



Title	Genomic insights into high exopolysaccharideproducing dairy starter bacterium <i>Streptococcus thermophilus</i> ASCC 1275
Author(s)	Shah, N; Wu, Q; Tun, HM
Citation	Joint Annual Meeting (JAM) of the American Dairy Science Association (ADSA), the American Society of Animal Science (ASAS), and the Canadian Society of Animal Science (CSAS), Kansas City, Missouri, USA, 20-24 July. In the Journal of Animal Science, 2014, v. 92 n. E-Suppl. 2, p. 164, abstract no. 0332
Issued Date	2014
URL	http://hdl.handle.net/10722/205072
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a pathogenic bacterium, *Escherichia coli* were investigated using Fourier transform infrared spectroscopy (FT-IR). A critical NaCl concentration that inhibited the growth of *E. coli* without significantly affecting the growth of probiotic bacteria was determined by monitoring cell growth and FT-IR spectra. To evaluate the effect of substitution of NaCl with KCl, substitution was performed at critical total salt concentration at varying concentrations (0%, 25%, 50%, 75% and 100% KCl). Furthermore, the effects of varying NaCl concentrations on viability, membrane integrity and metabolic activity of these probiotic bacteria were studied using conventional technique and flow cytometry. The findings revealed that in *Lb. acidophilus* degree of proteolysis increased with higher salt concentration at pH 5.0 and 6.0 and ACE-inhibitory activity was highest at pH 5.0 at all salt concentrations. Fourier transform infrared spectroscopy results demonstrated significant shifts occurring in amide-I and amide-III regions when *Lb. acidophilus* was subjected to varying salt concentrations. Further, the conventional technique revealed that 2.5% was the critical level of NaCl to inhibit the growth of *E. coli* without significantly affecting the growth of most probiotic bacteria. The FT-IR analysis also highlighted the changes that occurred mainly in amide regions on increasing NaCl concentration from 2.5 to 3% in most bacteria. The findings suggest that 50% substitution of NaCl with KCl at 2.5% total salt could inhibit *E. coli*, without affecting the probiotic bacteria. Lastly, the observations from conventional culture technique were compared with the findings from flow cytometric analysis on metabolic activities of the cells and it was revealed that there was a correlation between culturability and dye extrusion ability of *Lb. casei* and *B. longum*. However, a certain population of *Lb. acidophilus* was viable as per the plate count method but the efflux activity was compromised. The metabolic activity of *Lb. casei* was found to be highest among the three probiotic bacteria.

Key Words: FTIR, flow cytometry, probiotic bacteria

0332 Genomic insights into high exopolysaccharide-producing dairy starter bacterium *Streptococcus thermophilus* ASCC 1275.

N. Shah, Q. Wu* and H. M. Tun, *The University of Hong Kong, Hong Kong.*

Streptococcus thermophilus ASCC 1275 (ST 1275) is a typical dairy starter bacterium and produces the highest known amount (~1,000 mg/L) of exopolysaccharide (EPS) in milk within this species. This organism produces both capsular and ropy EPS and possesses textural modifying properties for yogurt and cheese. In this study, de novo shotgun paired-end pyrosequencing was applied to complete the whole genome of ST 1275. The genome size of ST 1275, a plasmid-free bacterium, was ~1.85 Mbp with an average GC content of 39.1%. A novel *eps* gene cluster for EPS assembly containing two-pair genes of *epsC-epsD* for determining the chain length of EPS was found in ST 1275 genome, which confirms that ST 1275 produces two types of EPSs as found in our previous studies. Compared with

other sequenced *S. thermophilus* strains, ST 1275 possessed the lowest numbers of 5 rRNA operons and 55 tRNAs suggesting that this organism may have a more effective protein synthesis machinery. The highest number of four separate CRISPR (clustered regularly interspaced short palindromic repeats)-Cas (CRISPR-associated) loci was found in ST 1275 genome indicating that this organism may have a better adaptive immunity against various bacteriophage infections. Further analysis including carbohydrate utilization, effective proteolytic system, sophisticated stress response systems and defense systems in ST 1275 was performed to provide genomic insights into its adaptation to milk and as a cell factory for EPS production during milk fermentation. The elucidation of ST 1275 genome makes this organism as a model dairy starter bacterium for the research of high EPS yield and capsular/ropy EPS producer from the species of *S. thermophilus*.

Key Words: genome sequencing; EPS biosynthesis; *Streptococcus thermophilus*

0333 Effectiveness of pulsed light treatment on the inactivation of pathogenic and spoilage bacteria on cheese surface.

J. Proulx*, L. Hsu¹, B. Miller¹, G. Sullivan¹, K. Paradis², and C. I. Moraru¹, ¹Cornell University, Ithaca, NY, ²McGill University, Montreal, QC, Canada.

Cheese products are susceptible to post-processing cross-contamination that can lead to both food safety issues and significant losses due to spoilage. Pulsed Light (PL) treatment, consisting of short, high-energy light pulses, could represent a solution to address this issue since it is a nondestructive technology that can effectively inactivate microorganisms on surfaces. This study examined the effectiveness of PL on the inactivation of the spoilage microorganism *P. fluorescens* and the pathogen surrogates *E. coli* ATCC 25922 and *L. innocua*. The effect of inoculum level, cheese surface topography, and the presence of clear polyethylene packaging were evaluated in a full factorial experimental design. The challenge microorganisms were grown to stationary phase: *P. fluorescens* 1150 was grown at 30°C in tryptic soy broth (TSB) while *E. coli* ATCC 25922 and *L. innocua* FSL C2-008 were grown at 37°C in TSB and brain heart infusion (BHI), respectively. White cheddar and processed cheese, chosen for their different surface topography, were cut into 2.5 cm × 5 cm slices. The samples were then spot inoculated using ten droplets of 10 µL per slice, resulting in an initial concentration of either 5 or 7 log CFU/slice. Inoculated samples were dried overnight at 4°C. For treatments through packaging, sterile UV-transparent low-density polyethylene packaging was placed on top of the inoculated cheese samples immediately before the PL treatment. Cheese samples were then exposed to PL doses of 1.1 to 13.2 J/cm². PL-treated samples were stomached for 2 min in Butterfield Phosphate Buffer, the extract then plated on selective media and survivors enumerated by standard plate