CHAPTER V

DISCUSSION

Klebsiella pneumoniae is well known to most clinician as a cause of community acquired bacterial pneumonia. But the vast majority of *Klebsiella* infections, however, are associated with hospitalization. Nosocomial *Klebsiella* infections are caused mainly by *Klebsiella pneumoniae*, the medically most important species of the genus. It was estimated that *Klebsiella* spp. caused 8% of all nosocomial bacterial infection in the United States and in Europe. The urinary tract and respiratory tract were the two most common sites of infection for *Klebsiella pneumoniae*. The organism accounts for 6 to 17 % of all nosocomial urinary tract infection and an even higher incidence was shown in a specific groups of patients at risk, such as patients with neuropathic bladders or with diabetes mellitus (4). This study on the *Klebsiella pneumoniae* prevalent rate from various types of specimens showed the concordant results with those previous studies that the organism could be isolated from urine (2.21%) and sputum (2.94 %) with the low prevalence rate. The carrier rate of *K. pneumoniae* in feces of the normal Thai persons was as high as 36 % which was also the same as those reported in the prrevious studies. (81,82)

The clinical problem from *K. pneumoniae* infection is the failure to response to the cephalosporins group particular the third generation cephalosporins due to the

production of the extended spectrum β - lactamase. At present, there are various methods for detection of ESBL producing organisms (21). However, the most acceptable methods worldwide are the methods described by the NCCLS including the initial screen test for the detection of the suspicious ESBL producing organisms and the phenotypic confirmatory test by the combination disk method for the confirmed detection of ESBL producing organisms. According to the NCCLS, the suspicious ESBL producing organisms could be detected when the inhibition zone sizes by either one of the three drugs; ceftazidime or cefotaxime or ceftriaxone was in the recommended range. This study showed the interesting result that among all 123 *K. pneumoniae* isolates that were suspicious ESBL producers determined by the above criterion, only 104 isolates were showed to be the suspicious ESBL producing by showing positive result to all three drugs while the other 19 isolates (11 isolates from one drug and 8 isolates from two drugs criteria) were also accounted as suspicious for ESBL producers. After the phenotypic confirmatory test was performed, 96.1 % of the 104 suspicious ESBL producers from the three positive drugs criteria were shown to be ESBL producing isolates while none of the other 19 isolates were ESBL producers. It has been indicated in this study that the most reliable intial screen test should be done by using the criteria with all three cephalosporins. However, more specific study on the enzyme detection or the study on the sensitivity and specificity of the methods using known ESBL producing and non ESBL producing *K pneumoniae* should be performed before any conclusive informations could be made. In addition, it was also shown that normal person were not carrier of ESBL

producing *K. pneumoniae* because all of the 36 isolates were shown to be non - ESBL producer by both test.

The E -test ESBL screen test has been suggested by many previous investigators as the very useful method in the detection of ESBL producing organisms. (55, 57, 58) However, this study showed that only 71 isolates out of the 104 ESBL producing isolates determined by the NCCLS phenotypic confirmatory test were shown to be ESBL producer. Therefore, many other investigators during these past few years, includes Vercauteren E *et al.* (56), Hadziyannis E *et al* (59) and Carter MW *et al.* (60) supported the use of the NCCLS ESBL phenotypic confirmatory test by showing the superiority of such test over the other methods including E - test ESBL screen test.

The prevalence of ESBL producing *K. pneumoniae* was also report in this study that among the 436 total *K. pneumoniae* isolates from the clinical specimens, there were as high as 100 ESBL producing isolates which was 24.6% of the total isolates. This recovery rate was very high as compared to the recovery rate of ESBL producing *K. pneumoniae* from Southern Taiwan in the year 2000 which was as low as 8.5% (53). However, the incidence of the organism was high when it was recovered in the intensive care unit. It was reported that approximately 20 - 25% of *K. pneumoniae* isolated from the ICU from various European hospital were ESBL producers. The high recovery rate of ESBL producing isolates in Thailand might due

to the fact that there has been a tremendous uses of cephalosporins particular the third generation cephalosporins in the hospital.

Because of the problem in the antimicrobial therapy in the infections caused by ESBL - producing *K. pneumoniae*, the information of result of ESBL producing test and antimicrobial susceptibility of the organisms are necessary. The MIC determination by E - test was done with 5 cephalosporins included ceftazidime, cefotaxime, ceftriaxone, cefuroxime, and cefoxitin. The imipenem which was the representative drug of carbapenem group was also included. Interesting result from this study showed that most of the isolates (79, 87 and 90%, respectively) were susceptible to ceftriaxone, cefotaxime and cefoxitin but very low numbers of isolates (5 - 30%) were susceptible to ceftazidime and cefuroxime, respectively. This might be due to the more frequent use of ceftazidime and cefuroxime in the treatment of the infectious diseases particularly due to the gram negative bacteria in the hospital. On the other hand, care must be taken in order to report the susceptibility of ESBL - producing *K. pneumoniae* to the other second and third generation cephalosporins when the isolates were shown to be ceftazidime resistance because number of the studies reported the failure of the use of those cephalosporins in the treatment of the infections due to this organism (83). Fortunately, all of ESBL producing was still sensitive to imipenem, the member of the carbapenems. However, the use of carbapenem against the ESBL - producing *K. pneumoniae* must be careful. The other groups of antimicrobial agents were tested using the Kirby-Bauer susceptibility test. It

was shown that the organisms were moderately susceptible to, were amikacin (71%), ciprofloxacin (67%) and gentamicin (50%). The ESBL producing isolates were poorly susceptible to trimethoprim - sulfamethoxazole (38%) and tobramycin (16%). Therefore, the percentage of the susceptible isolates in this study was low when compared to those reported from the routine Bacteriology laboratory of the hospital (84). In such report, it was indicated that 88% of *K. pneumoniae* were susceptible to amikacin while 80%, 80% and 55% were susceptible to ciprofloxacin, gentamicin and trimethoprim - sulfamethoxazole, respectively. However, these data were collected from all *K. pneumoniae* isolates which included both ESBL - producing and non ESBL - producing isolates.

From this point of view, detection of ESBL - producing organisms particular *K. pneumoniae* is necessary. More cooperation between the clinicians and the laboratory staffs to some extend is adviced.

It has been known for a long time that most of the ESBL had an ability to hydrolyze specific oxyimino - cephalosporins including ceftazidime cefotaxime and ceftriaxone (6). These enzymes have the ability to expand the hydrolytic activity more than the classical β -lactamase ; TEM - 1, TEM - 2 and SHV enzymes. This enhances the ability to cause resistance. However, the carbapenems are still resist to this enzymatic activity. More than 30 different types of ESBL have been described which occur predominantly in *Klebsiellae.* (6). The predominant types of the enzymes varied geographically such as TEM - 10 and TEM - 26 were common in the United States

(49), SHV - 5 in Germany (85) and SHV - 2 was widespread internationally (6). There still has been no report on the types of ESBL produced from *K. pneumoniae* isolated from the clinical specimens in Thailand. By the evaluation of the data from the study on the MICs of the two cephalosporins; ceftazidime and cefotaxime, preliminary results on the types of ESBL could be obtained. Thirty - two isolates produced ESBL with the broad type activity suggesting them to be related to TEM - 3, TEM - 4 and SHV - 4, SHV - 5 (80). The ceftazidimase activity which was related to TEM - 10 and TEM - 26 was produced by 13 isolates and 55 isolates produced undetermined type of enzyme which raised the MICs of ceftazidime to >2 μg/ml (80). However, further study on the characteristics of ESBL produced by the isolates obtained from this study should be performed before any conclusions about the types of enzymes could be made.

The molecular typing of all 100 ESBL producing *K. pneumoniae* was also performed using pulsed-field gel electrophoresis technique. The purpose to do this part of the study was to determine the variation of types of ESBL producing *K. pneumoniae* isolated from various specimens from different patient wards at Siriraj hospital during the 6 months of the study period. It seemed that there was neither particular endemic nor epidemic strain among the ESBL producing *K. pneumoniae* isolated from different patient units. There were as many as 86 pulsotypes of *K. pneumoniae* among all 100 isolates studied. Eventhough, it was found that there were few isolates obtained from the patients in the same patient unit that were the same pulsotype but the number of the isolates were too low to indicate that there

was an outbreak during the period of study. In addition, most of the isolates exhibited their unique profiles. The result from this part of the study indicated that the characteristics of *K. pneumoniae* was similar to that of *P. aeruginosa* and possibly some other gram - negative bacteria that the organism that colonized or infected the patients tended to originate from the patients themselves. (86) However, this study was the minute survey study and not designing to do the outbreak study in the specific patient units, it is still not conclusive that there is no the transmission of some epidemic strains in Siriraj Hospital. Further study on the pulsotyping of ESBL producing *K. pneumoniae* in each patient unit throughout the hospital should be done in order to make a final conclusion on the epidemiology of the organism.