

TITLE: COLORIMETRIC TEST FOR DETECTION OF BACTERIOPHAGES SPECIFIC TO *Streptococcus thermophilus*

AUTHORS: BATALHA, L.S.¹; SILVA, J.M.¹; GONTIJO, M.T.P.²; ELLER, M.R.¹

INSTITUTION: ¹ UNIVERSIDADE FEDERAL DE VIÇOSA, VIÇOSA, MG (AV. PETER HENRY ROLFS, S/N - CAMPUS UNIVERSITÁRIO, CEP 36570-900, VIÇOSA – MG, BRAZIL) ² UNIVERSIDADE ESTADUAL DE CAMPINAS, CAMPINAS, SP (CIDADE UNIVERSITÁRIA ZEFERINO VAZ – BARÃO GERALDO, CEP 13083-970, CAMPINAS – SP, BRAZIL)

ABSTRACT:

The production of fermented dairy products involves the use of microorganisms such as *Streptococcus thermophilus*. However, infection of these bacteria by bacteriophages causes fermentation failures, leading to losses of yield and quality. The early detection of these agents represents an opportunity for control measures to be taken before there is damage to the quality of fermented products and, eventually, to the consumer's health. Aiming at the rapid and simplified detection of these bacteriophages in the raw material, we developed a biosensor using polydiacetylene (PDA) vesicles. Bacteriophages specific for *S. thermophilus* were propagated to 1.0×10^8 PFU.ml⁻¹, concentrated to 5.0×10^9 UFP.ml⁻¹, and purified by ultracentrifugation with cesium chloride gradient. Six active protein fractions were used for the immunization of BALB/c mice to produce polyclonal anti-phage antibodies. The immune sera were collected and purified against bacterial cells to remove anti-bacterial antibodies. PDA vesicles were prepared with 1,2-dimyristoyl-sn-glycero-3-phosphatidylcholine (DMPC) added of an anti-phage antibody suspension (final concentration 1.0 mg/mL). The solution was exposed to UV light for 30 min para a formação da cadeia de polímero alternada eno-ino resulting in a lavender colored solution. An aliquot of the phage suspension was added to the vesicle-antibody conjugate suspension, and after 15 min a color change to light pink was observed. The color change of the solution was due to a specific antigen-antibody immunoreaction on the vesicle surface, which altered the polymer chain. The application of PDA vesicles for the detection of bacteriophages in the dairy industry is an alternative that, in addition to preventing large economic losses caused by the action of these phages in the starter culture, also stands out for having desirable characteristics, such as its rapid detection capacity, practical, specific and reproducible.

Keywords: dairy, biosensing, PDA liposome, colorimetric sensor

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