



CHITOSAN BASED FILMS AS AN ACTIVE PACKAGING OF MEXICAN FRESH CHEESE “QUESO RANCHERO” FOR PRESERVATION AND *Listeria monocytogenes* INHIBITION

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Introduction. The “queso ranchero” is fresh cheese of soft paste not pressed and produced in several states of Mexico. The short shelf life (up to 10 days) and its perishability are due to its high moisture content (45 and 55%), low salt content and near neutral pH (6.0–6.5) [1]. *Listeria monocytogenes* outbreak has been associated to consumption of fresh cheese [2]. Antimicrobial packaging using organic acids is an alternative for extending the shelf life of this cheese and reduction, inhibition or retard of microbial growth.

The aim of this study was the evaluation of four chitosan based films in grafting and blends with organic acids: chitosan (Q), chitosan-poly(lactic acid) (QAL), chitosan-co-citric (QC) and chitosan-co-citric acid-hydroxypropylmethyl cellulose (QH).

Methods. Chitosan was prepared by heterogeneous deacetylation from chitin extracted by biological method [3]. Biopolymers were characterized moisture content, ash, residual protein, soluble in acetic acid, degree of acetylation and molecular weight. The syntheses of QAL, QC and QH were carried out and characterized on their weight, thickness, water vapor permeability (WVP), mechanical properties and infrared spectra. Pouches were prepared by cutting and heating sealed chitosan film and wrapped commercial cheese ranchero (Celaya Guanajuato, Mexico). Cheeses were stored at 4°C with 20% relative humidity. Weight loss percentage (%W), moisture content, pH, total titratable acidity (ATT) and microbiological analysis aerobic mesophilic bacteria, molds and yeast were determined in cheeses. Further the WVP of QAL films were modified by using soy protein isolate (SPI) or rice flour (RS). The modified WVP films were employed for a second assay with cheeses inoculated with *L. monocytogenes* and stored at the above conditions. Cheeses were analyzed on pH, ATT and bacterial count using *Listeria* selective agar Oxford (Difco, France).

Results. The cross-linking reaction among organic acids and hydroxy propyl methyl cellulose into chitosan produced less rigid and fragile films than the solely chitosan. QAL and QH films showed higher values of WVP. The films of Q and QC presented smaller values of WVP (Table 1). It was observed that QAL showed higher inhibition for aerobic mesophilic bacteria, molds and yeasts (Fig. 1a, b) than other films. It is worth notice that the counts obtained for QAL were even lower than that determined at the initial. The cheeses packaged with QAL, QC and QH showed a decrease in pH after seven days storage might be caused by migration of the acidic components of the film. Due to none of the films prevented losses of moisture in the cheeses during storage, modification of WVP of QAL

films was carried out. The lowest WVP was determined in QAL employing 0.25% (w/v) of SPI (Table 2).

Table 1. Properties of Q, QAL, QC and QH films

Film	Thickness (mm)	WVP (gmm/m ² hkPa)	Young's Modulus extension (MPa)
Q	0.069±0.009	6.96X10 ⁻⁴ ±1.58X10 ⁻⁴	2226±561.8
QAL	0.071±0.012	1.53X10 ⁻³ ±5.22X10 ⁻⁴	151.2±34.0
QC	0.105±0.022	5.94X10 ⁻⁴ ±1.63X10 ⁻⁴	327.2±97.2
QH	0.151±0.024	1.46X10 ⁻³ ±2.29X10 ⁻⁴	222.3±25.1

Mean of measurements and their standard deviations. WVP was determined at gradient of HR 22/75 and 4°C±2.

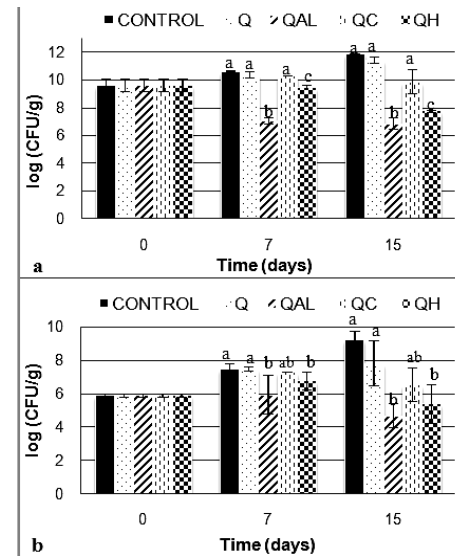


Fig. 1 Colonies enumeration of mesophilic microorganism (a) and fungi (b) of cheeses packed in bags made with chitosan based films, Mean of three measurements and their standard deviations. Histograms with the same letter did not display significant differences (P<0.05)

Table 2. WVP of QAL with addition of SPI and RS.

QAL/	WVP (g mm/ m ² hkPa)	QAL/	WVP (g mm/ m ² hkPa)
QAL	1.53X10 ⁻³ ±5.22X10 ⁻⁴ ^b	0.25%SPI	8.43X10 ⁻⁴ ±1.07X10 ⁻⁴ ^{a*}
2%RS	1.32X10 ⁻³ ±3.28X10 ⁻³ ^b	0.38%SPI	8.75X10 ⁻⁴ ±4.81X10 ⁻⁵ ^a
4%RS	2.33X10 ⁻³ ±1.76X10 ⁻⁴ ^c	0.50%SPI	9.26X10 ⁻⁴ ±1.94X10 ⁻⁴ ^a
0.14%SPI	1.13X10 ⁻³ ±4.29X10 ⁻³ ^b	0.75%SPI	1.18X10 ⁻³ ±6.93X10 ⁻³ ^b

Mean of measurements and their standard deviations. WVP was determined at gradient of HR 22/75 and 4°C±2. Values with the same letter did not display significant differences (P<0.05)

L. monocytogenes counts were significantly lower in QAL/SPI wrapped cheeses than the control (inoculated in LDPE bags) and those without wraps.

Conclusions. Microbial growth inhibition was observed in fresh cheeses packaged in QAL pouches. The addition of SPI in QAL resulted in a less permeable material that inhibited the *L. monocytogenes* growth.

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