USING NIR HYPERSPECTRAL IMAGING FOR DETECTION AND DIFFERENTIATION OF PATHOGENIC BACTERIA

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The lack of suitably accurate and rapid methods for pathogen detection in the food industry could lead to delays in product distribution or contaminated products entering the market. In March 2016 there were two *Listeria monocytogenes* scares in the USA where two shelf sensitive products, smoked salmon and packaged breakfast sandwiches, were distributed before tests could be completed and were recalled. Conventional culturing methods for pathogen detection and species differentiation is known to be time consuming, thus the aim of this work is to use NIR hyperspectral imaging a rapid tool for foodborne pathogen detection.

Four commonly encountered foodborne pathogens (*Salmonella enteritidis, Staphylococcus aureus, Bacillus cereus* and *Escherichia coli*), as well as non-pathogenic *Staphylococcus epidermidis* were studied All bacteria were imaged on Luria Bertani (LB) agar in glass petri dishes after 20 h incubation at 37 °C using a SisuChema short wave infrared (SWIR) camera, in the range 1000 to 2500 nm. Standard normal variate (SNV) correction and the Savitzky-Golay technique (2nd derivative, 3rd order polynomial; 25 point smoothing) was applied to wavelengths 1103 to 2471 nm. Principal component analysis (PCA) was applied to mean-centered data calculating three principal components (PCs). Score plots and score images were used interactively for removal of all irrelevant pixels and to explore the data for any clustering.

Chemical differences were evident, along PC1 (58.1 % SS) *B. cereus* (Gram positive) could be distinguished from *E. coli* and *S. enteritidis* (Gram negative), and *E. coli* from *S. enteritidis* in the direction of PC2 (7.75 % SS). *B. cereus* and *S. aureus* were separated from *S. epidermidis* along PC1 (37.5 % SS). The loading line plots and mean spectra showed that the main chemical contributors permitting distinctions between bacteria were variances in amino acids, carbohydrate and teichoic acid content. Teichoic acid consists of either glycerol or ribitol units [1]. The peak at 1405 nm (O-H stretch, ROH) was only present in loading lines where Gram positive bacteria were present, leading to the assumption that this peak represented teichoic acid. Partial least squares discriminant analysis (PLS-DA) models were used to confirm the PCA data. The best predictions were made for the identification of *B. cereus* and the two *Staphylococcus* species, where results ranged from 82.0-99.96% correctly predicted pixels.

With exploratory techniques, it was possible to distinguish between similarly coloured colonies, Gram positive and Gram negative bacteria and pathogenic and non-pathogenic species using NIR hyperspectral imaging.

References

[1] W.W. Navarre, O. Schneewind, Surface proteins of gram-positive bacteria and mechanisms of their targeting to the cell wall envelope, Microbiol. Mol. Biol. Rev., 63 (1999) 174-229.