

## REFERENCES

1. Grundmann H., Kropee A., Hartong D., Berner R., and Daschner F. 1993. *Pseudomonas aeruginosa* in a Neonatal Intensive Care Unit : Reservoirs and Ecology of the Nosocomial Pathogen. J. Infect. Dis. 168:943-947.
2. Whitby J.L., rampling A. 1972. *Pseudomonas aeruginosa* contamination in domestic and hospital environment. Lancet. 1:15-17.
3. Favevo M.S., Carson L. A., Bnd W.W., Peterson N.J. 1971. *Pseudomonas aeruginosa* : growth in distilled water from hospitals. Science. 173:836-838.
4. Burden D. W., Whitby J.L., 1967. Contamination of hospital disinfectants with *Pseudomonas* species. Br. Med. J. 2:153-155.
5. Lechi A., arosio E., Pancera P., et al. 1984. *Pseudomonas septicemia*. A review of 60 cases observed in a university hospital. J. Hosp. Infect. 5:29-37.
6. Olson B., Weinstein R.A., Nathan C., Chamberlin W., Kabins S. A. 1984. Epidemiology of endemic *Pseudomonas aeruginosa*. Why infection control efforts have failed. J. Infect. Dis. 150:808-816.
7. Cobben N.A.M., Drent M., Jonkers M., Wouters E.F.M., Vaneechoutte M., and Stoberingh E.E. 1996. Outbreak of severe *Pseudomonas aeruginosa* respiratory infections due to contaminated nebulizers. J. Hosp. Infect. 33:63-70.
8. Spencer R.C. 1994. Epidemiology of infection in ICUs. Intensive Care Med. 20:S2-S6.
9. Trila A. 1994. Epidemiology of nosocomial infections in adult intensive care units. Intensive Care Med. 20:S1-S4.
10. Bailey and Scott's. 1986. Nonfermentative gram-negative bacilli and coccobacilli, p.422-433. In S.M. Finegold and E.J. Baron (ed), *Diagnostic Microbiology*, 7<sup>th</sup> ed. St. Louis, Toronto, Princeton.
11. Arbeit R.D. 1995. Laboratory procedures for the epidemiologic analysis of

- microorganism, p. 190-208. In P. R. Murray, E.J. Baron, A.A. Pfaller, F. C. Tenover, and R. H. Tenover (ed), *Manual of clinical microbiology*, 6<sup>th</sup> ed. American Society for microbiology, Washington, D.C.
12. Rossello J., et al. 1992. Investigation of an outbreak of nosocomial infection due to a multiply drug resistant strain of *Pseudomonas aeruginosa*. *J. Hosp. Infect.* 20:87-96.
13. Jumaa P. and Chattopadhyay B. 1994. Outbreak of gentamicin, ciprofloxacin-resistant *Pseudomonas aeruginosa* in an intensive care unit, traced to contaminated quivers. *J. Hosp. Infect.* 28:209-218.
14. Arbeit, R. D., A. Slutsky, T. W. Barber, J.N. Maslow, S. Niemczyk, J. O. Falkinham III, G. T. O'Connor, and C. F. von Reyn. 1993. Genetic diversity among strains of *Mycobacterium avium* causing monoclonal and polyclonal bacteremia in patients with AIDS. *J. Infect. Dis.* 167: 1384-1390.
15. Tenover F.C. et al. 1995. Interpreting chromosomal DNA restriction patterns produced by pulsed-field gel electrophoresis: criteria for bacterial strain typing. *J. Clin. Microbiol.* 33: 2233-2239.
16. Bingen E. et al. 1996. Molecular epidemiology provides evidence of genotypic heterogeneity of multidrug-resistant *Pseudomonas aeruginosa* serotype O:12 outbreak isolates from a pediatric hospital.
17. Botzenhard K, Doring G. 1993. Ecology and epidemiology of *P. aeruginosa*, p. 7-13. In Campa M., Bendinelli M., and Friedman H. (ed), *Pseudomonas aeruginosa as an opportunistic pathogen*, 1<sup>st</sup> ed. Plenum, New York.
18. Folkhard, W., et al. 1979. X-ray diffraction and electron microscopic studies on the structure of bacterial F-pili. *J. Mol. Biol.* 130: 145-160.
19. Paranchych, W., and Frost, L. S., 1988. The physiology and biochemistry of pili. *Adv. Microbiol. Phys.* 29: 53-114.
20. Pasloske, B. L., Finlay, B. B., and Paranchych, W., 1985. Cloning and sequencing of *Pseudomonas aeruginosa* PAK pilin gene. *FEBS Lett.* 183: 408-412.

21. Irvin R. T. 1993. Attachment and colonization of *Pseudomonas aeruginosa*: role of the surface structures, p. 19-36. In Campa M., Bendinelli M., and Friedmam H. (ed), *Pseudomonas aeruginosa as an opportunistic pathogen*, 1<sup>st</sup> ed. Plenum, NewYork.
22. Schmidt, M. A., and O'Hanley, P., 1988. Synthetic peptides corresponding to protective epitopes of *Escherichia coli* digalactoside-binding pilin prevent infection in a murine pyelonephritis model. Proc. Natl. Acad. Sci. USA 85:1247-1251.
23. Linker, A., and Jones, R. S., 1966. A new polysaccharide resembling alginic acid isolated from *Pseudomonas*. J. Biol. Chem. 241: 3845-3856.
24. Hoiby, N., 1974. *Pseudomonas aeruginosa* infection in cystic fibrosis. Relationship between mucoid strain of *P. aeruginosa* and humoral immune response. Acta. Pathol. Microbiol. Scand. Sect. B 82: 321-327.
25. Cheg, K. J., Irvin R. T., and Costerton, J. W., 1981. Autochthonous and pathogenic colonization of animal tissues by bacteria. Can. J. Microbiol. 27: 461-490.
26. Govan, J. R. W., and Fyfe, J. A. M., 1978. Mucoid *Pseudomonas aeruginosa* and cystic fibrosis: Resistance of the mucoid form to carbenicillin, flucloxacillin and tobramycin and the isolation of mucoid variants in vitro. J. antimicrob. Chemother. 4: 233-240.
27. Schwarzmann, S., and Boring, J. R., III, 1971. Antiphagocytic effect of slime from a mucoid strain of *Pseudomonas aeruginosa*. Infect. Immun. 3: 762-767.
28. Learn, D. B., Brestel, E. P., and Seetharama, S., 1987. Hypochlorite scavenging by *Pseudomonas aeruginosa* alginate. Infect. Immun. 55: 1813-1818.
29. Darzins, a., and Chakrabarty, A. M., 1984. Cloning of genes controlling algiate biosynthesis from a mucoid cystic fibrosis isolate of *Pseudomonas aeruginosa*. J. Bacteriol. 159: 9-18.
30. Pier, G. B., et al. 1986. Polysaccharide surface antigens expressed by nonmucoid isolates of *Pseudomonas aeruginosa* from cystic fibrosis patients. J. Clin.

- Microbiol. 24: 189-196.
31. Pugashetti, K. B., Metzger, H. M., Jr., Vadas, L., and Feinold, D. S., 1982. Phenotypic differences among clinically isolated mucoid *Pseudomonas aeruginosa* strains. J. Clin. Microbiol. 16: 686-691.
  32. Piggott, N. G., Sutherland, I. W., and Jarman, T. R., 1982. Alginate synthesis by mucoid strains of *Pseudomonas aeruginosa* PAO. Eur. J. App. Microbiol. Biotechnol. 16: 131-135.
  33. Pier, G. B., Matthews, W. J., and Eardley, D. D., 1983. Immunochemical characterization of the mucoid exopolysaccharide of *Pseudomonas aeruginosa*. J. Infect. Dis. 147: 494-503.
  34. McArthur, H. A. I., and Ceri, H., 1983. Interaction of a rat lung lectin with the exopolysaccharides of *Pseudomonas aeruginosa*. Infect. Immun. 42: 574-578.
  35. Pavlovskis, O. R., Iglewski, B. H., and Pollack, M., 1978. Mechanism of action of *Pseudomonas aeruginosa* exotoxin A in experimental mouse infections: Adenosine diphosphate ribosylation of elongation factor2. Infect. Immun. 19: 29-33.
  36. Bjorn, M. J., Basil, M. L., Sadoff, J. C., and Iglewski, B. h., 1977. Incidence of exotoxin production by *Pseudomonas* species. Infect. Immun. 16: 362-366.
  37. Pavlovskins, O. R., and Gordon, F. B., 1972. *Pseudomonas aeruginosa* exotoxin: Effect on cell cultures. J. Infect. Dis. 125: 631-636.
  38. Cross, A. S., Sadoff, J. C., Iglewski, B. H., and Sokol, P. A., 1978. Evidence for the role of toxin A in the pathogenesis of infection with *Pseudomoas aeruginosa* in humans, J. Infect. Dis. 142: 538-546.
  39. Coburn, J., Wuatt, R. T., Iglewski, B. H., and Gill, D. M., 1989. Several GTP-binding proteins, including p21 c-H-ras, are preferred substrates of *Pseudomonas aeruginosa* exoenzyme S. J. biol. Chem. 264: 9004-9008.
  40. Woods, D. E., and Que, J. U., 1987. Purification of *Pseudomonas aeruginosa* exoenzyme S. Infect. Immun. 55: 579-586.

41. Nicas, T. I., et al. 1985. Role of exoenzyme S in chronic *Pseudomonas aeruginosa* lung infections. Eur. J. Clin. Microbiol. 4: 175-179.
42. Nicas, T. I., and Iglewski, B. H., 1984. Isolation and characterization of a transposon induced mutant of *Pseudomonas aeruginosa* deficient in exoenzyme S. Infect. Immun. 45: 470-474.
43. Wretling, B., and Pavlovskis, O. R., 1983. *Pseudomonas aeruginosa* elastase and its role in *Pseudomonas* infections. Rev. Infect. Dis. 5: 998-1004.
44. Nicas, T., and Iglewski, B. H., 1986. Production of elastase and other exoproducts by environmental isolates of *Pseudomonas aeruginosa*. J. Clin. Microbiol. 23: 967-969.
45. Wretling, B., and Wadstrom, T., 1977. Purification and properties of a protease with elastase activity from *Pseudomonas aeruginosa*. J. Gen. Microbiol. 103: 319-327.
46. Heck, L. D., Morihara, K., McRae, W. B., and Miller, E. J., 1986. Specific cleavage of human type III and IV collagens by *Pseudomonas aeruginosa* elastase. Infect. Immun. 51: 115-118.
47. Azghani, A. O., et al. 1990. Effects of *Pseudomonas aeruginosa* elastase on alveolar epithelial permeability in guinea pigs. Infect. Immun. 58: 433-438.
48. Woods, D. E., et al. 1986. Phenotypic comparison of *Pseudomonas aeruginosa* strains isolated from a variety of clinical sites. J. Clin. Microbiol. 24: 260-264.
49. Liu, P. V., 1966. The roles of various fractions of *Pseudomonas aeruginosa* in its pathogenesis. II. Effects of lecithinase and protease. J. Infect. Dis. 116: 112-116.
50. Doring, G., et al. 1983. Proteases of *Pseudomonas aeruginosa* in patients with cystic fibrosis. J. Infect. Dis. 147: 744-750.
51. Klinger, J. D., Straus, D. C., Hilton, C. B., and Bass, J. A., 1987. Antibodies to proteases and exotoxin A of *Pseudomonas aeruginosa* I patients with cystic fibrosis : Demonstration by radioimmunoassay. J. Infect. Dis. 138: 49-58.

52. Mull, J. D., and Callahan, W. S., 1965. The role of the elastase of *Pseudomonas aeruginosa* in experimental infection. Exp. Mol. Pathol. 4: 567-575.
53. Liu, P. V., 1979. Toxins of *Pseudomonas aeruginosa* p. 63-88 In : *Pseudomonas aeruginosa*-Clinical manifestations of infection and current therapy (R. G. Doggett, Ed.), Academic Press, New York.
54. Berka, R. M., and Vasil, M., 1982. Phospholipase C (heat-labile hemolysin) of *Pseudomonas aeruginosa* : Purification and preliminary characterization. J. Bacteriol. 152: 239-245.
55. Ostroff, R. M., and Vasil, M. L., 1987. Identification of a new phospholipase C activity by analysis of an insertional mutation in the hemolytic phospholipase C structural gene of *Pseudomonas*. J. Bacteriol. 169: 4597-4601.
56. Vasil, M. L., 1989. Molecular Biology of exotoxin A and phospholipase C of *Pseudomonas aeruginosa*, p. 3-14 In : *Pseudomonas* : Biotransformations, Pathogenesis, and Evolving Biotechnology, (S. Silver, A. M. Chakrabarty, B. H. Iglewski and S. Kaplan, eds.), Am. Soc. Microbiol., Washington, D. C.
57. Pritchard, A. E., and Vasil, M. L., 1986. Nucleotide sequence and expression of a phosphate-regulated gene encoding a secreted hemolysin of *Pseudomonas aeruginosa*. J. Bacteriol. 167: 291-298.
58. Coutinho, I. R., Berk, R.S., and Mammen, E., 1988. Platelet aggregation by a phospholipase C for *Pseudomonas aeruginosa*. Thromb. Res. 51: 1728-1730.
59. Meyers, D. J., and Berk, R.S., 1990. Characterization of phospholipase C from *Pseudomonas aeruginosa* as a potent inflammatory agent. Infect. Immun. 58: 659-666.
60. Bergmann, U., et al, 1989. Induction of inflammatory mediators (histamine and leukotrienes) from rat peritoneal mast cells and human granulocytes by *Pseudomonas aeruginosa* strains from burn patients. Infect. Immun. 57: 2187-2195.
61. Ostroff, R. M., Wretling, B., and Vasil, M. L., 1989. Mutations in the hemolytic-



- phospholipase C operon result in decreased virulence of *Pseudomonas aeruginosa* PAO1 grown under phosphate-limiting conditions. Infect. Immun. 57: 1369-1373.
62. Finland, M., Hones, w. F., and Barnes, M. W., 1959. Occurrence of serious bacterial infections since the introduction of antibacterial agents. J.A.M.A. 170: 2188-2197.
63. Stanley, M. M., 1947. *Bacillus pyocyaneus* infections: A review, report of cases and discussion of newer therapy including streptomycin. Am.J. Med. 2: 253-277.
64. Kerby, B. P., 1947. *Pseudomonas aeruginosa* bacteremia: summary of literature with report of a case. Amer. J. Dis. Child. 74: 610-615.
65. Whimbey, E., et al. 1987. Bacteremia and fungemia in patients with neoplastic disease. Am. J. Med. 82: 723-730.
66. Cooper, G. S., Havilir, D. S., Shlaes, D. M., and Salata, R. A., 1990. Polymicrobial bacteremia in the late 1980s: Predictors of outcome and review of the literature. Medicine. 69: 114-123.
67. Saroff, A. L., Armstrong, D., and Johnson, W. D., 1973. *Pseudomonas* endocarditis. Am. J. Cardiol. 32: 234-237.
68. Shekar, R., et al. 1985. Outbreak of endocarditis caused by *Pseudomonas aeruginosa* serotype O11 among pentazocine and tripeleennamine abusers in Chicago. J. Infect. Dis. 151: 203-208.
69. Botsford, K. B., et al. 1985. Selective survival in pentazocine and tripeleennamine of *Pseudomonas aeruginosa* serotype O11 from drug addicts. J. Infect. Dis. 151:209-216.
70. Cohen, P. S., Maguire, J. H., and weinstein, L., 1980. Infective endocarditis caused by gram-negative bacteria: A review of literature, 1945-1977. Prog. Cardiovasc. Dis. 22: 205-242.
71. Reyes, M. P., Palutke, W.A., Wylin, R. F., and Lerner, A. M., 1973. *Pseudomonas*

- endocarditis in the Detroit Medical Center 1969-1972. Medicine. 52: 173-194.
72. Rajashekaraiyah, K. R., Rice, T. W., and Kallick, C.A., 1981. Recovery of *Pseudomonas aeruginosa* from syringes of drug addicts with endocarditis. J. Infect. Dis. 144:482.
73. Rose, H. D., Heckman, M. G., and Unger, J. D., 1973. *Pseudomonas* pneumonia in adults. Am. Rev. Respir. Dis. 107: 416-422.
74. Hughes, J. M., 1988. Epidemiology and prevention of nosocomial pneumonia, p. 241-259 In : Current Clinical Topics in Infectious Diseases, Volume 9 (J. S. Remington, M. N. Swartz, eds.), McGrawHill, New York.
75. Pennington, J. E., Reynolds, H. Y., and Carbone, P. P., 1973. *Pseudomonas* pneumonia: A retrospective study of 36 cases, Am. J. Med. 55: 155-160.
76. Morrison, A. J., and Wenzel, R. P., 1984. Epidemiology of infections due to *Pseudomonas aeruginosa*. Rev. Infect. Dis. 6: S627-S642.
77. Montgomerie, J. A., and Morrow, J. W, 1980. Long-term *Pseudomonas* colonization in spinal cord injury patients. Am. J. Epidemiol. 112: 508-517.
78. Tong, M. J., 1972. Septic complications of war wounds. J. A. M. A. 219: 1044-1047.
79. Klein, R. S., berger, S. A., and Yekutieli, P., 1975. Wound infection during the Yom Kippur war: Observation concerning antibiotic prophylaxis and therapy. Ann. Surg. 182: 15-21.
80. Pruitt, B. A., Lindberg, R. B., McManus, W.F., and Mason, A. D., 1983. Current approach to prevention and treatment of *Pseudomonas aeruginosa* infections in burned patients. Rev. Infect. Dis. 5: S889-S897.
81. Harris, J. H., and Nadler, H. L., 1983. Incidence, genetics, heterozygote, and antenatal detection of cystic fibrosis, p.1-7 In : Textbook of Cystic Fibrosis, (J. D. Lloyd-Still, ed.), John Wright, Boston.
82. Rommens, J. M., et al. 1989. Identification of the cystic fibrosis gene: Chromosome walking and jumping. Science. 245: 1059-1065.



83. Tsui, L. C., 1991. Molecular genetics of cystic fibrosis and possible mechanisms of protein function, World Health Organization, Hereditary diseases program. Report of a joint WHO/ICF(M) A task force on cystic fibrosis. Annex 1, pp. 21-30.
84. Hoiby, N., 1982. Microbiology of lung infections in cystic fibrosis patients. Acta Paediatr. Scand. Suppl. 301: 33-54.
85. Wood, R. E., Boat, T. F., and doershuk, C. F., 1976. Cystic fibrosis: state of the art. Am. Rev. Respir. Dis. 113: 833-878.
86. Krogh-Johansen, H., Nir, M., Hoiby, N., Koch, C., and Schwartz, M., 1991. Severity of cystic fibrosis in patients homozygous and heterozygous for delta F508 mutation. Lancet. 337: 631-634.
87. Jensen, T., Pedersen, S. S., Hoiby, N., Koch, C., Flensburg, E. W. 1989. Use of antibiotics in cystic fibrosis. The Danish approach. Antibiol. Chemother. 42: 237-246.
88. Erika D., Lata V., Paola D., and Matthew S. 1997. Molecular epidemiology of acquisition of Ceftazidime-Resistant Gram-negative bacilli in a nonoutbreak setting. J. Clin. Microbiol. 35: 2602-2605.
89. Wachsmuth, K. 1985. Genotypic approaches to the diagnosis of bacterial infections: plasmid analyses and gene probes. Infect. Control. 6: 100-109.
90. Mekalanos, J. J. 1992. Environmental signals controlling expression of virulence determinants in bacteria. J. Bacteriol. 174: 1-7.
91. Eisenstein, B. I. 1990. New molecular techniques for microbial epidemiology and the diagnosis of infectious disease. J. Infect. Dis. 161: 595-602.
92. Mickelsen, P. A., et al. 1985. Instability of antibiotic resistance in a strain of *Staphylococcus epidermidis* isolated from an outbreak of prosthetic valve endocarditis. J. Infect. Dis. 152: 50-58.
93. Edouard B. et al, 1996. Molecular epidemiology provides evidence of genotypic heterogeneity of multidrug-resistant *Pseudomonas aeruginosa* serotype O:12

- outbreak isolates from a pediatric hospital. J. Clin. Microbiol. 34: 3226-3229.
94. Pfaller, M. A. 1991. Typing methods for epidemiologic investigation, p. 171-182. In A. Balows, W. J. Hausler, Jr., K. L. Herrmann, H. D. Isenberg, and H. J. Shadomy (ed.), Manual of Clinical Microbiology, 5<sup>th</sup> ed. American Society for Microbiology, Washington, D.C.
95. Pitt, T. L. 1988. Epidemiological typing of *Pseudomonas aeruginosa*. Eur. J. Clin. Microbiol. Infect. Dis. 7: 238-247.
96. Clabots, C. R., S. Johnson, M. M. Olson, L.R. Peterson, and D. N. Gerding. 1992. Acquisition of *Clostridium difficile* by hospitalized patients: evidence for colonized new admissions as a source of infection. J. Infect. Dis. 166: 561-567.
97. Southern, E. M. 1975. Detection of specific sequences among DNA fragments separated by gel electrophoresis. J. Mol. Biol. 98: 503-517.
98. Arthur, M., R. D. Arbeit, C. Kim, P. Beltran, H. Crowe, S. Steinbach, C. Campanelli, R.A. Wilson, R. K. Selander, and R. Goldstein. 1990. Restriction fragment length polymorphisms among uropathogenic *Escherichia coli* pap-related sequences compared with *rrn* operons. Infect. Imm. 58: 471-479.
99. Rabkin, C., et al. 1989. *Pseudomonas cepacia* typing systems: collaborative study to assess their potential in epidemiologic investigations. Rev. Infect. Dis. 11: 600-607.
100. Robert, D. A. 1995. Laboratory procedures for the epidemiologic analysis of microorganisms, p. 190-208. In E.J. Baron, M. A. Bfaller, F. C. Tenover, and R.H. Tenover (ed), Manual of Clinical Microbiology, 6<sup>th</sup> ed. Washington, D. C.
101. Schwartz, D. C., and C. R. Cantor. 1984. Separation of yeast chromosome-sized DNAs by pulsed field gradients gel electrophoresis. Cell. 37: 67-75.
102. Birren, B., and E. Lai. 1993. Pulsed Field Gel Electrophoresis: a Practical Guide. Academic Press, Inc., San Diego.
103. Arbeit, R. D., et al. 1990. Resolution of recent evolutionary divergence among *Escherichia coli* from related lineages: the application of pulsed field

- electrophoresis to molecular epidemiology. J. Infect. Dis. 161: 230-235.
104. Talon D., Capellier G., Billot A. and Michel-Briand Y. 1995. Use of pulsed-field gel electrophoresis as an epidemiologic tool during an outbreak of *Pseudomonas aeruginosa* lung infections in an intensive care unit. Intens. Care. Med. 21: 996-1002.
105. Boukadida J. et al. 1993. Molecular epidemiology of chronic pulmonary colonization by *Pseudomonas aeruginosa* in cystic fibrosis. J. Med. Microbiol. 38: 29-33.
106. Horan, T. C., et al. 1986. Nosocomial infections surveillance. Morbid. Mortal. Wkly. Rep. 35: 17-29.
107. Brawley, R. L., et al. 1989. Multiple nosocomial infections, an incidence study. Am. J. Epidemiol. 130: 769-780.
108. Horan, T. C., et al. 1988. Pathogens causing nosocomial infections. Antimicrobio. Newsletter. 5: 65-67.
109. McManus, A. T., 1989. *Pseudomonas aeruginosa*: A controlled burn pathogen. Antibiot. Chemother. 42: 103-108.
110. Tummler, B., et al. 1991. Nosocomial acquisition of *Pseudomonas aeruginosa* by cystic fibrosis patients. J. Clin. Microbiol. 29: 1265-1267.
111. Maki, D. G. , Alvarado, C. J., Hassemer, C. A., Zilz, M. A., 1982. Relation of the inanimate hospital environment to endemic nosocomial infection. New Engl. J. Med. 307: 1562-1566.
112. Wolz, C., et al. 1989. *Pseudomonas aeruginosa* cross-colonization and persistence in patients with cystic fibrosis. Use of a DNA probe. Epidemiol. Infect. 102: 205-214.
113. Kern, W., Wolz, C., and Doring, G., 1990. Molecular epidemiological study of *Pseudomonas aeruginosa* isolates from patients with acute leukemia. Eur. J. Clin. Microbiol. Infect. Dis. 9: 257-261.
114. Griffith, S. J., et al. 1989. The epidemiology of *Pseudomonas aeruginosa* in

- oncology patients in a general hospital. J. Infect. Dis. 160: 1030-1036.
115. Worlitzsch, D., et al. 1989. Molecular epidemiology of *Pseudomonas aeruginosa* urinary tract infections in paraplegic patients. Zentralbl. Hyg. 189: 175-184.
116. Widoner A. F., Wenzel R. P., Trilla A., Bale M. J., and Jones R. N., 1993. Outbreak of *Pseudomonas aeruginosa* infections in a surgical intensive care unit: probable transmission via hands of a healthcare worker. Clin. Infect. Dis. 16: 372-376.
117. Horan T., Culver D., and Harvis W. R., 1988. Pathogens causing nosocomial infections. Antimicrob Newsletter. 5: 65-67.
118. Cobben N. A. M., et al. 1996. Outbreak of severe *Pseudomonas aeruginosa* respiratory infections due to contaminated nebulizers. J. Hosp. Infect. 33: 63-70.
119. Kerr J. R., et al. 1995. Investigation of a nosocomial outbreak of *Pseudomonas aeruginosa* pneumonia in an intensive care unit by random amplification of polymorphic DNA assay. J. Hosp. Infect. 30: 125-131.
120. Zembrzuska-Sadkowska E., et al. 1995. Epidemiology of *Pseudomonas aeruginosa* infection and the role of contamination of the environment in the Danish Cystic Fibrosis Center. J. Hosp. Infect. 29:1-7.
121. Doring, G., et al. 1991. Generation of *Pseudomonas aeruginosa* aerosols during handwashing from contaminated sink drains, transmission to hands of hospital personnel, and its prevention by use of a new heating device. Zentralbl. Hyg. 191: 494-505.
122. Brown, D. G., and Baublis, J., 1977. Reservoirs of *Pseudomonas* in an intensive care unit for newborn infants: mechanisms of control. J. Pediatr. 90: 453-457.
123. Chadwick, P., 1973. Relative importance of airborne and other routes in the infection of tracheostomised patients with *Pseudomonas aeruginosa*, p. 481-489 In *Airborne Transmission and Airborne Infection*, 6<sup>th</sup> Intern. Symp. On Aerobiology, (J. F. Hers and K. C. Winkler, eds.), Oosthock Publ. Co., Utrecht,

The Netherlands.

124. Gohn G et al. 1987. Laboratory diagnosis of lower respiratory tract infections. *Cumitech* 7a. Sep. : 9-10.
125. Gillgan, P.H. 1995. *Pseudomonas* and *Burkholderia*. p. 509-519. In P. R. Murray, E.J. Baron, A.A. Pfaller, F. C. Tenover, and R. H. Tenover (ed), *Manual of clinical microbiology*, 6<sup>th</sup> ed. American Society for microbiology, Washington, D.C.
126. Bauer A.N., Kirby W.M.M., Sherris J. C., Tenover M. 1966. Antibiotic susceptibility testing and standardized single disc method. *Am. J. Clin. Pathol.* 45: 493-496.
127. Jorgensen, J. H. et al. 1993. Performance standards for antimicrobial disk susceptibility tests fifth edition; approved standard. NCCLS Document M2-A5.
128. Maslow et al. 1993. The application of Pulsed-Field Gel Electrophoresis to molecular epidemiology, p. 563-572. In D. H. Persing, T. F. Smith, F. C. Tenover, and T. J. White (ed), *Diagnostic Molecular Microbiology : Principles and Applications*. American Society for Microbiology, Washington, D.C.
129. Poh, C. L., Yeo, C.C., Tah, L. 1992. Genome fingerprinting by pulsed-field gel electrophoresis and ribotyping to differentiate *Pseudomonas aeruginosa*. *Eur. J. Clin. Microbiol. Infect. Dis.* 11: 817-822.
130. Kersulyte D. et al. 1995. Comparison of arbitrarily primed PCR and macrorestriction (Pulsed-field gel electrophoresis) typing of *Pseudomonas aeruginosa* strains from cystic fibrosis patients. *J. Clin. Microbiol.* 33: 2216-2219.
131. Griffith S. J., et al. 1989. The epidemiology of *Pseudomonas aeruginosa* in oncology patients in a general hospital. *J. Infect. Dis.* 160: 1030-1036.
132. Grundmann. H., et al. 1993. *Pseudomonas aeruginosa* in a neonatal intensive care unit: reservoirs and ecology of the nosocomial pathogen. *J. Infect. Dis.* 168: 943-947.

133. Strulens, M. J., Schwam, A.V., Deplano, A., and Baran, D. 1993. Genome macrorestriction analysis of diversity and variability of *Pseudomonas aeruginosa* strains infecting cystic fibrosis patients. J. Clin. Microbiol. 31: 2320-2326.



สถาบันวิทยบริการ  
จุฬาลงกรณ์มหาวิทยาลัย



## APPENDIX I

### Media, Chemical agents, Materials and Instruments

#### A. Media

##### 1. Trypticase soy broth medium

Casein peptone	17.0	g.
Soy peptone	3.0	g.
Glucose (dextrose)	2.5	g.
Sodium chloride	5.0	g.
Dipotassium phosphate	2.5	g.
Distilled water	1000.0	ml.

##### 2. Trypticase soy agar medium

Casein peptone	15.0	g.
Soy peptone	5.0	g.
Sodium chloride	5.0	g.
Agar	15.0	g.
Distilled water	1000.0	ml.

##### 3. Nutrient agar medium

Meat (beef) extract	10.0	g.
Peptone	10.0	g.
Sodium chloride	5.0	g.
Nutrient agar	15.0	g.

#### 4. Mueller-Hinton agar medium

Beef, Infusion from	300.0	g.
Casamino acid, Technical	17.5	g.
Bacto soluble starch	1.5	g.
Bacto agar	17.0	g.
Distilled water	1000.0	ml.

#### 5. Tryptose blood agar base medium

Bacto tryptose	10.0	g.
Bacto beef extract	3.0	g.
Sodium chloride	5.0	g.
Bacto agar	15.0	g.
Distilled water	950.0	ml.

#### Media preparation

All of ingredients were dissolved in distilled water and then sterilized by autoclaving at 121°C, 15 pounds/inch<sup>2</sup> pressure, for 15 minutes. The sterilized medium was cooled to 55°C, and dispensed into sterile plates or tubes. For sterile tryptose blood agar base medium, 50 ml of sterile defibrinated blood was added aseptically into sterile plates before dispensation.

#### 6. Tryptone sugar iron agar (TSI) medium

Meat extract	3.0	g.
Yeast extract	3.0	g.
Peptone	20.0	g.
Glucose	1.0	g.
Lactose	10.0	g.
Sucrose	10.0	g.

FeSO <sub>4</sub> .7H <sub>2</sub> O	0.2 g.
OR [Ferric citrate]	[0.3] g.
NaCl	5.0 g.
Na <sub>2</sub> S <sub>2</sub> O <sub>3</sub> .5H <sub>2</sub> O	0.3 g.
Agar	15.0 g.
Distilled water	1000.0 ml.
Phenol red 0.2% aq. solution	12.0 ml.

All of ingredients except indicator were heated to dissolve the solids in distilled water. The indicator solution was added and then mixed and dispensed into tubes. The tubes were sterilized by autoclaving at 115°C, 15 pounds/inch<sup>2</sup>, for 20 minutes and then cooled to form slopes with deep butts about 3 cm along.

#### 7. O-F carbohydrate base medium

##### 7.1 10% aqueous carbohydrate solutions (glucose and lactose)

carbohydrate (glucose and lactose) 2.0 g. was added into distilled water 20 ml.

And then immediately sterilized by passing them through a 0.2 μm membrane filter.

##### 7.2 O-F carbohydrate base medium

Peptone or tryptone	2.0 g.
Sodium chloride	5.0 g.
Agar	2.5 g.
Dipotassium phosphate	0.3 g.
Bromthymol blue	0.03 g.
Distilled water	1000.0 ml.

All of ingredients were dissolved in distilled water and divided the solution into several smaller flasks with known volume and then sterilized by autoclaving at

121 °C, 15 pounds/inch<sup>2</sup> pressure, for 15 minutes. The sterile medium was cooled to 55°C.

Twenty ml of the sterile 10% carbohydrate solution was added aseptically into the flask of O-F base. The O-F carbohydrate base medium was dispensed into 16-125 mm screw cap test tubes. The tube was allowed to solidify upright, tighten caps, and refrigerated.

#### B. Chemical agents

- InCert agarose (Promega, USA)
- SeaKem agarose (Gibco BRL, Spain)
- Brij-58 (Sigma, USA)
- Sodium deoxycholate (Sigma, USA)
- Sodium lauroyl sarcosine (Sigma, USA)
- Rnase A (Amresco, USA)
- Proteinase K (Amresco, USA)
- Tris (Amresco, USA)
- Sodium chloride (Merck, Germany)
- EDTA (Amresco, USA)
- Boric acid (Bio-Rad, USA)

#### C. Materials

- 15-ml snap-top tubes (Fisher, USA)
- 5-ml snap-top tubes (Fisher, USA)
- 15-ml round bottom tube, screw cap (Pyrex, USA)
- 16x125 mm, screw cap test tube (Pyrex, USA)
- Insert mold (Bio-Rad, USA)
- Glass tray
- Metal tray

#### D. Instruments

Incubator 37°C, 42°C (Mettler, Germany)

Shaking water bath (United Instrument, USA)

Turbidity meter

Mixer Vortex (Scientific, USA)

Caliper

Roller (Life Science, USA)

Refrigerator centrifuge (4°C) (Sigma, USA)

Refrigerator (-70°C) (Forma Scientific, USA)

Refrigerator (-20°C) (Listed Household Freezer, USA)

Automatic pipette, p20/p200/p1000 (Gilson Medical Electronic, France)

pH meter (Beckman, USA)

Micropore filter (Pyrex, USA)

Pulsed-Field Gel Box (Bio-Rad, USA)

Pump, Gel Molds (Bio-Rad, USA)

Cooling water bath (Bio-Rad, USA)

Power supply, Pulse wave switcher (Bio-Rad, USA)

Gel Doc 1000 (Bio-Rad, USA)

Standard Woods' lamp (CAMAGE, Schweiz)

#### E. Enzyme and Molecular Marker

*SpeI* (Boehringer, Germany)

$\lambda$  ladder marker (Bio-Rad, USA)

## APPENDIX II

### Reagents

#### 1. PIV buffer

- 10 mM Tris base (pH 7.6)
- 1M NaCl

Tris-base and NaCl were dissolved in ultra pure water and adjusted the pH to 7.6 by adding 1N HCl and then steriled by autoclaving at 121°C, 15 pounds/inch<sup>2</sup> pressure, for 15 minutes. The buffer was stored at 4°C until used.

#### 2. Lysis buffer

- 6 mM Tris base (pH 7.6)
- 1M NaCl
- 100mM EDTA (pH 7.6)
- 0.5% Brij-58
- 0.2% Sodium deoxycholate
- 0.5% Sodium lauryl sarcosine

All of ingredients were dissolved in ultra pure water and adjusted the pH to 7.6 by adding 1N HCl and then steriled by passing them through a 0.4 µm membrane filter.

#### 3. Lysis solution

- 20 µg of Rnase per ml

Proteinase K was dissolved in ultra pure steriled water and then dispensed into microcentrifuge tube. The RNase solution was stored at -20°C until used.



- 1 mg of lysozyme per ml

Lysozyme was dissolved in ultra pure steriled water and then dispensed into microcentrifuge tube. The lysozyme solution was stored at  $-20^{\circ}\text{C}$  until used.

#### 4. ES buffer

- 0.5M EDTA (pH 8.0)

- 10% Sodium lauryl sarcosine

EDTA and sodium lauryl sarcosine were dissolved in ultra pure water and adjusted the pH to 8.0 by adding concentrated HCl. The buffer was steriled by passing them through a  $0.4\ \mu\text{m}$  membrane filter.

#### 5. ESP solution

- 20X proteinase stock solution

Fifty gram of proteins K was dissolved in 50 ml of ES buffer and then incubated in  $50^{\circ}\text{C}$  water bath for 1 hr. The proteinase K stock solution was stored at  $4^{\circ}\text{C}$  until used.

The ESP buffer was freshly prepared by adding aseptically 5 ml of the proteinase K stock solution into 95 ml of ES buffer. The ESP buffer was stored at  $4^{\circ}\text{C}$  until used.

#### 6. 1X TE buffer

- 10 mM Tris base (pH 7.6)

- 0.1 M EDTA (pH 7.6)

Tris-base and EDTA were dissolved in ultra pure water and adjusted the pH to 7.6 by adding 1N HCl. The buffer was steriled by autoclaving at  $121^{\circ}\text{C}$ , 15 pounds/inch<sup>2</sup> pressure, for 15 minutes. The buffer was stored at  $4^{\circ}\text{C}$  until used.

#### 7. 10xTBE buffer (Tris borate buffer)

- 10 M Tris base (pH 7.6)
- 0.1 M Boric acid
- 4 mM EDTA (pH 8.5)

All of ingredients were dissolved in ultra pure water and adjusted the pH to 7.6 by adding 1N HCl. The buffer was sterilized by autoclaving at 121°C, 15 pounds/inch<sup>2</sup> pressure, for 15 minutes. The buffer was stored at room temperature until used.

#### 8. Ethidium bromide solution

Ethidium bromide stock solution (5 mg/ml in water) was diluted to 0.5 µg/ml in water.



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