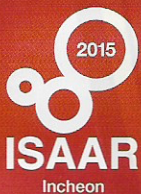


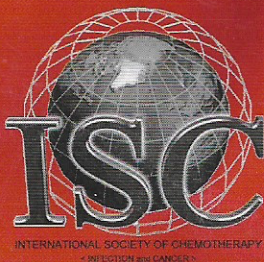
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strains were corrected from 93 hospitals nationwide surveillance program for VISA/VRSA using brain heart infusion agar with 4 mg/L oxacillin testing systems, VISA strains were identified by MIC values of agar, broth dilution or disk diffusion guidelines (M100-S24, 2014). Molecular typing was performed using toxin genes detection, pulsed-field gel electrophoresis (PFGE). Screened isolates were identified as VISA strains with MIC of 3 mg/L or more. The isolates were screened for enterotoxin genes of *sec*, *seg*, and *sei* with *sec*, *seg*, *sei* with SCCmec III and *agr* I or with SCCmec IV and *agr* I (1 isolates). In addition, TSST-1 gene was detected

of MRSA strains were isolated in Korea, but no VISA strains (81.8%) showed SCCmec II

Staphylococcal binding protein in ST5 methicillin-resistant

Yong, J.L. Yoo, C. Park, I.H. Kim. Division of Microbiology, National Institute of Health, Korea

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a major cause of infections, and the ST5 type is dominant in Korea. Like many other *S.aureus* strains, MRSA produce a series of virulence factors. The functions and mechanisms of the genes are not clear. We investigated the contribution of elastin-binding protein to virulence using knockout system in

MRSA. *Ebp* shuttle vector (pKOR) was used for *ebp* gene construction. The *ebp* knockout strains S3659 and S3670 were constructed by homologous recombination methods; S3659 Δ *ebp* and S3670 Δ *ebp*. The virulence expression was analyzed by real time pcr, adherence, cytotoxicity, binding to extracellular matrix and antimicrobial susceptibility of mutants were compared to parent strains.

Expressions of *lgrA*, *icaA*, *sspA*, and *efb* were analyzed in the *ebp* knockout strains. On the other hand, the parent strains did not show any difference in the adhesion and invasion activity, and showed lower cytotoxicity compared to the parent strains. Deletion of *ebp* in both knockout strains caused a decrease of ECM binding capacity including the binding to FN, pFN, and type IV collagen. The MIC of imipenem indicated that S3670 Δ *ebp* was susceptible.

Deletion of *ebp* gene in ST5 MRSA may affect the expression of some virulence genes and ECM binding. This study showed the cytotoxicity and the antimicrobial susceptibility of the S3670 Δ *ebp* strain.

Molecular characterization of methicillin-resistant *Staphylococcus aureus* from pigs and farm workers

Yong, S.-R. Kim, G.-C. Jang, H.-S. Lee, Division of Microbiology, Animal and Plant Quarantine

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a major cause of nosocomial infections and resistant bacteria. Furthermore incidence

of MRSA in animals has increased in Korea recently. Therefore, the aims of this study were to investigate the prevalence and to characterize the MRSA isolates from pigs and farm workers in Korea.

Methods: Between February 2012 and May 2013, a total of 671 nasal cavity swab samples were collected from pigs and farm workers of 31 pig farms throughout Korea. The types of *mec* gene complex, toxin genes and host specific genes were determined by PCR. In addition, multilocus sequence typing and *spa*-typing were performed to determine the genetic relatedness of the MRSA strains.

Results: The prevalence of MRSA was 5.3% (31/580) in pigs and 17.6% (16/91) in farm workers, respectively. Highest prevalence (9.9%, 14/142) of MRSA was observed in weaned piglets among the age groups. Two different lineages were found among the 47 MRSA isolates from pigs and workers: 31 and 11 strains of livestock-associated type (ST398 or ST541/ *spa* t034 or t034 variant) and 0 and 5 strains of human-associated type (ST72/ *spa* t5440 or t664 or t148), respectively. All HA MRSA isolates carried enterotoxin, leukotoxin genes and/or host specific genes, whereas LA MRSA not. However, all LA MRSA isolates were multi-drug resistant, whereas HA types were susceptible or resistant to less than two antimicrobials. Furthermore, uncommon antimicrobial resistance genes such as *fexA*, *cfi*, and *tetL* were detected in LA MRSA isolates. Biofilms were formed by 42.9% (18/42) of LA MRSA isolates, whilst HA MRSA isolates not.

Conclusion: To our knowledge, this is the first report of LA MRSA in farm workers in Korea. This result suggests that MRSA in pigs may pose health risk especially for people who frequently come into contact with pigs.

P1-SP11

The different effect of concentration and contact time of chlorhexidine and triclosan on the growth of methicillin resistant *Staphylococcus aureus* (MRSA)

E. Suswati. Laboratory of Microbiology, Medical Faculty, University of Jember, Indonesia

Staphylococcal infection spread specially in hospital as nosocomial infection could be controlled by the use of proper antiseptic to medical staff. This study investigated inhibition effect of chlorhexidine and triclosan to Methicillin Resistant *Staphylococcus aureus* (MRSA) growth using various concentration and contact time. Each antiseptic and its concentration have been tested to 4 MRSA isolate i.e. MRSA I, II, III, IV for various contact time i.e. 30", 60", 90" and 120". Concentration of chlorhexidine were 0.25%, 0.5%, 1%, 2% and 4%. Concentration of triclosan were 0.125%, 0.25%, 0.5%, 1% and 2%. All treatments were replicated 4 times. Inhibition to each antiseptic compared to control group.

Statistical analyses mean of multivariate ANOVA continued to LSD showed significant different in all treatment ($p < 0.01$). There was significantly different between concentrations. Significance different in contact time only happened between 30", 60" and 90". Result showed different inhibition effect of MRA growth by chlorhexidine compared to triclosan in various concentration and contact time. Antiseptic concentration and contact time were major influent to inhibit the growth of MRSA.

The conclusion of this investigation showed that 0.5% concentration of chlorhexidine, 0.25% concentration of triclosan and 90" contact time were the most effective concentration and contact time for inhibition Methicillin Resistant *Staphylococcus aureus* growth.

Keywords: chlorhexidine, triclosan, concentration, contact time, MRSA

ABSTRACT

Staphylococcal infection spread specially in hospital as nosocomial infection could be controlled by the use of proper antiseptic to medical staff. This study investigated inhibition effect of chlorhexidine and triclosan to *Methicillin Resistant Staphylococcus aureus* (MRSA) growth using various concentration and contact time. Each antiseptic and its concentration have been tested to 4 MRSA isolate i.e. MRSA I, II, III, IV for various contact time i.e. 30", 60", 90" and 120". Concentration of chlorhexidine were 0.25%, 0.5%, 1%, 2% and 4%. Concentration of triclosan were 0.125%, 0.25%, 0.5%, 1% and 2%. All treatments were replicated 4 times. Inhibition to each antiseptic compared to control group.

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Key Word: chlorhexidine, triclosan, concentration, contact time, MRSA

INTRODUCTION

Staphylococcal infection spread in the hospital especially as a nosocomial infection could be controlled by the using of proper antiseptic by the medical staff. Methicillin-resistant *Staphylococcus aureus* (MRSA) continues to be a global problem in infection control. For many years it has been a major cause for nosocomial infections in many countries. The proportion of methicillin resistance among clinical isolates of *S. aureus* is still increasing. In southern European countries, the proportion may be as high as 55%. MRSA now even becomes an increasing problem in the community. Transmission of MRSA in community has been shown to be as high as 60%. Family members who are living with MRSA carriers are in danger of MRSA transmission. Dermal colonization with MRSA may be persistent, especially in the groin. That is why attempts are often undertaken to treat colonized MRSA patients. Antibiotics were shown to be effective in uncontrolled and controlled trials with eradication rates between 53% and 85%. But antibiotics are considered to be inappropriate for patients who are only colonized and not infected with MRSA. One reason is their potential to cause adverse effects, especially allergy, which can not be justified for patients who do not have an infection. More important is the risk of emergence of vancomycin-resistance in *S. aureus*. Topical antiseptic measures, however, are normally employee. The nasal cavity is usually treated with mupirocin or with tolerable antiseptic. Dermal colonization is eradicated with antiseptic liquid soap. Only few studies have addressed the question of MRSA eradication among colonized patients with liquid soaps in combination with nasal treatment. All of them are uncontrolled trials and most of them have different types of biases (Kampf & Kramer, 2004).

MATERIAL AND METHODE

Triclosan 0.125%, 0.25%, 0.5%, 1%, 2% and Chlorhexidine 0.25%, 0.5%, 1%, 2%, 4% had been tested to MRSA isolate in various contacttime i.e. 30", 60", 90" and 120". Control groups were MRSA contacted each to 0% chlorhexidine. All treatment replicated 4 times. Chlorhexidine inhibition effect compared to control group.

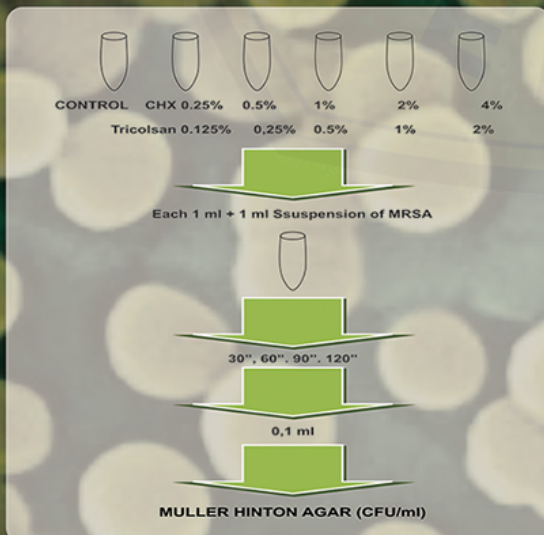


Table 1. Mean of MRSA growth after contact with chlorhexidine

Contact Time	Mean of MRSA colony count					
	Concentration of chlorhexidine					
	0%	0.25%	0.50%	1%	2%	4%
30	100	20.31	5.4	0	0	0
60	100	4.24	1.3	0	0	0
90	100	1.05	0	0	0	0
120	100	0.23	0	0	0	0

Table 2. Mean of MRSA growth after contact with triclosan

Contact Time	Mean of MRSA colony count					
	Concentration of triclosan					
	0%	0.125%	0.25%	0.5%	1%	2%
30	100	54,91	15,80	0	0	0
60	100	18,81	6,36	0	0	0
90	100	0,00	0,00	0	0	0
120	100	0,91	0,00	0	0	0

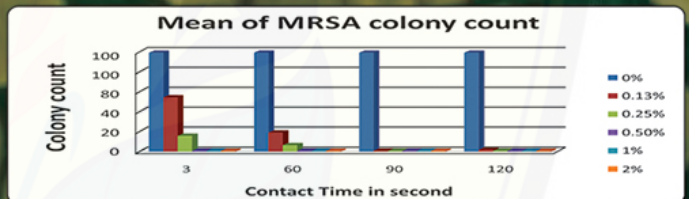


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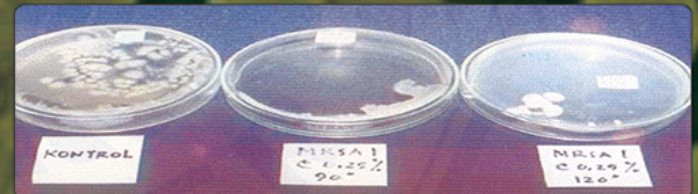


Fig 2. MRSA growth after contact with chlorhexidine 0.25% , contact time 90" and 120"

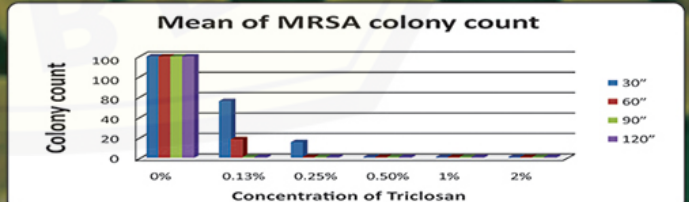


Fig 3. Mean of MRSA growth after contact with Triclosan



Fig 4. Mean of MRSA growth after contact with Triclosan

RESULT AND DISCUSSION

The result showed there were different inhibition effects by chlorhexidine and triclosan to MRSA growth in the control groups of all concentrations. Significance different in contact time only happened between 30", 60" and 90".

CONCLUSION

The conclusion of this investigation showed that 0.5% concentration of chlorhexidine, 0.25% concentration of triclosan and 90" contact time were the most effective concentration and contact time for inhibition *Methicillin Resistant Staphylococcus aureus* growth.

Certificate of Attendance

We herewith certify that

Eddy Suswati

has attended and presented the abstract "*The Different Effect of Concentration and Contact Time of Chlorhexidine and Triclosan on the Growth of Methicillin Resistant Staphylococcus (MRSA)*" at the 10th International Symposium on Antimicrobial Agents and Resistance (ISAAR 2015) from May 14 to 15, Incheon, Korea.

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