Title: Purification and characterization of a lipase A from *Bacillus amyloliquefaciens*

Po-Shu Shih, Whei-Fen Wu

Department of Agricultural Chemistry, College of Bio-resource and Agriculture, National Taiwan University, Taipei, Taiwan, R.O.C.

We isolated a strain *Bacillus amyloliquefaciens* from the environment and select its lipase gene. Then, a lipase gene in pET21a, into *E.coli* and the expressing protein was produced by an induction with IPTG. Through the purification of the lipase protein (LipA) by the Nickle column method, its molecular weight is about 25kDa. The optimal temperature and pH value for LipA lipase activity is 35℃ and pH-10, respectively. Also, the LipA lipase is stable under 35℃ and pH value 9-10. The LipA lipase prefers *p*-nitrophenyl laurate as substrates. When the buffer contain a final concentration of 10 mM Zn^{2+}、Fe^{2+}、Cu^{2+}、Ca^{2+}, SDS、Triton-X-100 would strongly suppress the enzyme activity. However, 10% of DMSO and isopropanol would slightly enhance the enzyme activity. From the results of the thin-layer-chromatography (TLC), the degradation product of olive oil contain fatty acid and diglyceride. The degradation rate of olive oil after 96 h by *B. amyloliquefaciens*, transformant or LipA lipase protein was 38.8%、45.3%、53.5%, respectively; as for soybean oil it was about 35.5%、43%、62%, respectively. Taken together, our results revealed that LipA lipase has the potential application of biodiesel production, used-oil treatment and oil modification.

Whei-Fen Wu has completed her PhD from University of Iowa in USA and postdoctoral studies from National Institute of Health. She is an Associate Professor in NTU and she has published 25 papers in the professional Journal.