High-throughput differentiation of *Streptococcus thermophilus* and *Lactobacillus* spp. strains used in Swiss cheese production by infrared microspectroscopy

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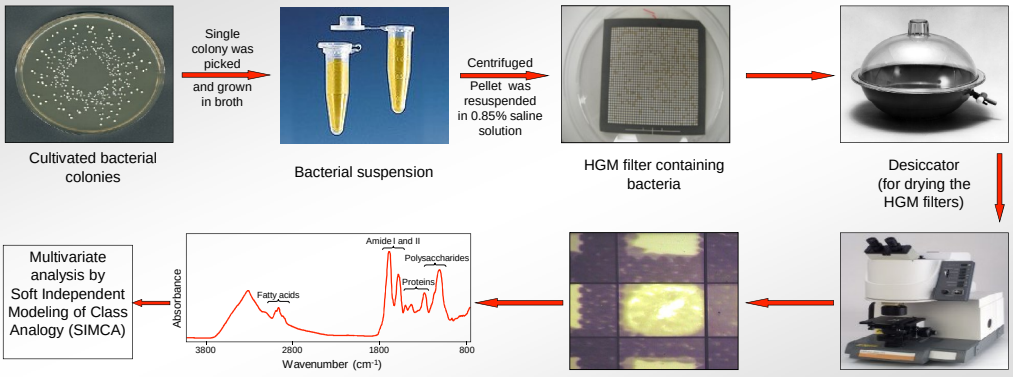
ABSTRACT

Swiss cheese production in US was 310 million lbs (3.2% of total cheese production) in 2008. Ohio is the leading Swiss cheese producer in the US, supplying over 42% of the total US production. Strain variations of starter cultures have an impact on final quality and hence the price of Swiss cheese. A rapid and cost-effective method for identification and differentiation of starter cultures at strain level could help maintain uniform quality of Swiss cheese. The study was focused on developing a high-throughput technique to identify and differentiate *Streptococcus thermophilus* and *Lactobacillus* spp. strains involved in Swiss cheese production using the combination of hydrophobic grid membrane (HGM) filters and Fourier transform infrared (FTIR) microspectroscopy. Strains of *S. thermophilus* (14) and *Lactobacillus* spp. (15) were analyzed. Aliquots (2 µL) of each strain were transferred onto a grid of the HGM filter vacuum dried in a desiccator and analyzed using FTIR microspectroscopy. The procedure was repeated on six different days to account for growth variability. The spectra were analyzed by soft independent modeling of class analogy. The pattern recognition analysis showed tight clustering at the strain level for models developed for *S. thermophilus* and *Lactobacillus* spp. The models showed unique patterns in the spectral region from 1150 to 1000 cm^{-1} for the major discrimination in the *S. thermophilus* and *Lactobacillus* spp. that can be attributed to differences C-O stretching of polysaccharides. This method could be an effective tool to identify and monitor activity of dairy cultures.

INTRODUCTION

The acceptability of Swiss cheese depends largely on the flavor formed during ripening. Production of Swiss cheese employs starter cultures composed of *Streptococcus thermophilus*, *Lactobacillus helveticus* (or *Lactobacillus delbrueckii*), and *Propionibacterium* freudenreichii [1]. In addition, non-starter lactic acid bacteria contribute to the flavor of cheeses [2], being considered the last uncontrolled element in the modern cheese manufacture [3]. Thus, several species and strains are involved in the formation of flavor compounds. Therefore, an understanding of role of cheese microflora and their interactions in the formation of flavor compounds is necessary to control cheese flavor development. Time and money can be saved through identification of these microflora whereby quality control of cheese could be established early in the manufacturing process. Common methods used in the identification of starter cultures are time-consuming and may give misleading results if applied to unknown bacteria [4,5]. Infrared spectroscopy is a novel technology for the rapid, cost-effective, sensitive and high-throughput analysis of food components. Recent advances in Fourier transform infrared (FTIR) spectroscopy, sample preparation methods like HGM filters and multivariate techniques have shown the potential for the successful discrimination, classification and identification of microorganisms. **This research is focused on developing a method that enables rapid identification and differentiation of Swiss cheese starter culture *S. thermophilus* and *Lactobacillus* spp. strains using FTIR microspectroscopy.**

METHODS



RESULTS

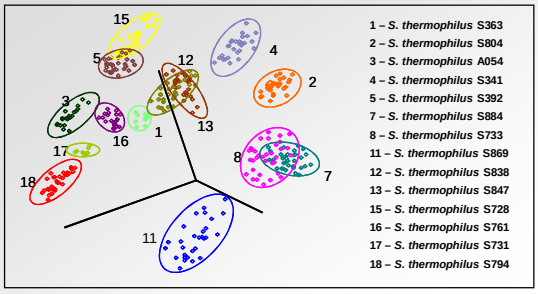


Figure 1. SIMCA classification model of 14 *S. thermophilus* strains obtained by FTIR microspectroscopy

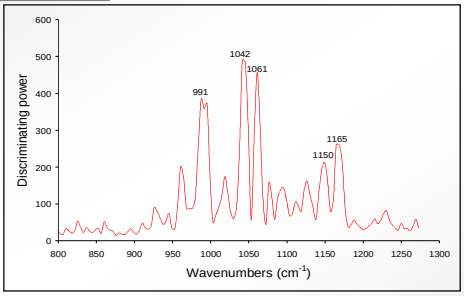


Figure 2. Discriminating power plot for the *S. thermophilus* SIMCA model

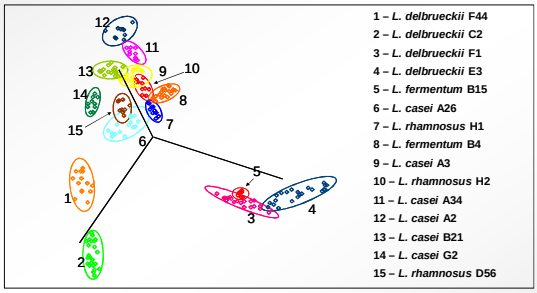


Figure 3. SIMCA classification model of 15 *Lactobacillus* spp. strains obtained by FTIR microspectroscopy

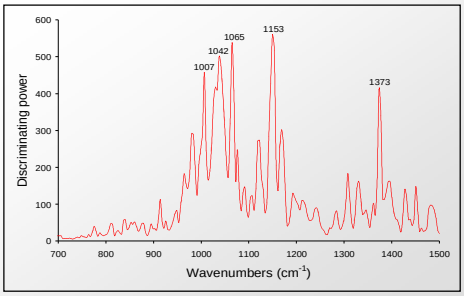


Figure 4. Discriminating power plot for the *Lactobacillus* spp. SIMCA model

DISCUSSION

- Classification models were developed to discriminate several strains of *S. thermophilus* and *Lactobacillus spp.* used in Swiss cheese production.
- Figure 1 and 3 shows the SIMCA classification model of *S. thermophilus* and *Lactobacillus spp.* respectively.
- Figure 2 and 4 shows the important wavenumbers that contributed in discrimination of *S. thermophilus* and *Lactobacillus spp.* classification models respectively.
- Spectral region 1150 to 1000 cm^{-1}, that can be attributed to C-O stretching of polysaccharides, was important for discrimination in the models.
- HGM filters enhanced the spectral signal by confining the bacterial colonies and increasing colony density.
- Generation of FTIR spectral libraries for starter and non-starter cultures will allow for rapid and cost-efficient monitoring of cultures at strain level during cheese manufacture and storage.

CONCLUSIONS

An infrared spectroscopy method was developed for the differentiation of Swiss cheese starter and non-starter cultures at the strains level. This technique could be a potential tool for identification of dairy cultures at strain level and monitoring its activity during cheese ripening and storage.

REFERENCES

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