

# Occurrence of *Escherichia coli* O157 in foods from animal origin in Belgium since 1997

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## Introduction

Enterohemorrhagic *Escherichia coli*, implicated in aqueous diarrhoea, hemorrhagic colitis or hemolytic uremic syndrome (HUS), is a serious health problem in various countries. In Belgium, all cases are sporadic and no outbreak has been detected. In USA and UK, the consumption of poorly cooked minced beef is an essential risk factor. The knowledge of the rate and the level of contamination are essential for an efficient risk assessment program.

## Material and Methods

In 1997, a large number of matrixes have been investigated for the presence of *E. coli* O157. These matrixes were: cattle and pork (carcasses, livers, retail cuts and minced meat); broiler (carcasses, livers and breasts); layer, turkey and rabbit (carcasses). All samples were 600cm<sup>2</sup> or 25g.

In 1998, priority was given to cattle for further investigations. In order to increase the number of samples, 6000 carcasses of beef, 1% of the annual beef's slaughtering in Belgium, were swabbed. In four zones of 100cm<sup>2</sup> (10x10cm each for a total sample size of 400cm<sup>2</sup>) These carcasses were collected from the 40 greatest Belgian companies and were pooled 5 by 5. If a pool was detected positive, each individual sample was assessed.

In 1999, on account of interpretative difficulties, pooling system was given up and 1984 carcasses were swabbed in four zones of 400cm<sup>2</sup> (20x20cm each for a total sample size of 1600cm<sup>2</sup>) and processed individually. Minced meat was also investigated by the analyse of 974 samples directly taken in the butchers.

In 1997, seeing that it was no normalised method, the one that was used was a double method with a pre-enrichment in buffered peptone water during 7h at 37°C followed by an immuno-magnetic separation (Dynabeads anti-*E. coli* O157, Dynal, Norway). The first half of the concentrate was then plated onto sorbitol MacConkey agar incubated during 18h at 42°C, the other half was cultured into 10ml of modified *E. coli* broth with novobiocin and incubated 18h at 37°C. After the incubation period, an ELISA test was performed (Vidas ECO, bio-Mérieux, France) and in case of positive result, agglutination test (*E. coli* latex test, Oxoid, United Kingdom) was performed on several colonies and then tested for biochemical characterisation (Api20E, bio-Mérieux, France). All isolates were then tested for virulence factors.

Since 1998, it exists an official method from the Ministry of Public Health (SP-VG M001) which is based on a short pre-enrichment in modified trypticase soy broth with novobiocin incubated 6-7h at 42°C. The incubation was followed by the enrichment of 1ml in 9ml of MacConkey broth with cefixime and tellurite, incubated 18h at 37°C and then tested with an ELISA test as described above (Vidas ECO, bio-Mérieux, France). From the positive broth, an immuno-magnetic separation was performed and the concentrate was plated onto SMAC and SMAC with cefixime and tellurite. Further detection and confirmation were realised as describe above.

## Results and discussion

In 1997, no *E. coli* O 157 was detected but some matrixes have given a lot of false positive results with the ELISA test which suggest us to change the used protocol.

In 1998, Enterohemorrhagic *E. coli* O157 were detected in 10 of the 1202 pooled carcasses but it was nearly impossible to find which individual sample was positive and the estimated rate of contamination was supposed to be 0,2% (10 positive results from 1202 pools of 5 carcasses). New changes in the sampling strategy were necessary.

In 1999, 25 carcasses (1,3%) and 1 minced meat (0,1%) were found positive.

## Conclusion

- These studies show that more and more *E. coli* O157 are detected on beef carcasses. The increase of prevalence may be due to either a better detection level of the method or to a real increase of the presence of this pathogen. Nevertheless, one of the companies investigated in 1999 show an epidemic peak in the month of august which lead to a contamination rate of 13% (data not shown).
- Hygienic measures should be taken in slaughterhouses in which highly faecal contaminated carcasses should be reserved for cooked meat.
- The Belgian diet contains raw or undercooked beef meat, which could endanger the risk population, children and elderly, if they eat contaminated meat, even at a low rate of contamination.
- Because of the repercussions for public health, surveillance must be maintained in Belgium and should also assess the contamination rate of meat (for raw consumption) in butchers.

## References

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- Piérard D. and al., Virulence factors of verocytotoxin-producing *Escherichia coli* isolated from raw meats, Appl. And Environ. Microbiol.; 1997; Vol 63; 11; 4585-4587.