

Evaluation of media and test kits for the detection and isolation of *Escherichia coli* O157 from cattle faeces

A.V. Tutenel, L. De Zutter and J. Van Hoof

Department of Veterinary Food Inspection and Public Health, Faculty of Veterinary Medicine, RUG, Salisburylaan 133, B-9840, Belgium

Presented at the symposium: "Escherichia coli: friend and foe" at the University of York (UK), 13th - 16th of July 1999.

ABSTRACT

Four strains of enterohemorrhagic *E. coli* O157:H7 (one reference strain, one human strain, one strain from meat and one strain from carcasses) were used to inoculate feces from cattle. Each strain was added to 25 g of sample at a level of 5 to 14 cfu. Three selective enrichment media were used: (1) buffered peptone water (BPw), (2) modified tryptone soya broth with 1% novobiocine (mTSB-n) and (3) mTSB-n followed by cefixime-tellurite MacConkey (CT-MAC). Five different test kits were applied to these media: (1) Dynabeads® anti *E. coli* O157 (Dynal); (2) Captive® O157 (Lab M); (3) Vidas® *E. coli* O157 ECO (Biomérieux); (4) Vidas® *E. coli* O157 ICE (Biomérieux) and (5) Transia Plate® *E. coli* O157 (Diffchamb). These kits were tested after 6 h and/or 24 h of incubation. Plating on CT-SMAC plates occurred direct and after using the kits 1, 2 and 5.

This study showed that isolation of *E. coli* O157 was very good by using the Dynal Dynabeads® after 6h of incubation in BPw by 37°C. For Captive®, the results depended on the strain tested. The Transia Plate® O157 was very useful for the detection of *E. coli* after 22h incubation. Successful confirmation was only obtained using an immunomagnetic separation after incubation in BPw and CT-MAC. Vidas® ECO yielded less positive results than the test kits 1, 2 and 5. With Vidas® ICE, not all Vidas® ECO positive samples could be confirmed.

Introduction

Escherichia coli O157:H7 is a foodborne pathogen and can cause haemorrhagic colitis, haemolytic uraemic syndrome and thrombocytopenia.

Most outbreaks are associated with the consumption of raw meat and raw meat products. It is believed that cattle is the main reservoir for this pathogen.

Since the first outbreak of *E. coli* O157:H7 in America in 1982, lots of different isolation and detection kits and enrichment media are developed.

Isolation and detection can occur by: (1) chromogenic and/or fluorogenic media, (2) biochemical test kits, (3) electrical, (4) ELISA, (5) immunochromatography, (6) immunocapture, (7) latex agglutination, (8) nucleic acid hybridization probe, and (9) PCR

The aim of this work was to compare different detection and isolation media and test kits for the isolation of *E. coli* O157:H7 from faeces of cattle.

The different **isolation and detection kits** used in this recourse are:

(1) Dynabeads[®] anti *E. coli* O157 (Dynal); (2) Captive[®] O157 (Lab M); (3) Vidas[®] *E. coli* O157 ECO (Biomérieux); (4) Vidas[®] *E. coli* O157 ICE (Biomérieux) and (5) Transia Plate[®] *E. coli* O157 (Diffchamb). These kits were tested after 6 h and/or 24 h of incubation.

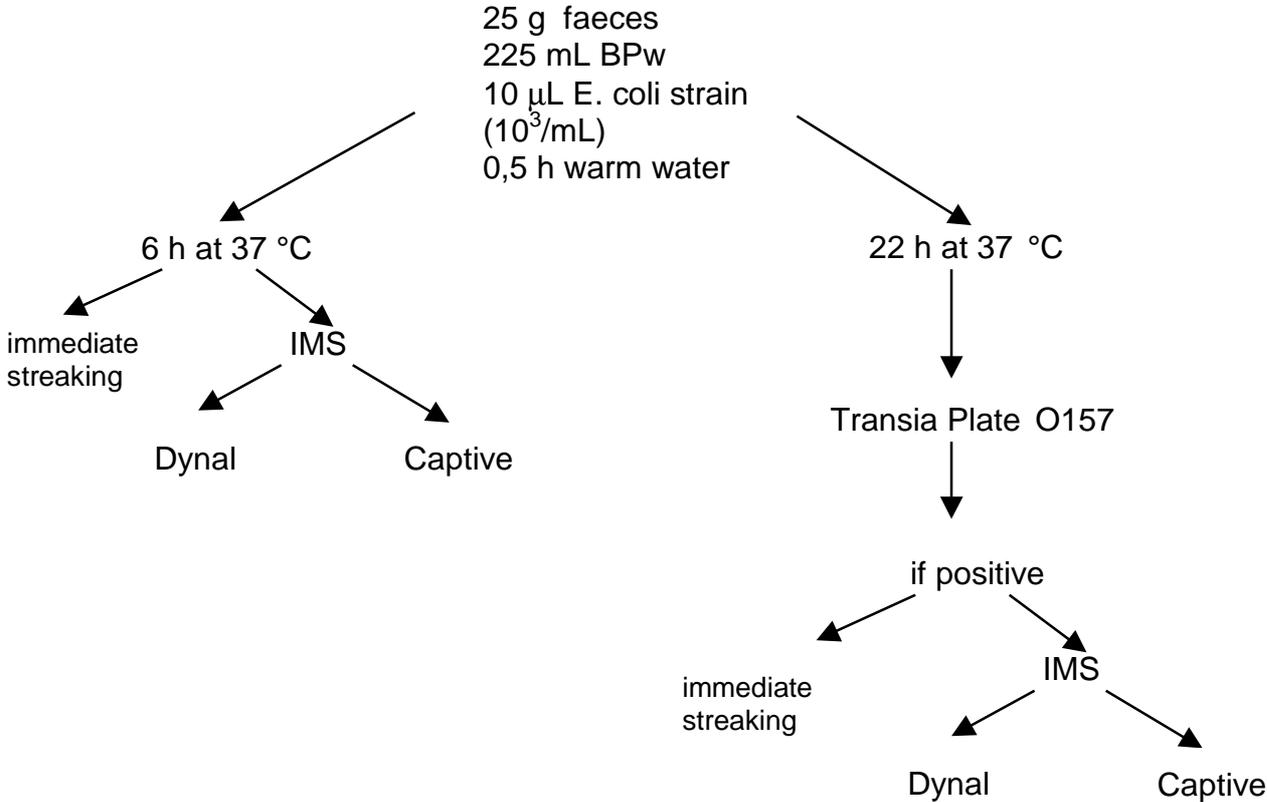
We used four different ***E. coli* O157 strains** to test the different kits: (1) LMG 15068, a reference strain isolated from hamburgers; (2) VUB EH1, the first strain isolated from human; (3) B032, isolated from carcasses; (4) 17762/1, isolated from minced beef.

Enrichment occurred in (1) BPw, (2) mTSBn and (3) CT-Mac.

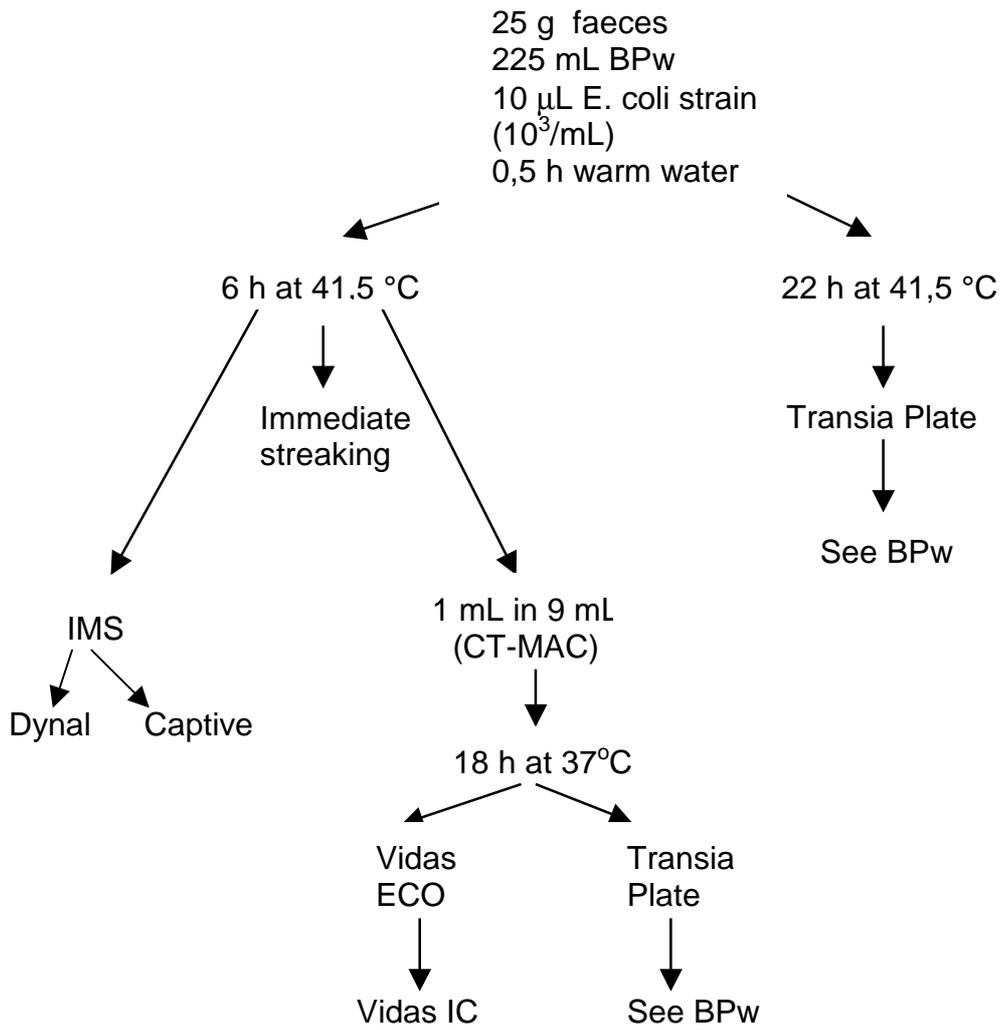
Everything was **streaked** onto CT-SMAC plates.

Materials and Methods

(1) Incubation in BPw



(2) Incubation in mTSBn



Results

Four different strains were tested on five different faeces samples.

enrichment time strain	6 h					
	1	2	3	4	T	%
Immediate streaking						
BPw	4	3	5	5	17	85
mTSBn	2	1	1	1	5	25
Dynal						
BPw	5	5	5	5	20	100
mTSBn	3	2	5	5	15	75
Captive						
BPw	5	5	3	0	13	65
mTSBn	2	0	2	0	4	20

enrichment time strain	22 h					
	1	2	3	4	T	%
Immediate streaking						
BPw	4	3	3	4	14	70
mTSBn	4	2	1	1	8	40
After Dynal						
BPw	4	4	5	5	18	90
mTSBn	5	4	1	2	12	60
After Captive						
BPw	5	5	5	5	20	100
mTSBn	5	5	1	2	13	65
Transia Plate						
BPw	5	4	5	5	19	95
mTSBn	5	5	5	5	20	100
CT-MAC						
Dynal	5	5	3	5	18	90
ECO	5	5	4	1	15	75
ICE	4	3	4	0	11	55
Transia Plate	5	5	5	5	20	100
Captive	5	5	4	5	19	95

Discussion

(A) after 6 h incubation

- all positive samples were found back using Dynabeads
- less positive samples were found back using mTSBn

(B) after 22 h incubation

- enrichment in BPw: Captive gave a 100% result
- enrichment in mTSBn: Transia Plate gave a 100% result
- enrichment in CT-Mac: Transia plate gave a 100% result
- less positive results yielded with Vidas

General conclusion

- (1) immediate streaking gave never a 100% result
- (2) after 6 h incubation, using BPw as enrichment broth gives a 100 % using Dynabeads
- (3) after 22 h incubation, using mTSBn as enrichment broth gives a 100% result using the Transia Plate
- (4) successful isolation of *E. coli* after the Transia Plate was only obtained using an IMS
- (5) Vidas ECO yielded less positive results comparing with the other techniques
- (6) Vidas ICE could not confirm all Vidas ECO positive samples