豬霍亂紗門氏菌與小鴨沙門氏菌攜帶Ceftriaxone抗藥基因質體之物理圖譜分析

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摘 要

近年來臨床上抗藥性細菌大幅出現，尤其是對第三代頭孢菌素 (cephalosporins) 抗藥性腸內菌越來越多。篩選對第三代頭孢菌素抗藥之菌株，自林口長庚醫院受感染病人的檢體中分離出Salmonella enterica serotype Choleraesuis (SC72) 和Salmonella enterica serotype Anatum (SA323)，都攜帶ceftriaxone抗藥性之質體 (分別為pSC72-1及pSA323-1)，這是台灣地區罕見具ceftriaxone抗藥性之沙門氏菌。針對兩質體進行聚合?鏈鎖反應及序列分析之研究，發現兩質體都帶有blaCTX-M-3抗藥基因，且抗藥基因是由transposon所攜帶。兩質體分別和Salmonella enterica serotype Typhimurium (LBNP4417) 及E. coli (HB101) 進行接合實驗，顯示有接合能力 (conjugative)。進一步分析質體間之差異，pSC72-1大小約為74 kb，而pSA323-1大小約為82 kb，分別利用三種不同限制?，以單一或混合兩種限制?做切割，呈現限制?多樣性，再以限制?切割片段之差異性構築其物理圖譜。由南方墨點雜合法結果顯示兩質體除了攜帶同種抗藥基因之外，其相關性不高。 由於臨床上分離出攜帶blaCTX-M-3抗藥基因的菌株多為E. coli 和Klebsiella pneumoniae，從林口長庚醫院受感染病人的檢體中分離出4株E. coli及5株K. pneumoniae，由聚合?鏈鎖反應、序列分析及南方墨點雜合法的結果顯示其抗藥基因blaCTX-M-3位於質體上，也是由transposon所攜帶。利用接合實驗分離出具有接合能力的質體pKP104-1和pKP116-1，以限制?做切割，兩質體之限制?片段一致，和pSC72-1、pSA323-1的限制?片段比較，亦呈現限制?多樣性。由pKP104-1和pKP116-1限制?切割的結果顯示攜帶blaCTX-M-3抗藥基因之質體已經在K. pneumoniae菌株之間傳播。由已知nikA基因序列設計引子，進行聚合?鏈鎖反應及序列分析，其結果顯示除了HB101/323-1之質體 (pSA323-1) 外，其他臨床分離菌株質體的接合系統 (conjugation system) 都一樣。 根據我們的了解，此研究應是台灣地區第一個構築S. Choleraesuis 和S. Anatum抗藥質體之物理圖譜。希望藉由此研究進一步了解其抗藥性及流行病學上的意義，並控制由 S. Choleraesuis 和S. Anatum所引起的感染。

關鍵詞 : Salmonella enterica serotype Choleraesuis ; Salmonella enterica serotype Anatum ; 抗藥基因 ; ceftriaxone ; 接合 ; 物理圖譜

目錄

參考文獻

Ambler, R. P. 1980. The structure of β-lactamases. Philos. Trans. R. Soc. London B. 289:321-331. Baraniak, A., J. Fiett, A. Sulikowska, W. Hyniewicz, and M. Gniadkowski. 2002. Countrywide spread of CTX-M-3 extended-spectrum β-lactamase —producing microorganism of the family Enterobacteriaceae in Poland. Antimicrob. Agents Chemother. 46:151-159. Barnass, S., J. Franklin, and S. Tabaqchali. 1990. The successful treatment of multiresistant non-enteric salmonellosis with seven day oral ciprofloxacin. J. Antimicrob. Chemother. 25:299-300. Bauernfeind, A., H. Grimm, and S. Schweighart. 1990. A new plasmidic cefotaximase in a clinical isolate of Escherichia coli. Infection. 18:294-298. Bauernfeind, A., I. Stemplinger, R. Jungwirth, S. Ernst, and J. M. Casellas. 1996. Sequences of beta-lactamase genes encoding CTX-M-1 (MEN-1) and CTX-M-2 and relationship of their amino acid sequences with those of other beta-lactamases. Antimicrob. Agents Chemother. 40:509-513. Bonnet, R. 2004. Minireview. Growing group of extended-spectrum β-lactamases: the CTX-M enzymes. Antimicrob. Agents Chemother. 48:1-14. Bradford, P. A. 2001. Extended-spectrum β-lactamases in the 21st century: characterization, epidemiology, and detection of this important resistance threat. Clin. Microbiol. Rev. 14:933-951. Bush, K., G. A. Jacoby, and A. A. Medeiros. 1995. A functional classification scheme for β-lactamases and its correlation with molecular structure. Antimicrob. Agents Chemother. 39:1211-1233. Cao, V., T. Lambert, and P. Courvalin. 2002. ColE1-like plasmid pIP843 of Klebsiella pneumoniae encoding extended-spectrum beta-lactamase CTX-M-17. Antimicrob. Agents Chemother. 46:1212-1217. Chiu, C. H., T. Y. Lin,and J. T. Ou. 1999. Prevalence of the virulence plasmids of nontyphoid Salmonella in the serovars isolated from humans and their association with bacteremia. Microbiol. Immunol. 43:899-903. Chiu, C. H., T. L. Wu, L. H. Su, C. Chu, J. H. Chia, A. J. Kuo, M. S. Chien, and T. Y. Lin. 2002. The emergence in Taiwan of fluoroquinolone resistance in Salmonella enterica serotype Choleraesuis. N. Engl. J. Med. 346:413-419. Doublet, B., A. Carattoli, J. M. Whichard, D. G. White, S. Baucheron, E. C. Dancla, and A. Cloeckaert. 2004. Plasmid-mediated florfenicol and ceftriaxone resistance encoded by the floR and blaCMY-2 gene in Salmonella enterica serovars Typhimurium and Newport isolated in the United States. FEMS Microbiol. Lett. 233:301-305. Eckert, C., V. Gautier, M. S. Allard, N. Hidri, C. Verdet, Z. O. Hocine, G. Barnaud, F. Delisle, A. Rossier, T. Lambert, A. Philippon, and G. Arlet. 2004. Dissemination of CTX-M-Type β-lactamase among clinical isolates of Enterobacteriaceae in Paris, France. Antimicrob. Agents Chemother. 48:1249-1255. Fisker, N., K. Vinding, K. Molbak, and M. K. Hornstrup. 2003. Clinical review of nontyphoid Salmonella infections from 1991 to 1999 in a Danish county. Clin. Infect. Dis. 37:e47-e52. Giraud, E., A. Cloeckaert, S. Baucheron, C. Mouline, and E. Chaslus-Dancla. 2003. Fitness cost of fluoroquinolone resistance in Salmonella enterica serovar Typhimurium. J. Med. Microbiol. 52:697-703. Hsu, R. B., Y. G. Tsay, R. J. Chen, and S. H. Chu. 2003. Risk factors for primary bacteremia and endovascular infection in patients without acquired immunodeficiency syndrome who have nontyphoid salmonellosis. Clin. Infect. Dis. 36:829-834. Kado, C. I. and C. T. Liu. 1981. Rapid procedure for detection and isolation of large and small plasmids. J. Bacteriol. 145:1365-1373. Kariuki, S., J. E. Corkill, G. Revathi, R. Musoke, and C. A. Hart. 2001. Molecular characterization of a novel plasmid-encoded cefotaximase (CTX-M-12) found in clinical Klebsiella pneumoniae isolates from Kenya. Antimicrob. Agents Chemother. 45:2141-2143. Klein, N.

C., and B. A. Cunha. 1995. The selection and use of ceplalosporins: a review. Adv. Ther. 12:83-101. Li, C. R., L. Yan, and P. A. Zhang. 2003. Dissemination and spread of CTX-M extended-spectrum β-lactamase among clinical isolates of Klebsiella pneumoniae in central China. Int. J. Antimicrob. Agents. 22:521-525. Ling, M. L., K. T. Goh, G. C. Wang, K. S. Neo, and T. Chua. 2002. An outbreak of multidrug-resistant Salmonella enterica subsp. enterica serotype Typhimurium, DT104L linked to dried anchovy in Singapore. Epidemiol. Infect. 128:1-5. Livermore, D. M. 1998. β-lactamase-mediated resistance and opportunities for its control. J.Clin. Microbiol. 41:25-41. McEvoy, G. K. 1998. Cephalosporins, P. 125-205. In McEvoy, G. K. (ed.), AHFS Drug Information 98. Bethesda; American Society of Health-System Pharmacists, Inc. Mead, P. S., L. Slutsker, and V. Dietz. 1999. Food-related illness and death in the United States. Emerg. Infect. Dis. 5:607-625. Medeiros, A. A. 1997. Evolution and dissemination of β-lactamases accelerated by generation of β-lactam antibiotics. Clin. Infect. Dis. 24:S19-S45. Moland, E. S., J. A. Black, A. Hossain, N. D. Hanson, K. S. Thomson, and S. Pottumarthy. 2003. Discovery of CTX-M-like extended-spectrum beta-lactamase in Escherichia coli isolates from five U.S. states. Antimicrob. Agents Chemother. 47:2382-2383. Molbak, K., D. L. Baggesen, F. M. Aarestrup, J. M. Ebbesen, J. Engberg, K. Frydendahl, P. G. Smidt, A. M. Petersen, and H. C. Wegener. 1999. An outbreak of multidrug-resistant, quinolone-resistant Salmonella enterica serotype Typhimurium DT104. N. Engl. J. Med. 341:1420-1425. Mulvey, M. R., E. Bryce, D. Boyd, M. O. Agostini, S. Christianson, A. E. Simor, S. Paton, and The Canadian Hospital Epidemiology Committee of The Canadian Nosocomial Infection Surveillance Program, Health Canada. 2004. Ambler class a extended-spectrum beta-lactamase-producing Escherichia coli and Klebsiella spp. In Canadian hospital. Antimicrob. Agents Chemother. 48:1204-1214. Olsen, S. J., E. E. Debess, T. E. Mcgivern, N. Marano, T. Eby, S. Mauvais, V. K. Balan, G. Zirnstein, P. R. Cieslak, and F. J. Angulo. 2001. A nosocomial outbreak of fluoroquinolone-resistant Salmonella infection. N. Engl. J. Med. 344:1572-1579. Oskoui, R., W. A. Davis, and M. N. Gomes. 1993. Salmonella aortitis. Arch. Intern. Med. 106:485-488. Pai, H., E. H. Choi, H. J. Lee, J. Y. Hong, and G. A. Jacoby. 2001. Identification of CTX-M-14 extended-spectrum beta-lactamase in clinical isolates of Shigella sonnei, Escherichia coli, and Klebsiella pneumoniae in Korea. J. Clin. Microbiol. 39:3747-3749. Palucha, A., B. Mikiewicz, W. Hryniewicz, and M. Gniadkowski. 1999. Concurrent outbreaks of extended-spectrum β-lactamase-producing organism of the family Enterobacteriaceae in a Warsaw hospital. J. Antimicrob. Chemother. 44:489-499. Partridge, S. R., and R. M. Hall. 2003. In34, a complex In5 family class 1 integron containing orf513 and dfrA10. Antimicrob. Agents Chemother. 47:342-349. Patrice, N. 1998. Trends in β-lactam resistance among Enterobacteriaceae. Clin. Infect. Dis. 27:S100-S106. Popoff M. Y., J. Bockemuhl, and L.L. Gheesling. 2003. Supplement 2001 (no. 45) to the Kauffmann-White scheme. Res. Microbiol. 154:173-174. Russell, A. D. 2002. Antibiotic and biocide resistance in bacteria: Introduction. J. Appl. Microbiol. Sym. Suppl. 92:1S-3S. Saladin, M., V. T. Cao, T. Lambert, J. L. Donay, J. L. Herrmann, Z. Ould-Hocine, C. Verdet, F. Delisle, A. Philippon, and G. Arlet. 2002. Diversity of CTX-M beta-lactamases and their promoter regions from Enterobacteriaceae isolated in three Parisian hospital. FEMS Microbiol. Lett. 209:161-168. Salmon, S. A., J. L. Watts, C. A. Case, L. J. Wegener, and R. J. Yancey Tr. 1995. Comparison of MICs of ceftiofur and other antibiotic agents against bacterial pathogens of swine from the United Stated, Canada, and Denmark. J. Clin. Microbiol. 33:2435-2444. Soto, S. M., B. Guerra, A. del Cerro, M. A. Gonzalez-Heria, and M. C. Mendoz. 2001. Outbreaks and sporadic cases of Salmonella serovar panama studied by DNA fingerprinting and antimicrobiol resistance. Int. J. Food Microbiol. 71:34-43. Su, L. H., C. H. Chiu, C.Chu, M. H. Wang, J. H. Chia, and T. L. Wu. 2003. In vivo acquisition of ceftriaxone resistance in Salmonella enterica serotype Anatum. Antimicrob. Agents Chemother. 47:563-567. Tassios, P. T., M. Gazouli, E. Tzelepi, H. Milch, N. Kozlova, S. Sidorenko, N. J. Legakis, and L. S. Tzouvelekis. 1999. Spread of a Salmonella typhimurium clone resistance to expanded-spectrum cephalosporins in three European countries. J.Clin. Microbiol. 37:3774-3777. Tauxe, R. V. 1997. Emerging foodborne diseases: an evolving public health challenge. Emerg. Infect. Dis. 3:425-434. Wang, H., S. Kelkar, W. Wu, M. Chen, and J. P. Quinn. 2003. Clinical isolates of Enterobacteriaceae producing extended-spectrum β-lactamases: prevalence of CTX-M-3 at a hospital in China. Antimicrob. Agents Chemother. 47:790-793. Ward, L. R., E. J. Threlfall, and B. Rowe. 1990. Multiple drug resistance in Salmonellae in England and Wales: a comparison between 1981 and 1988. J. Clin. Pathol.43:563-566. White, D. G., S. Zhaos, R. Sudler, S. Ayers, S. Friedman, S. Chen, S. McDermott, D. D. Wagner, and J. Meng. 2001. The isolation of antibiotic-resistant Salmonella from retail ground meats. N. Engl. J. Med. 345:1147-1154. Womble, D. D., and R. H. Rownd. 1988. Genetic and physical map of plasmids NR1: comparison with other IncFII antibiotic resistance plasmids. Microbiol. Rev. 433-451. Yamasaki, K., M. Komatsu, T. Yamashita, K. Shimakawa, T. Ura, H. Nishio, K. Satoh, R. Washidu, S. Kinoshita, and M. Aihara. 2003. Production of CTX-M-3 extended-spectrum β-lactamase and IMP-1 metallo β-lactamase by five gram-negative bacilli: survey of clinical isolates from seven laboratiories collected in 1998 and 2000, in the Kinki region of Japan. J. Antimicrob. Chemother. 51:631-638. Yan, J. J., W. C. Ko, S. H. Tsai, H. M. Wu, Y. T. Jin, and J. J. Wu. 2000. Dissemination of CTX-M-3 and CMY-2 β-lactamase among clinical isolates of Escherichia coli in southern Taiwan. J. Clin. Microbiol. 38:4320-4325. Yan, J.J., W. C. Ko, C. H. Chiu, S. H. Tsai, H. M. Wu, and J. J. Wu. 2003. Emergence of ceftriaxone-resistance Salmonella isolates and rapid spread of plasmid-encoded CMY-2-like cephalosporinase, Taiwan. Emerg. Infect. Dis. 9:323-328. Yang, Y. J., C. C. Liu, S. M. Wang, J. J. Wu, A. H. Huang, and C. P. Cheng. 1998. High rates of antimicrobial resistance among clinical isolates of nontyphoidal Salmonella in Taiwan. Eur. J. Clin. Microbiol. Infect. Dis. 17:880-883. Yu, W. L., L. T. Wu, M. A. Pfaller, P. L. Winokur, and R. N. Jones. 2003. Confirmaton of extended-spectrum β-lactamase-producing Serratia marcescens: preliminary report from Taiwan. Diagn. Microbiol. Infect. Dis. 45:221-224.