



CHARACTERIZATION OF EDIBLE CASEINATE FILMS WITH ANTIMICROBIAL ACTIVITY FROM Streptococcus sp. CULTURE BROTHS

Bethsua Mendoza-Mendoza^{1,2}, Adriana-Inés Rodríguez-Hernández¹, Gloria Díaz-Ruiz³, Rafael Montiel-Duarte⁴, Apolonio Vargas-Torres¹, Norberto Chavarría-Hernández¹

¹Cuerpo Académico de Biotecnología Agroalimentaria. Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo. Tulancingo, Hidalgo, CP 43600 <u>norberto@uaeh.edu.mx</u>; ²Instituto Tecnológico Superior del Oriente del Estado de Hidalgo, Apan, Hidalgo, CP 43900; ³Departamento de Alimentos y Biotecnología, Facultad de Química, Universidad Nacional Autónoma de México, México DF, CP 04510; ⁴Laboratorio Nacional de Genómica para la Biodiversidad, CINVESTAV, Irapuato, Guanajuato, CP 36821

Key words: bacteriocins, food packaging, sustainability

Introduction. The food industry is interested in the availability of food packaging materials with additional functions like the antimicrobial one. This is the case of the biopolymer-based edible films incorporating the antimicrobial activity from microbial products like bacteriocins (1). The present work deals with the elaboration and characterization of caseinate based edible films with the inclusion of antimicrobial substances (AM) produced by *Streptococcus* sp., acid lactic bacterium (LAB) isolated from *pozol*, a Mexican fermented food.

Methods. AM substances were produced by Streptococcus sp. (Dra. C. Wacher, FQ, UNAM) in MRS. Then, the supernatant was recovered after centrifugation, treated and conserved at -20°C until use (2). Edible films. Sodium caseinate (4%) and glycerol (30% db) with different AM concentrations: 0, 0.5, 0.7, 0.9 and 1.25 mg_{AM}/g_{film}. Antimicrobial activity. Listeria monocytogenes was the indicator. It was recorded the listeria-colonial growth in Oxford agar in contact with the films, to estimate the colonial growth kinetic parameters: a) colony lag phase (λ_c , h) (time when the colonies were first observed). b) Maximum specific colony growth rate (μ_{c-max} , h^{-1}) which is the maximum value of the function $\mu_c = \frac{d(A/A_0)}{dt} \times \frac{1}{(A/A_0)}$ in the exponential growth region, where A is the colonial area and A₀ is the corresponding A value to a colony when it is first observed; t is time (h). c) The maximum listerial biomass that was accumulated within each monitored colony was considered to be proportional to the corresponding maximum (A/A₀) value recorded (i.e., (A/A₀)_{max}, dimensionless). Films were also characterised concerning the Water vapor permeability (WVP), Oxygen permeability (PO₂), and <u>Mechanical properties</u> (i.e., rupture stress, σ_{max} , Young modulus, EM, and deformation percentage, %E)

Results. Figure 1 exhibits the colonial growth of listeria; when it grew with no film, the lag phase, λ , was 19 h. The same occurred with the film without AM. In contrast, λ values for treatments T1 and T2 were 35 h, being 45 h for T3 whereas in T4 there was no growth during the 72 h that the experiment lasted. On the other hand, μ_{max} decreased when AM concentration increased. Values of μ_{max} for listeria growth with no film (0.77 h⁻¹) and film with no AM (0.74 h⁻¹) did not exhibit statistically differences.

Nonetheless, there were important differences with treatments T1 (0.16 h⁻¹), T2 (0.10 h⁻¹) and T3 (0.9 h⁻¹). Concerning the achieved maximum biomass concentration, both the maximum and minimum values for $(A/A_0)_{max}$ were 1593 and 3 (-) corresponding to no-film condition and T3 treatment, respectively (without considering T4, where there was no listeria growth). The $(A/A_0)_{max}$ value for film with no AM was notably higher than the corresponding ones for T1, T2 and T3, being (459.8/9.1)=50.5, (459.8/3.3)=139.3 and (459.8/3)=153.3 times, respectively.



Fig.1 Effects on *Listeria monocytogenes* growth in contact with sodium caseinate films within antimicrobial (AM) substances produced by *Streptococcus* sp. Key: ●, no film; ◇, film without AM; ▼, T1 (0.5 mg_{AM}/g_{lim}); □, T2 (0.7 mg_{AM}/g_{lim}), and ○, T3 (0.9 mg_{AM}/g_{lim}).

Conclusions. It was characterised the antimicrobial effects of edible caseinate films against *L. monocytogenes*, in terms of the effects on the listeria colony growth kinetics. The determination of all kinetic parameters (colony lag phase, λ_c ; maximum specific colony growth rate, $\mu_{c\text{-max}}$, and maximum biomass accumulation [proportional to $(A/A_0)_{\text{max}}$]) was useful to estimate the magnitude of the inhibitory effects against *L. monocytogenes*.

Acknowledgements. PROMEP 2012 *"Biotecnologías basadas en biomoléculas funcionales para el sector agroalimentario"* y PIFI-2012-13MSU0017T-04, UAEH.

References.

1. Cao-Hoang L, Chaine A, Grégoire L, Waché Y (2010). *Food Microbiol.* 27(7):940-944.

2. Mendoza-Mendoza B (2012). Caracterización de películas a base de caseinato de sodio, con la inclusión de sustancias antimicrobianas producidas por bacterias ácido lácticas aisladas de pozol. Tesis de Maestría. ICAp, UAEH. Hidalgo, México.