Expression and crystallization of Toxoflavin lyase(TflA) & TRP

Woo-Suk Jung, Du-kyo Jung, Sangkee Rhee, *Department of agricultural biotechnology, Seoul National University. Seoul, Korea.*
E-mail: srheesnu@snu.ac.kr

TRP (transthyretin-related protein) from soil bacterium *Bacillus subtilis* was suggested to be involved in the ureide pathway. In order to investigate its enzymatic role in the pathway, we have carried out structural study of TRP protein (121 amino acid). TRP gene from *Bacillus subtilis* was subcloned into the expression vector, pET15b and expressed as the His-tagged protein, followed by purification using immobilized metal affinity chromatography (IMAC). Crystals were formed within 6 days at concentration of 12 mg/ml with hanging drop.

TflA (222 amino acid) is an enzyme degrading a phytotoxin, toxoflavin. This toxin which is produced from *Burkholderia glumae* causes rice grain rot in rice nursery boxes. TflA gene subcloned into pET14b vector was overexpressed and purified by using IMAC and ion-exchange chromatography. Crystals of TflA were observed in 2M ammonium sulfate, 0.1M MES, pH7.2. Preliminary crystallographic data will be presented.

Keywords: TflA, TRP, enzyme mechanism