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Since bacterial resistance has been a major problem in Korea, we monitored antibiotic resistance of *Staphylococcus aureus* and *Streptococcus pneumoniae* strains isolated from hospital patients in Korea and studied resistance mechanisms of them in relation to stress proteins.

From minimum inhibitory concentrations (MICs) of 107 *S. aureus* strains isolated from hospital patients in the year 2000, the resistance rates were as follows; penicillin resistant, 99%; oxacillin resistant (MRSA), 80%; vancomycin resistant (VRSA), 0%. In the presence of Triton X-100, bacterial lysis of ATCC25923 (methicillin-susceptible *S. aureus*) and STA007 (methicillin-resistant *S. aureus*) were suppressed after heat shock (culture temperature was shifted from 30 °C to 40 °C for 10 minutes) and the suppression of lysis by heat shock was greater in the STA007 than in the ATCC25923.

When lysis of the wild type SKP3026 and its *clpL* mutant of *S. pneumoniae* strains by tetracycline were compared, lysis of the *clpL* mutant was faster than that of the wild type.

Heat shock suppressed bacterial autolysis in *S. aureus* and 84-kDa stress protein (ClpL) of *S. pneumoniae* suppressed autolysis by tetracycline. Therefore stress proteins do not seem to be the major mechanism of antibiotic resistance, but contribute to increase viability in resistant strains of *S. aureus* and *S. pneumoniae*.

[PC2-10] [04/19/2001 (Thr) 15:30 – 16:30 / Hall 4]

Bacterial Arylsulfate Sulfotransferase as a Reporter System

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In order to investigate whether the arylsulfate sulfotransferase (ASST) is suitable as a sensitive reporter system for Gram-positive bacteria, a reporter vector carrying the fragments of the *astA* structural region was constructed and designated as pSY815. To test the utility of the ASST reporter system in *Bacillus subtilis*, the regulatory regions of *ermC* and *ermAMR* were inserted upstream of the coding region of the reporter gene, to generate the vectors pSY815-EC and pSY815-ER, respectively. In the absence of an inserted regulatory region, the plasmid displayed very low background activity. The ASST activity under the control of the *ermC* regulatory region was increased 4.42-fold when induced by 0.1 µg/ml of erythromycin. Under the *ermAMR* regulatory control, the activity was increased 1.66-fold when induced by 0.2 µg/ml of tylosin. These results were consistent with a *lacZ* reporter gene assay of the *ermC* and *ermAMR* regulatory regions. This indicates that this reporter system is very sensitive.

The lack of endogenous activity, the simple detection of enzyme activity in the living cell, the commercially available non-toxic substrates, and the high sensitivity make ASST a useful genetic reporter system for monitoring gene expression and understanding gene regulation in Gram-positive bacteria.

[PC2-11] [04/19/2001 (Thr) 15:30 – 16:30 / Hall 4]

Genetic Characterization of Vancomycin-Resistant Enterococci from Raw Milk

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