

# Microbial update

## faecal indicators

by Dr Roy Betts, Head of the Microbiology Dept, Campden-BRI, Chipping Campden, Gloucestershire, GL55 6LD, UK. Produced as a service to the food industry by Oxoid, part of Thermo Fisher Scientific.

Microbiological testing of foods and ingredients is done to give some indication of the microbiological status of those materials. It is important that before setting out to do testing, companies understand what they want to get out of a test plan, and what they will do with the results they receive.

The type of tests done may include those for general microbial levels for example Total Viable Count (TVC), tests for potential spoilage micro-organisms such as Pseudomonads, lactic acid bacteria or yeasts and moulds, and tests for specific pathogens like salmonella or Listeria monocytogenes.

All of these tests give the food producer an indication of the microbiological quality of a food product, and must be viewed with a great understanding of what each microbial group are responsible for within a product.

As an example, tests for spoilage organisms are done to gain confidence that a food will not spoil within its designated shelf life, TVC can give a general indication of total viable microflora and may be very useful if trended over time, to show improving (or perhaps problematic) production hygiene, or perhaps issues occurring seasonally or after a change of supplier.

Pathogen tests are there as a part of the verification that HACCP plans are operating correctly, and that pathogens are not detected within the analysed portions of the foods tested.

There is, however, one other set of micro-organisms that the food producer may wish to consider testing for, in order to help them get a view of the quality of a food and the food production environment, these are the indicator organisms.

### History of indicators

Reading past publications, or even considering some producer/customer specifications, we may see the frequent use of the term 'indicator organisms' or even sometimes 'index organisms' but very few descriptions of what such tests are trying to 'indicate' or provide an index of.

These terms are frequently misunderstood



**Enterococci are not exclusively of faecal origin and exist in soil, some animals and on plants, with no adverse hygienic significance.**

and this can result in the data from such tests being misused. In order to undertake testing for 'indicator organisms' we must understand what these tests are trying to 'indicate', and what any positive results may mean.

In order to understand the use of tests for indicator organisms, we must first understand the problems of testing foods for enteric food pathogens.

Many foods will be tested for enteric pathogens, such as salmonella. Such tests are not difficult for a food microbiology laboratory to perform, but they are more complex than simple plating tests used for TVC, and the results are only available after many days of incubation.

Additionally, enteric pathogens are rarely present in foods and if they are, they will usually be at very low levels, and will be non-homogeneously distributed throughout a food product.

This means that even in the unlikely event that a food contains an enteric pathogen, it may be very difficult to detect, even though it would put the consumer at risk.

Indicator organisms provide one test that

can help to overcome these issues with enteric pathogen testing.

By choosing the correct microbial group, it is possible to use a fairly simple and rapid test that will give a possible indication that food has been produced or stored under less than optimal, hygienic conditions.

Two important things must, however, be kept in mind:

- Firstly the presence of levels of indicator organisms does not automatically mean that a food also contains enteric pathogens.

- Secondly the absence of indicator organisms does not mean that a food does not contain enteric pathogens.

Indicators are a rough guide that can be used to quickly determine the general hygienic status of a food production, storage, distribution chain, and thus the hygienic status of the food itself.

### E. coli and coliforms

The use of E. coli as an indicator of potential faecal contamination was first proposed in the 1890s. E. coli detection methods were and are simple, and as the organism is found in some considerable numbers in faecal matter, it is more easily detected than enteric pathogens, which if present, are likely to be present in much lower numbers.

Historically, however, E. coli was quite difficult to detect due to the presence of large numbers of other enteric bacteria which interfered with detection methods.

The move to consider detection and enumeration of coliforms provided a way around this issue. The coliforms can generally be defined as organisms within the genera, escherichia, klebsiella, citrobacter, and enterobacter, however as the group are generally defined by their ability to ferment lactose, and as such other members of the enterobacteriaceae can sometimes fall into this group.

The coliforms are established indicators of faecal pollution of water and their use as indicators of hygienic conditions in the food industry is an extension of this.

Coliforms grow well at 37°C, however in the past there has been a designation of coliforms able to grow at higher temperatures (44-45°C) as 'faecal coliforms'.

Tests for faecal coliforms are now not often used with food testing. Testing for coliforms is still done in some parts of the world, however, there is a general move towards replacing the coliform test, with tests for enterobacteriaceae.

The reasons for this are multiple, but include the fact that coliforms are not a defined 'group' within microbial taxonomy (the enterobacteriaceae are a defined family), and that organisms within the coliform group can be 'naturally' found in some raw foods types.

A good example would be on fresh produce, where members of the Genus *Enterobacter* are found as common natural commensal organisms on plants, and have no link with faecal contamination or unhygienic conditions.

*E. coli*, is still widely used as a test within the food industry. Many different types of *E. coli* are recognised by microbiologists, most are not considered pathogenic, but there are some types that are known pathogens, for example the verocytotoxin producing *E. coli* (VTEC) group that includes *E. coli* O157:H7 and *E. coli* O104 (the cause of the large food poisoning outbreak linked to sprouted seeds in Germany in 2011).

General counts of *E. coli* done on foods tend to be used as 'indicators' of potential unhygienic conditions within food production, and usually microbiological criteria require this organism to be absent or present at only very low levels in most types of food.

Low levels of *E. coli* are not usually considered in themselves to constitute a food poisoning risk. If food producers wish to look for known pathogenic types of *E. coli*, then very specific types of test have to be done, these look only for the pathogenic types.

The tests are complex and infrequently done, and any isolation of these potential pathogenic organisms results in a requirement for very special laboratory containment requirements.

As noted before, is important for food producers to understand that the presence

of general indicator *E. coli* does not mean that pathogens or even pathogenic *E. coli* are present in a food, and likewise the absence of *E. coli* does not mean that pathogenic types are absent.

## Faecal enterococci

The genus enterococcus contains a number of species that are linked with faecal contamination including *E. faecalis* and *E. faecium*.

Tests for faecal enterococci are commonly done in food testing laboratories, but again enterococci are not exclusively of faecal origin and exist in soil, some animals and on plants, with no adverse hygienic significance.

It is known, however, that the enterococci are slightly more resistant to heat and some stresses than coliforms and in some conditions are better indicators than the latter group.

## Significance of indicators

The significance of 'indicator organisms' in food is not easy to define. Whilst it is known that *E. coli*, coliforms and faecal enterococci can be found in animal faecal material, they can also originate from other sources, with some coliforms in particular, being part of the natural microflora of plants.

Results must therefore be treated with care. High levels of these organisms are not desirable in foods, however it is hard to find any published scientific articles, which clearly link the presence of 'indicators' with the presence of enteric pathogens.

Likewise, we should guard against any consideration that the absence of indicators automatically means that there are no pathogens present in a food.

Testing for indicator organisms provides a relatively simple and quick way to gain some understanding of the general hygienic status of the food production environment.

Generally, food producers that would like to employ indicator organism tests need to generate trending graphs of these groups to fully understand, and take appropriate actions based on results.

By generating trending graphs, food producers will understand the general levels of these organisms usually present in their product, and thus will be able to see any gradual or sudden changes in levels of these groups.

This will allow the producer to consider whether levels are static, or gradually changing, and perhaps to link any change with specific actions (changes in raw materials, processing conditions or cleaning and hygiene inputs).

Overall, indicator organisms provide a useful range of tests for food producers. The testing methods tend to be simple and give results in a fairly short time period (particularly if compared to pathogen tests). The results of these tests must, however, be treated with caution.

Whilst elevated levels of any indicator group or organism are not desirable within foods and must be investigated, they do not automatically give any direct information about the presence of specific pathogens, and as noted previously, absence of these groups does not mean that pathogens can be considered to be absent.

Only specific testing for pathogenic organisms can give this information. The indicator groups do, however, prove useful in trend analysis.

Allowing food producers to monitor changing microbiological trends over time and link these trends to specific actions that they may have taken.

This may allow a better control of food production conditions, and a better microbiological quality of food products. ■

✉ val.stroud@thermofisher.com

***E. coli*, coliforms and faecal enterococci can be found in animal faecal material, but can also originate from other sources.**



## References

- The Use of Indicators and Surrogate microorganisms for the Evaluation of Pathogens in Fresh and Fresh-Cut Produce <http://www.fda.gov/Food/ScienceResearch/ResearchAreas/SafePracticesforFoodProcesses/ucm091372.htm>. Accessed 28/01/2012.
- Pierson, M. D., Zink, D. L. and Smoot, L.M. (2007) Indicator Microorganisms and Microbiological Criteria. Chapter 4 (p.69-85) In: Food Microbiology, Fundamentals and Frontiers. 3rd Edn. Eds: M. P. Doyle and L. R. Beuchat. ASM Press 2007.
- Photographs copyright Shutterstock.