

C001

### Co-culturing of *Bifidobacterium Bifidum* for Cholesterol Removal is Related to Bile Salt Hydrolase Activity.

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To determine the validity of the hypothesis of assimilation and/or precipitation of cholesterol, three different strains of *Bifidobacterium bifidum* (NRRL1976, 1917 & KCCM 12096) and their co-culturing were undertaken in TPY medium containing oxgall or taurocholic acid. In the presence of taurocholic acid containing medium cholesterol uptake was 10% higher as compared to Oxgall containing medium in mixture after 24 and 32h, there was not much difference in reduction of cholesterol observed after next 4 to 8h of incubation. All cultures deconjugated 60 to 98% was detected by HPLC of the bile salts and 64 to 80% reduction in cholesterol respectively. Co-culturing only demonstrates the differences in the amount of bile salts deconjugation and cholesterol reduction. Small amount of cholesterol was found inside the cells indicating that assimilation phenomena exist but on a very lower level.

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C002

### Detection of *Escherichia coli* O157:H7, *Salmonella* spp., *Staphylococcus aureus* and *Listeria monocytogenes* in Kimchi by Multiplex Polymerase Chain Reaction (mPCR)

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We developed a mPCR assay for the simultaneous detection of *Escherichia coli* O157:H7, *Salmonella* spp., *Staphylococcus aureus* and *Listeria monocytogenes* in one tube by using species-specific primers targeted to the genes encoding the target gene. mPCR employed *E. coli* O157:H7-specific primer Stx2A, *Salmonella* spp.-specific primer Its, *Staphylococcus aureus*-specific primer Cap8A-B, and *Listeria monocytogenes* specific primer Hly and produced products of 553, 312, 405 and 210 bp, respectively. All PCR products were easily detected by agarose gel electrophoresis, and the sequences of specific-primers were assessed, respectively. The potential pathogenic bacteria in laboratory-prepared and four commercial selected kimchis were isolated and detected by the mPCR assay, respectively, and the results showed perfect correlation with the results of sequencing of primer products, respectively. The sensitivity of the assay was determined for purified pathogen DNA from four bacterial chromosomal DNA, respectively.

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C003

### A Novel Approach to the Synthesis of Insoluble Dye-Labeled Inulins as Chromogenic Substrates

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Inulin is a widespread polyfructan in plants, consisting of linear chains of  $\beta$ -(2,1)-linked fructofuranose molecules. The ordinary method for the synthesis of insoluble chromogenic substrate by using reactive dye and cross-linking reagent such as 1,4-butanediol diglycidyl ether is not applicable for inulin due to its low molecular weight. A novel approach was applied for the preparation of insoluble dye-labeled inulins. For this purpose methacrylated inulin (MA-Inulin) was synthesized by using glycidyl methacrylate and 4-(dimethylamino)-pyridine as catalyst. Aqueous solutions of MA-Inulin were converted into cross-linked hydrogels by free radical polymerization. Finally, inulin hydrogels were dyed by using reactive dyes and diversely colored insoluble cross-linking inulins were obtained. These dye-labeled inulins were degraded by inulinase from *Aspergillus niger* in a time- and concentration-dependent manner and they can be applied as chromogenic substrates for the assay of endonulinase.

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C004

### Enantioselective Production of Levofloxacin by a Microbial Lipase : Screening and Characterization of the Lipase from the Yeast *Yarrowia lipolytica* 180

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Levofloxacin, (*S*)-isomer of the (*R*),(*S*)-ofloxacin, is a new fluoroquinolone antibiotic. To screen strains producing a lipase that hydrolyze the (*R*),(*S*)-ofloxacin ester to levofloxacin, 151 isolates showing lipolytic activity on tributyrin agar plate were tested for the hydrolysis of ofloxacin ester. 3 strains among them were selected, and one of them, identified as the yeast *Yarrowia lipolytica* 180, was found to enantioselectively hydrolyze the (*S*)-enantiomer of the (*R*),(*S*)-ofloxacin ester. The enantioselective lipase-encoding gene was cloned by screening genomic library of *Y. lipolytica* 180 and the sequence was determined. The lipase gene consisted of 1431-bp nucleotides, encoding a polypeptide of 476 amino acids with a molecular mass of 52 kDa. The consensus sequence G-X<sub>1</sub>-S-X<sub>2</sub>-G in most serine lipases was well conserved. The catalytic apparatus involving the triad serine, aspartate, and histidine was also found in the protein sequence. The amino acids comparison showed a high similarity with the yeast lipases of type B1 carboxylesterase/lipase family.

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