The Gas-liquid chromatography in the detection of bacteria fatty acids

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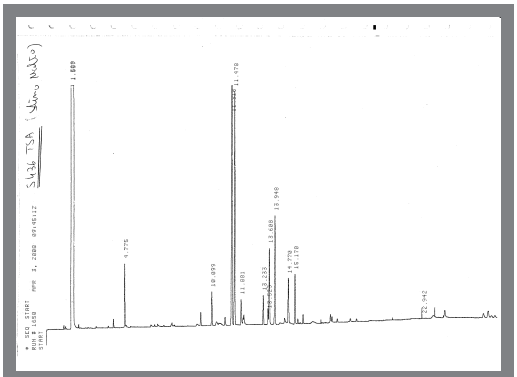
Introduction

Although most characteristics, such as shape or biochemical parameters, are essential to study a bacterium, it is sometimes necessary to look at fatty acids present in its membrane. Following a chemical break of the cell wall and preparation of the sample, it is possible to use gas-liquid chromatography in order to separate these fatty acids.
Thus, the fatty acid quantity can be determinated in accordance with the medium on which the bacterium had grown.
A large number of chromatograms allow the experimenter to read into the results with several statistical tools.
Then, in terms of acid secretion, according to the nutrients provided, additional information about bacteria appear.
Many studies have been done on acids produced by the metabolism of glucose by bacteria. Studies of acids within the membrane are more infrequent.
Thus, this method is unusual in a laboratory of medical analysis, as it constitutes a part of biological research on the properties of the bacterium.



Fatty Acids Chromatogram of Stenotrophomonas maltophilia cultured on TSA agar.

Work description

Different species of bacteria (Acinetobacter baumannii, Stenotrophomonas maltophilia, Pseudomonas aeruginosa) were isolated in different culture media (TSA agar, CSB agar and Drigalski medium). After the preparation was made, in order to detect fatty acids, the solution was injected into the chromatograph. Due to the retention time, a peak on the graph is associated with one acid.
The experiment was done using a column (HP 1909 J 202) with a diameter of 0.33 µm and a length of 25 m on a CPG Hewlett Packard 5890 Series II. The detection system was a FID detector. This separation was performed with a temperature gradient of 170°C to 270°C.
The results were analysed using statistical tools (average, standard deviation, etc.) with a focus on different topics.
First, the best agar for the vast development of a specific bacterium can be found within the area of the different peaks. It appears to be CSB agar for Acinetobacter baumannii and Stenotrophomonas maltophilia, and Drigalski agar for Pseudomonas aeruginosa.
Secondly, in order to generate a big quantity of a specific acid, it is possible to find the combination of specific bacteria and specific agar. Moreover, this argument can be used to show which medium of culture is homogeneous (i.e. which gives the same quantity of each acid for one bacteria). In this study it appears to be Stenotrophomonas maltophilia growing on Drigalski agar.
Another interest that can be put brought to attention, regarding the analysis of fatty acids present in the membrane of bacteria: as each type of cell has a specific chromatographic profile for a specific culture medium, the chromatograms can give the experimenter the identity of an unknown micro-organism.
Usually, this identification procedure is performed with fatty acids produced by the metabolism of glucose. Then, results are more certain and more accurate. By comparing them to literature data, the name and the family can be discovered.

Discussion

The gas-liquid chromatography is a very useful method for analysis. In the case of bacteriology, it provides the biologist with considerable information, which doesn't necessarily appear with other techniques.
Some results given by the chromatograms can be interpreted directly. Others must be combined with biochemical tests to give identification. This constitutes a weakness of that type of analytical science.

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