MRSA-IV, ST30-MRSA-IV, ST59-MRSA-IV/V and more recently ST772-MRSAV.

The use of epidemiological typing of CAMRSA can provide important information on evolutionary events - questions such as

- Have CA MRSA arisen by introduction of SCC*mec* into successful lineages of MSSA with dissemination? or
- Has the SCC*mec* been introduced into multiple less common MSSA lineages with subsequent adaption and increase in fitness resulting in endemicity?
- Have internationally recognised clones been introduced into a region?
- If introduced have evolutionary changes occurred?

Western Australia (WA) is well placed to answer these questions. MRSA has been a notifiable organism in WA since 1984 with all isolates sent to a central laboratory. The Australian

Collaborating Centre for *Enterococcus* and *Staphylococcus* Species (*ACCESS*) Typing and Research performs a wide variety of typing techniques (PFGE, MLST, DNA microarray, *spa* typing, and direct repeat unit (*dru*) typing as well as PVL detection and characterisation.)

A study (O'Brien et al 2009) on 11 remote indigenous populations of Western Australia between 1995 and 2003 revealed that there were numerous MSSA strains representing 13 clonal complexes, 23 singletons and 56 sporadic isolates. Five MSSA lineages also contained MRSA strains but surprisingly these were not the predominant MSSA strains in the communities. For example the predominant MSSA was ST93 but there were no ST93 MRSA detected. CA-MRSA were found in different CCs indicating that horizontal transmission of the SCC*mec* element into *S. aureus* has occurred on at least six occasions. Based on the *spa* type and DNA microarray profile six evolutionary events have subsequently occurred on at least three occasions from these clones (i.e. vertical transmission of the SCC*mec*).

A subsequent study (Coombs et al 2011) on MRSA isolated across WA showed more extensive horizontal transfer of the SCC*mec* into different CCs and within the same CC. In addition for many single and double locus variant CA-MRSA clones only a few isolates were detected. This suggests the successful evolution of a CA-MRSA clone may not only depend on the mobility of the SCC*mec* element but also on other genetic determinants. The selective pressure for the emergence and maintenance of multiple MRSA clones likely comes from high beta-lactam and macrolide usage for skin disease and sexually transmitted disease in the lower socioeconomic groups of the population although this requires further study. This study also demonstrated the importation of several PVL positive clones, including the CC59 strain ST59-MRSA-V_T [5C2&5] (Taiwan CA-MRSA clone), and the CC8 strain ST8-MRSA-IV [2B] (USA300). Genetic analysis of these strains indicated they are distinct from WA CA-MRSA clones.

Two of these apparently imported clones were analysed further. (Coombs et al 2010, 2009) Although ST59-MRSA-V_T [5C2&5] (Taiwan CA-MRSA clone) was found to be the most prevalent CC59 clone isolated in WA, independent evolution of PVL-negative CC59 CA-MRSA has occurred. Using a variety of molecular techniques, six distinct groups of CC59 were differentiated. Within these groups at least seven different variants of SCC*mec* elements were distinguished. This suggests rapid evolution and/or multiple transfer events of SCC*mec* have

occurred within this CC. Although some CC59 isolates in WA have overseas origins (eg Taiwan CA-MRSA clone and possibly USA1000), PVL-negative CC59 lineages unique to WA have acquired various SCC*mec* types on multiple occasions. The PVL-positive ST8-MRSA-IV [2B] strain isolated in WA was found to be closely related to USA300, with most isolates unable to be distinguished from USA300-TC1516. Some isolates however varied in their carriage of resistance and virulence determinants and therefore USA300 in Australia cannot be regarded as being genetically homogeneous. Altogether 16 variants were identified indicating continuing evolution of this strain. Notably some isolates did not harbour the ACME locus, which is intriguing because this locus is assumed to be involved in facilitating the spread of USA300 by skin contact.

ST93-MRSA-IV is the predominant CA-MRSA in Australia but has only recently appeared in WA where it has rapidly become the dominant PVL positive clone. Molecular typing of 58 isolates over 16 years from around Australia (Coombs et al 2012) has demonstrated that the emergence of Queensland CA-MRSA (ST93-MRSA-IVa [2B]) has been due to independent acquisitions of different *dru*-defined type IV and type V SCC*mec* elements in several *spa*-defined ST93-MSSA backgrounds. Rearrangement of the *spa* sequence in ST93-MRSA has subsequently occurred in some of these strains. Although multiple ST93-MRSA strains were identified, PVL-positive ST93-MRSA-IVa [2B]-t202-dt10 was the predominant strain. Whether this strain arose from one PVL-positive ST93-MSSA-t202 or by independent acquisition of SCC*mec*-IVa [2B]-dt10 into multiple PVL-positive ST93-MSSA-t202 strains is yet to be determined.

These studies (PhD thesis by Dr Geoffrey Coombs) have demonstrated many MRSA have most likely evolved due to multiple acquisitions of SCC*mec* elements into MSSA with subsequent expansion of adapted genetically fit MRSA clones rather than insertion of the SCC*mec* into successful MSSA clones. This is not so for the successful local CA-MRSA, ST93 MRSA IV, which has arisen due to independent acquisition of the SCC*mec* into the highly successful ST93-MSSA with subsequent further evolution. Similarly further evolutionary changes have been demonstrated in imported international clones.

References

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