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DNA microarray를 이용한 Whole-cell Biosensor 개발 Development of whole-cell based biosensor using DNA microarray

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The selection of promoters based on the DNA microarray analysis and the construction of cell-based biosensors were conducted to develop chemical- or stress-specific whole-cell biosensors in a high-throughput manner. Two superoxide-stress responsive bioluminescent E. coli biosensors (fpr'-lux/RFM443 and zwf'-lux/RFM443) and the other three DNA damaging responsive bioluminescent E. coli biosensors stress (sulA'-lux/RFM443, alkA'-lux/RFM443 and gltA'-lux/RFM443) constructed by fusing promoters to luxCDABE operon for the genes fpr, zwf, sulA, alkA and gltA) respectively. These genes were selected based on the gene expression analysis of genome-wide DNA microarray data for E. coli RFM443 treated with a superoxide radical generating agent, paraquat, or DNA damaging agents such as mitomycin C (MMC) and N-Methyl-N'-Nitro-N-Nitroso-Guanidine (MNNG). The fpr (126. 4 fold induction), and zwf (7.4 fold induction) genes were found to be highly upregulated with paraquat in DNA microarray experiments. The results

obtained from the bacterial biosensors showed that the fpr::luxCDABE and zwf::luxCDABE fusion bacteria were strongly induced specifically by superoxides generated by paraquat but failed to respond to  $H_2O_2$ , therefore, distinguishing oxidative stress caused by  $O_2-$  from  $H_2O_2$ . Also, sulA::luxCDABE, alkA::luxCDABE and gltA::luxCDABE fusion bacteria were shown to be strongly induced specifically by MMC and MNNG, respectively.