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METHODS OF ANALYSIS OF LISTERIA MONOCYTOGENES (Abstract of the lecture given by Mrs Rollier at CECALAIT's Annual General Meeting)

Since the beginning of 1999, public attention in France has been focused on safety problems in food associated to *Listeria monocytogenes*.

Listeriosis is a rather rare disease (225 cases in France in 1997), mostly associated to food. Most healthy people show no or influenza-like symptoms without any consequence. But, for target people, complications may be severe, usually meningitis or septicemia, which are often fatal. The main target populations are elderly people, new-borns, immunocompromised persons, pregnant women. Infections during pregnancy may result in spontaneous abortion (2nd/3rd trimester) or stillbirth. Listeriosis is caused by *L. monocytogenes*, of which the infective dose is unknown but is believed to vary with the strain, susceptibility of the victim and contamination conditions. This bacterium is ubiquitous and very hardy.

In any case, dairy regulation, eg EC directive 92/46 specifies the absence of *Listeria monocytogenes* usually in 25 g of product (cheese, milk...). So the detection of this bacterium is of great importance in food hygiene and faster, easier and more specific methods are expected.

UBIQUITOUS AND VERY HARDY BACTERIUM

It is a Gram-positive bacterium, motile at 22°C, but not at 37°C, aero-anaerobic. It is quite hardy and resists remarkably well to chemicals and severe temperature, pH or salinity conditions as is shown in table 1 in La Lettre de CECALAIT.

Therefore, it can be found everywhere : air, soil, plants, food, healthy carriers....In food, incidence data collected France showed (1993 to 1995 data) a rather high incidence in meat products, then decreasing from seafood to vegetables, pastries and dairy products. However, in these latter products, contamination is often specific to soft cheeses, more heavily contaminated than other foods.

The epidemiologic survey since the 80s shows that *L. monocytogenes* has been associated to very different kinds of foods, including cabbage, raw-meat sausages, raw and smoked fish, and dairy products such as soft cheeses, pasteurized milk...It also shows that serotype 4b is usually responsible. In the last years, the duration and the number of cases in epidemics has tended to decrease. This is certainly linked to the more frequent official and self controls during food processing and marketing and to the improvement in methods of analysis.

EVOLUTION OF THE METHODS

It was necessary to evaluate their evolution as the classical methods are very complex and time consuming. Table 2 in La Lettre de CECALAIT shows the evolution in the methods of detection and enumeration of *L. monocytogenes*.

Reference methods, such as ISO 11290-1 and -2 (1997 and 1998) and IDF 143A :1995 also improved, as is shown in table 2. But, they still remain much too time consuming for routine

purposes in dairy laboratories. So a lot of alternative, quick methods appeared as shown in table 3 in La Lettre de CECALAIT. Most of them are validated (AFNOR, AOAC). However, as the enrichment steps still remains necessary, it is difficult to take less than 48h.

As CECALAIT has been organizing ringtests for *Listeria* detection since 1993, we could observe how laboratories followed the evolution of methods. Figure 1 in La Lettre de CECALAIT thus compares enrichment, isolation media and identification techniques used in the first ringtest (March 1993) and in the latter one (April 1999).

However, even quick method users often use reference methods for confirmation. So the conclusions of the European Project of validation of ISO 11290-1 & -2 methods of detection and enumeration of *L. monocytogenes* are particularly interesting.