

Cloning and Expression of Hepatitis B Surface Antigen with Elastin-Like Polypeptides in *Pichia pastoris*

Hye Jin Kang, Yoon Mo Koo*

Department of Biological Engineering, ERC for Advanced Bioseparation
Technology,

Inha University, Incheon 402-751, Korea

TEL: +82-32-872-2679, FAX: +82-32-872-4046

Elastin-like polypeptides (ELPs) undergo a reversible phase transition upon an increase in temperature, forming hydrophobic aggregate. This thermally triggered phase transition allows for a simple and rapid means of purifying the fusion protein. Recovery of ELPs fusion protein was easily achieved by aggregation triggered either by raising temperature or by adding salt.

Hepatitis B virus 's' gene coding for surface antigen was cloned into *Pichia pastoris* transformation vector pPIC3.5 with and without ELPs tag, respectively. Transformants were analyzed for the integration of the transgene by colony PCR. Expression level was determined by RPHA (reversed passive hemagglutination). Western blot analysis confirmed the presence of 24 kDa and 33 kDa band specific to HBsAg and HBsAg-ELP[V-20], respectively.

References

1. Dan E. Meyer and Ashutosh Chilkoti, "Purification of recombinant proteins by fusion with thermally-responsive polypeptides"(1999), *Nature Biotechnology*, 17, 1112-1115.
2. Joan L. Cereghino and James M. Cregg, "Heterologous protein expression in the methylotrophic yeast *Pichia pastoris*"(2000), *FEMS Microbiology Reviews*, 24, 45-66.