

Pattaraporn Thongthai 2009: Detection of *Staphylococcus aureus* in foods by dot-ELISA Technique using Polyclonal Antibody. Master of Science (Microbiology), Major Field: Microbiology, Department of Microbiology. Thesis Advisor: Associate Professor Patcharee Sunthornandh, Ph.D. 105 pages.

Staphylococcus aureus can cause many kinds of disease include food poisoning in raw and ready-to-eat foods. It can produce enzymes, toxins and contain protein A at the cell wall which is the surface antigen using for indicate the present of this bacteria in any samples. Detection of *Staph. aureus* by conventional microbiological method take much more time than immunological method. The immunological method by dot-ELISA was developed by using polyclonal antibody; produced from rabbit injected by whole cell of *Staph. aureus* and purified protein A. Antibodies (IgG) were purified by using affinity chromatography and purity were characterized by SDS-PAGE. Amount of purified IgG from whole cell of *Staph. aureus* and purified protein A were 0.58 mg/ml and 18.96 mg/ml, respectively. Both of purified IgG were used to detect *Staph. aureus* by dot-ELISA at the optimum concentration of 50 µg/ml and the optimum conjugate of 1:5,000, which could detect *Staph. aureus* at 10^3 CFU/ml.

The cross reaction with other *Staphylococcus* and enteric bacteria were eliminated by using Baird-Parker broth for enrich and selection of *Staph. aureus* and followed by same condition of dot-ELISA. This technique can be detected *Staph. aureus* at 10^3 CFU/ml, the same result as mention before.

Dot-ELISA technique was used for detection of *Staph. aureus* in food sample by inoculated the food samples in selective Baird-Parker broth for 24 hours, then followed by dot-ELISA. It revealed the same result when compared with conventional method, but detection by dot-ELISA include cultured in selective broth took only 30 hours, shorter than conventional method which took about 5-6 days. This method was easy and less amount of reagent was used, high sensitivity and might be a good option for detection of *Staph. aureus* in food and water.

Student's signature

Thesis Advisor's signature

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