Fatty Acid Methyl Ester (FAME)

Overview

Fatty acids have long been molecules of environmental, biomedical, agricultural, and industrial importance. The analysis or identification of bacteria based on the ability to differentiate one type of microorganism from another by using fatty acid content and distribution is well known.

Fatty acid analysis using Gas Chromatography (GC) has been a challenge because of their high molecular weight and low volatility. The conversion of the fatty acid to the fatty acid methyl ester (FAME) is performed to increase volatility: A method of extracting, methylating, and analyzing fatty acid content has been available for some years using a bench top commercial GC. The advantages of a miniaturized GC system with microfabricated device elements over a traditional GC are low power requirements, low cost, hand-held, and lightweight.

Chemistry Overview

Lipids are water-insoluble biomolecules of cells that have high solubility in non-polar organic solvents. Lipids have many biological roles: they serve as fuel molecules, as highly concentrated energy stores, and as components of membranes. We are interested in the lipids that are components of membranes. There are three kinds of membrane lipids, phospholipids, glycolipids, and cholesterol.

Glycolipids and phospholipids are composed of fatty acids chains that are connected to a glycerol backbone. The fatty acid chains usually contain an even number of carbon atoms in a linear fashion (the 16- and 18-carbon chains are the most common). Fatty acids can be saturated or unsaturated (containing one or more double bonds).

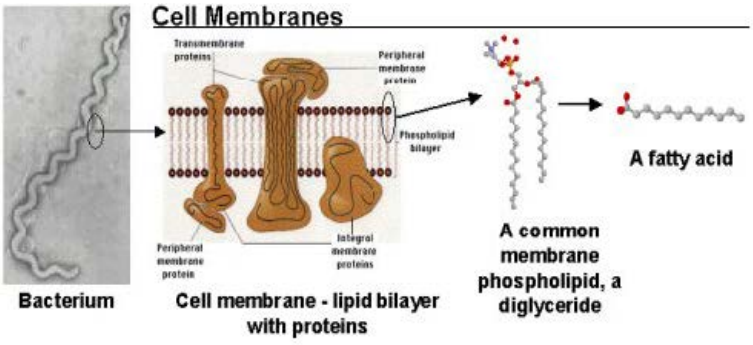


Figure 1. Cell membranes.

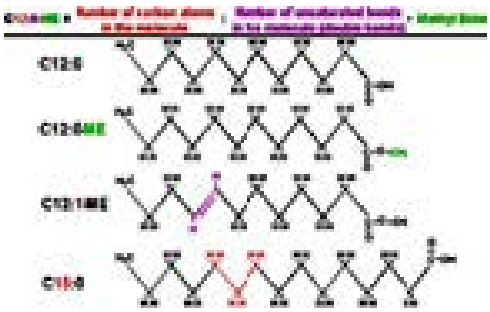


Figure 2. Biochemical reasoning.

Analytical Systems

A hand-held analysis system is being developed using miniaturized pyrolysis/GC instrumentation as shown in schematic below. This system will be based on the microfabricated stages already developed for MicroChemLab at Sandia National Laboratories. See MicroChemLab for additional information on the microfabricated stages.

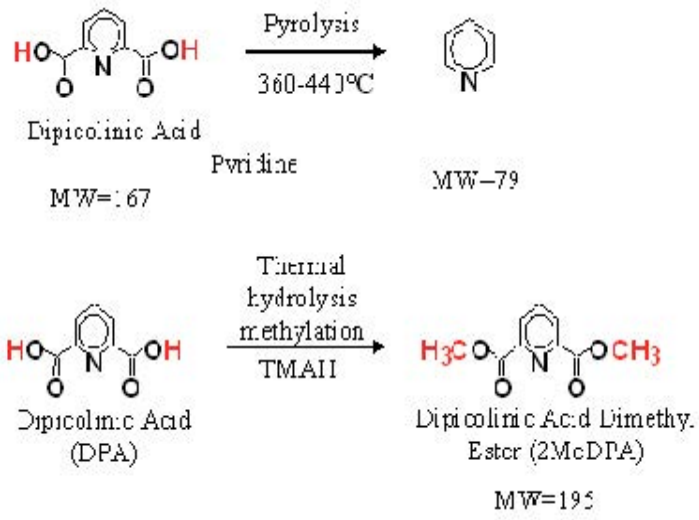


Figure 3. Structures, nomenclature and reactions.

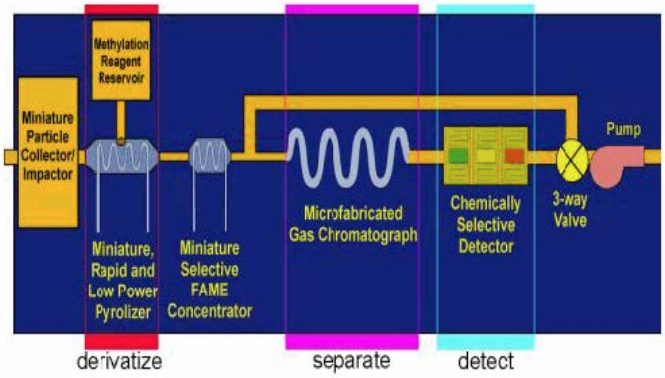


Figure 4. Cartoon schematic to describe the incorporation of biodetection using FAME pyrolysis into the *MicroChemLab* fixture. The additional components are those necessary to derivatize and pyrolyze the membrane fatty acids. It is envisioned that analysis of FAMEs will be accomplished using a micro gas chromatographic (GC) column for separation and a SAW (surface acoustic wave) for the detection of FAMEs *(see technical info of MicroChemLab for additional information)*.

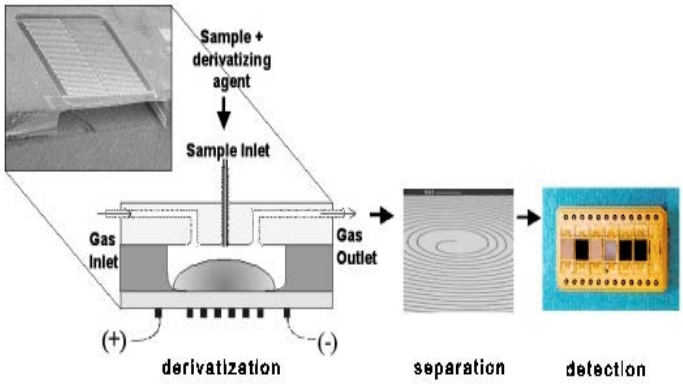


Figure 5. Schematic showing more details of the pyrolysis fixture (this is the location of where derivatization and pyrolysis of the fatty acids will occur).

Micropyrolyzer has been shown to convert whole cell bacteria into their FAME signature pattern with less than 1 W (GC/MS detection).

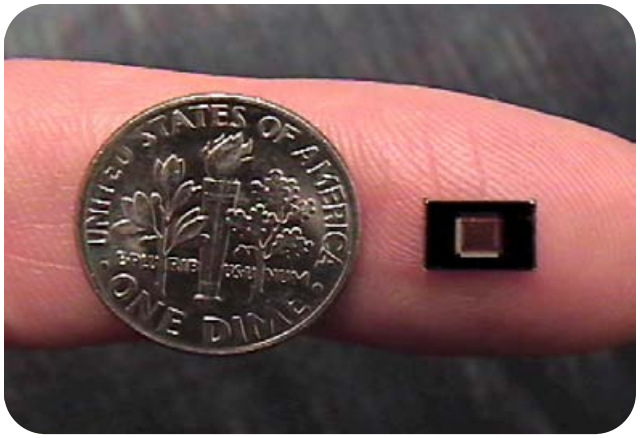


Figure 6. Micropyrolyzer.

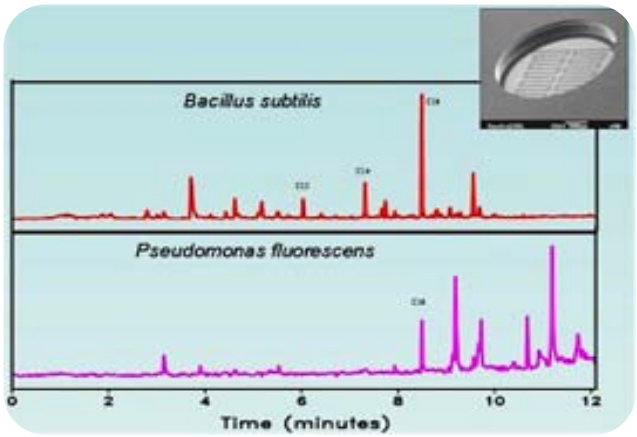
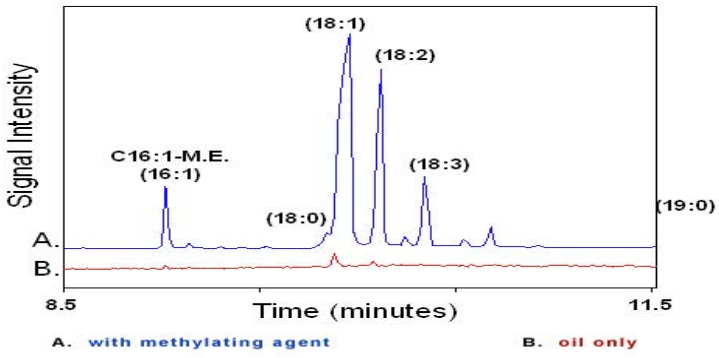


Figure 7.

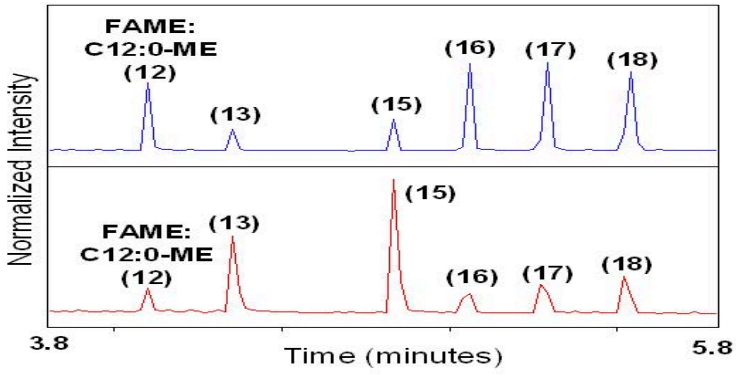
Performance

We have performed experiments to determine whether microfabricated components could facilitate a pyrolysis/methylation reaction and therefore demonstrate the potential for a hand-held FAME sensor. Pyrolysis/methylation of an edible oil has been demonstrated and FAMEs have been produced by pyrolysis/methylation of whole cell bacteria using the micro-pyrolyzer.



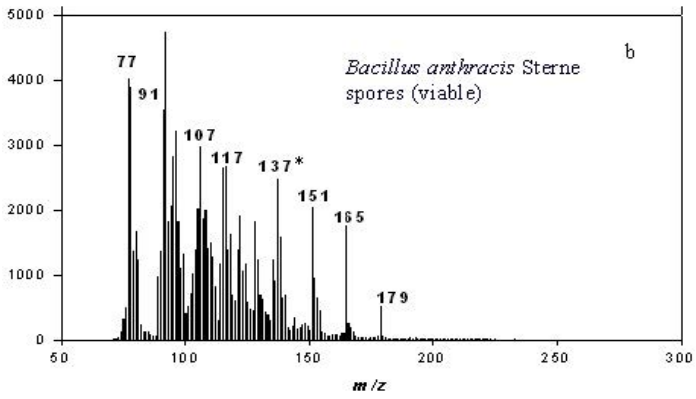
This figure illustrates the increase in volatility that happens to a fatty acid upon methylation. This figure also demonstrates that methylation does occur in our pyrolysis fixture.

Figure 8. Pyrolysis of an edible oil.



This figure demonstrates that in our fixture we are able to achieve effective separation and detection of pyrolysis products. The figure also demonstrates the ability to differentiate bacteria based on signal pattern from two different Bacillus species.

Figure 9. Pyrolysis of whole cell bacteria.



Thermal hydrolysis methylation of B. anthracis spores.

Figure 10. Mass spectra.

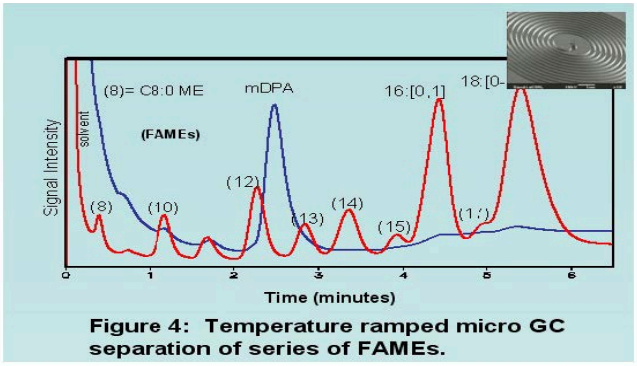


Figure 4: Temperature ramped micro GC separation of series of FAMEs.

FAMEs and mDPA can be separated using microfabricated columns, although more resolution is needed to separate saturated and unsaturated fatty acids.

Figure 11. Micropyrolysis results.

Sandia's Goal and Results

Utilize unique microfabrication facilities / skills to reduce size, weight, power of existing concepts for improvements in field portability and performance.

A prototype system incorporating the µ-pyrolyzer, µ-GC column, and miniature IMS has been assembled.

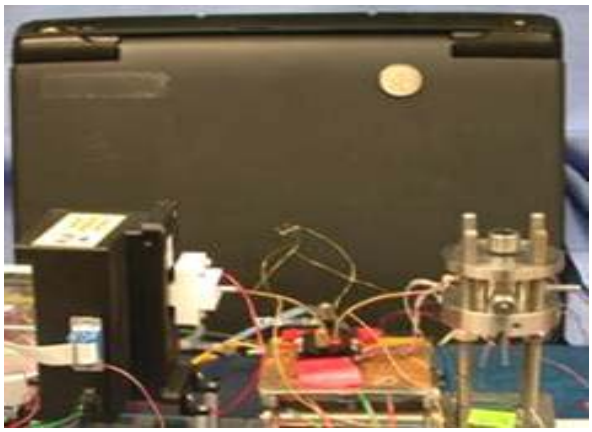


Figure 12. SNL system.

For additional information or questions, please email us at MGA@sandia.gov.

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