

stress.

**H202**

### Scale-up and Purification of Plant Chitinase induced from Hypersensitive Stress

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We induced chitinase from soybean, cabbage, corn under various condition and the chitinase was purified chitin-affinity chromatography and CM-cellulose chromatography. The optimal pH of purified chitinase under osmotic stress was  $5.0 \pm 0.05$  and that was highly activated at 10 mM  $Zn^{2+}$  ion. The inhibition rate was 86% at *Phytophthora capsici*. The induced chitinase by fungal elicitor has optimal pH of  $6.0 \pm 0.2$  and activation of high value at *Fusarium oxysporium* elicitor included 12% oilgosaccharide. The inhibition rate was 67.3% at *Trichocheum viridet*. The activity of expressive chitinase by abiotic stress was highly activated at salinity concentration of 50 mM. The inhibition rate was 72% at *Fusarium oxysporium*. Cabbage callus cell expressed high levels of the antifungal activity among the plant cell. The optimum time and pH of treated *F. moniliforme* elicitor for antifungal agent production were 48 hours to 168 hours and pH  $5.8 \pm 0.05$  in medium. Antifungal activity was detected in the crude extracellular solution, which was expressed as units/ml crude solution, was in descending order of 168 hours > 144 hours and 126 hours > 96 hours. The accumulation of antifungal activity during drought stress was confirmed by enzyme activity.

**H202**

### Inactivation of Blood-Born Model Viruses in Low pH Intravenous

### Immunoglobulin

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Viral safety is a prerequisite for manufacturing clinical immunoglobulin solution which is prepared from human plasma pools. The study presented here was thus designed to evaluate the efficacy of incubation of final immunoglobulin solution at pH 3.86 for 14 days at 25° C for inactivation of several blood-born model viruses. Bovine viral diarrhoea virus (BVDV), Bovine herpes virus (BHV), Murine encephalomyocarditis virus (EMCV) and Porcine parvovirus (PPV) were selected for this study. Immunoglobulin solutions were spiked with the viruses and the amount of virus during incubation was kinetically quantified by 50% tissue culture infectious dose (TCID<sub>50</sub>). BVDV titer was reduced from an initial titer of 5.75 log<sub>10</sub> TCID<sub>50</sub> to undetectable level within 9 days of incubation. BHV titer was reduced from an initial titer of 8.61 log<sub>10</sub> TCID<sub>50</sub> to undetectable level within 5 days of incubation. EMCV titer was reduced from an initial titer of 8.39 log<sub>10</sub> TCID<sub>50</sub> to 3.20 log<sub>10</sub> TCID<sub>50</sub> after 14 days of incubation. PPV titer was reduced from an initial titer of 6.28 log<sub>10</sub> TCID<sub>50</sub> to 4.32 log<sub>10</sub> TCID<sub>50</sub> after 14 days of incubation. The log clearance factors achieved during the low pH incubation were >3.86 for BVDV, >7.43 for BHV, 5.19 for EMCV, and 1.96 for PPV. These results indicate that low pH incubation process for immunoglobulin has sufficient virus reducing capacity to achieve a high margin of virus safety.

**H302**

### Physiological Role of $\beta$ -lactamase Inhibitory Proteins (BLIP-I) in *Streptomyces exfoliatus* SMF19

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Two  $\beta$ -lactamase inhibitory proteins (BLIP-I and BLIP-II) was identified from *Streptomyces exfoliatus* SMF19 (Kang et al, JBC 2000). The gene consisting of 558bp (bliA) for BLIP- I and 1116bp (bliB) for BILP- II were identified, respectively. The bliA and bliB genes in the wild strain were disrupted by inserting hygromycin or apramycin resistance gene in the corresponding genes. The mutants disrupted single gene (bliA::hyg, bliB::apr) and and the both genes (bliA::hyg with bliB::apr) showed a bald phenotype, indicating that the these genes play a role in morphological differentiation.

**H303**

### Cloning, Sequencing and Expression of Phosphohydrolyase Gene in *Streptomyces clavuligerus* ATCC27064

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In oder to elucidate the physiological role of guanosine tetraphosphate (ppGpp) on the biosynthesis of antibiotics in *S. clavuligerus*, genes (sclA and sclB) those may encode phosphohydrolyase were cloned and identified. Those genes are heterologously expressed in *E. coli* and the gene products were purified for the preparation polyclonal antibody. The characteristics of the genes and the products were compared with those from *E. coli* and from other *Streptomyces* spp. From

the current research, the role of ppGpp or mechanism of ppGpp on the onset of secondary metabolism in *Streptomyces* spp. are going to be discussed.

**H304**

### Expression and Proteolytic Modification of Protease Inhibitor (SFI) from *Streptomyces fradiae* SMF9

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A proteinous protease inhibitor (SFI) produced in *Streptomyces fradiae* SMF9 was found to be encoded from a gene (sfi). The molecular weight of the SFI deduced from the DNA sequence was 21,450Da, although that determined by SDS-PAGE was 18,000Da. Moreover amino acid sequence of the active form (SFI: 18,000Da) and the inactive protein (SFIp: 21,450Da) was identical except 3 amino acids from the N-terminal. Hence it is thought that the SFI may be activated from the SFIp through post-translational modification. In order to elucidate the differences in the molecular weight and also to determine the site of cleavage at the post-translational modification step, C-terminal sequence of SFI and the N-terminal sequence of the protein (3,000Da) cleaved from the SFIp are being determined.

**H305**

### Mannanase를 생산하는 *Bacillus* sp. WL-1 균주의 분리 및 mannanase의 생산

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토양으로부터 Mannase를 분리하는 미생