ISOLATION OF METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* (MRSA) AND *S. EPIDERMIDIS* (MRSE) FROM CHILDREN OF WELFARE HOMES IN IPOH, PERAK, MALAYSIA.

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• *Staphylococcus* is a gram positive that appear as a spherical shape and form a grape-like clusters. Reside on the skin and mucous membrane.

• *S. aureus* is an ubiquitous bacteria that has ability to develop antibiotic resistance (Jiang *et. al.*, 2019).

• *S. epidermidis* related with catheters and indwelling medical devices (Arciola *et. 2001*).

• Opportunistic pathogens and can cause serious infections:
  - Skin and soft tissue infections (SSTI)
  - MRSA

(Pantosti and Venditti, 2009)
Most of MRSA infections are mild, but they can change becoming deeper and more serious.

Bug bites, rashes, and other skin problems can be confused with MRSA.

Overcrowded facilities, close contact and lack of proper sanitation had contributed to the spread of MRSA (Wijaya and Liang, 2006).

Figure 1: Wound caused by spider bite

Figure 2: Wound infection caused by MRSA
1. To determine any potential MRSA and MRSE from children of welfare homes.

2. To detect susceptibility of isolates against Cefoxitin.

3. To detect mecA gene in Cefoxitin-positive isolates.
METHODS

SAMPLES COLLECTION

BACTERIAL ISOLATIONS

POLYMERASE CHAIN REACTION (PCR)

PHENOTYPIC IDENTIFICATION

ANTIBIOTIC SUSCEPTIBILITY TEST (AST)
1) SAMPLES COLLECTION
   - Nasal swab (consent obtained prior from guardians of children welfare homes)
   - Sociodemographic details collected

2) BACTERIAL ISOLATIONS
   - Mannitol Salt Agar (MSA)
   - Clumping factor & Protein A

Figure 3: BactiStaph Latex Agglutination Test Kit. No. 4: Isolates, No.5: Positive control and No. 6: Negative control
3) ANTIBIOTICS SUSCEPTIBILITY TEST (AST)

- Cefoxitin (30µg) disc diffusion
- Automatic colony counter
- Resistant ≤ 21cm

Figure 3: Antibiotic disc diffusion against Cefoxitin
4) PHENOTYPIC IDENTIFICATION

- Isolates that were Cefoxitin-resistant were confirmed for present of Penicillin Binding Protein 2A (PBP2 kit-Oxoid, USA).

![Figure 4: Detection of PBP2a as positive control - TL MRSA (Test Latex Methicillin-resistant Staphylococcus aureus - no agglutination) and CL MRSA (Control Latex Methicillin-resistant Staphylococcus aureus - agglutination)](image)
5) POLYMERASE CHAIN REACTION (PCR)

- *meca* gene (gold standard)

- Primer sequence
  Forward: 5’-AAAATCGATGGTGTAAGGTTGGC-3’
  Reverse: 5’-AGTTCTGCTGATACCGGATTTTG-3’

- Thermal cycling conducted was 5 minutes at 94°C; 30 seconds at 58°C and 30 seconds at 30°C for 30 amplification cycles (Siripornmongcolcháí *et al.*., 2002).

- Product separated on 1% agarose gel and view under UV Transilluminator.
• Of the 226 samples, 197 and 94 isolates of *S. aureus* and *S. epidermidis*.

• One out of 197 (0.6%) *S. aureus* isolates and 3 out of 94 (3.2%) *S. epidermidis* were positive for Cefoxitin-resistant.

• The *mecA* gene was detected in 1 (0.5%) out of 197 isolates *S. aureus* and 3 (3.2%) out of 94 isolates *S. epidermidis*. 
Figure 5: Analysis of PCR products with primers specific generating 286 bp fragment by conventional polymerase chain reaction using 1% agarose gel electrophoresis. Lane 1: 100 bp DNA Ladder, Lane 3: S. aureus, Lane 2,4,5: S. epidermidis, Lane 6: MRSA and MRSE (positive control), Lane 8: MSSA (negative control)
• Finding of *mecA* gene is the major evidence (gold standard) for the detection of Methicillin-resistant *Staphylococcus aureus* isolate (Olayinka *et. al.*, 2009).

• Previous study among university student in Malaysia showed 11.5% were positive for CA-MRSA (Mariana *et. al.*, 2008) among students in Medical School.

• Recent study on MRSA among healthy individual in Nigeria according to Bale *et. al.*, (2019) revealed 14% isolates; Japan 6.3% isolates (Hirose *et. al.*, 2019).
CONCLUSIONS

Low prevalence

Asymptomatic

Might contribute to CA-MRSA/CA-MRSE

Consider for further study: Type and other virulent/toxin genes
REFERENCES


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