Factors influencing cell count of a probiotic Lactobacillus crispatus strain. Kevin Bourzac*, Ann Bernard, Dr. M. E. Sanders, and Dr. Rafael Jimenez-Flores, California Polytechnic State University, San Luis Obispo, CA.

The probiotic bacteria Lactobacillus crispatus strain HP101 was derived from a fermented dairy product in eastern Europe. Commercialization of this strain has been a challenge due to its poor growth characteristics in standard Lactobacillus media. The objective of this work was to analyze factors influencing growth and cell count in different media to improve the commercialization potential of this strain. Compared to the successful industrial Lactobacillus acidophilus strain NCFM, HP101 exhibits 2-3 log cycle fewer CFU/ml in MRS media although optical density measurements are equivalent. HP101 cells were also observed to have different morphology than NCFM when grown in MRS. NCFM cells were short, compact rods where HP101 were long and spindly (often associated with unhealthy cells). A live/dead staining procedure also indicated that a high percentage of HP101 cells were damaged or dead when grown on MRS for 24hrs. Growth in milk completely reversed the negative HP101 growth parameters. Cell morphology, cell health (as determined by the live/dead stain), and final cell count became equivalent to that of NCFM. However, since cells are not easily recovered from milk media, it is unsuitable for industry use. Therefore, applicable media adjustments which mimicked results from growth in milk were determined. Our experimental methods included growth curve analysis, colony counts, live/dead stain analysis, peptide analysis of media and 2-D gel electrophoresis. Milk permeate supplemented with ≥0.3 % casein resulted in CFU and cell morphology that mimicked milk-grown cells.

Key Words: Probiotics, Permeate, Lactobacillus