Letters to the Editor

Is enrichment culture necessary for isolating Campylobacter jejuni from faeces?

There has been debate in your columns previously as to the necessity of including an enrichment stage when culturing human faecal samples for “thermophilic” Campylobacters.1–3 Between May and October 1985 19 of the 21 microbiology laboratories in Wales participated in a study under the auspices of the Welsh Standing Specialist Advisory Group in Microbiology to investigate this need.

Each laboratory used its own selective technique for both primary isolation and subculture after enrichment. Enrichment was performed as described previously.4 Only two of the participating laboratories used identical cultural techniques.

A total of 16 837 samples were cultured. One thousand and forty four (6.2%, range from 1.8–11.5%) yielded Campylobacters on direct culture and a further 166 after enrichment, representing an overall increase in yield on enrichment of 15.9%. The increase varied considerably between laboratories, from 3.2% in one to 85.7% in another. The laboratory which obtained the highest increase used commercially supplied ready poured plates for selective isolation. This observation led to a subsequent study comparing a ready poured medium with an identical freshly prepared one. Seven positive isolates were detected on the freshly prepared medium. Two of these failed to grow on the ready poured medium, and the remaining five grew less profusely.

Clearly, many variations in methodology exist in our region and this may be reflected nationwide. Furthermore, our results suggest that the different selective techniques are not all equally efficient. While enrichment may not be required for specimens cultured by optimal methods it would seem to be necessary in many cases in practice.

Doubt still exists as to the optimal selective technique for the isolation of Campylobacters from human faecal samples. Various workers have reported promising results with blood free media.6–8 Bolton et al have also been reassessing filtration methods.9 Until this question is resolved our experience suggests that microbiologists should continue to assess the need for an enrichment stage in the context of their own laboratories.

Before this investigation was undertaken just three laboratories in Wales routinely used an enrichment procedure while two others used it only on selected samples: as a result of the study a further 11 laboratories introduced it into their laboratory routine.

CD RIBEIRO*
DC FITZGERALD†

* Public Health Laboratory, University Hospital of Wales, Heath Park, Cardiff.
† Microbiology Department, Llandough Hospital, Penarth, Near Cardiff.

Participating laboratories (Welsh Standing Specialist Advisory Group in Microbiology)

Royal Infirmary, Cardiff; University Hospital of Wales, Cardiff; Public Health Laboratory, Cardiff; Llandough Hospital, Penarth; East Glamorgan Hospital, Near Pontypridd; Bridgend General Hospital; Caerphilly District Miners Hospital; Prince Charles Hospital, Merthyr Tydfil; Singleton Hospital, Swansea; Neath General Hospital; Morriston Hospital, Swansea; Withybush Hospital, Haverfordwest; Bronglas Hospital, Aberystwyth; Ysbyty Gwynedd, Bangor; Ysbyty Glan Clwyd, Near Rhyll; Maelor Hospital, Wrexham; Public Health Laboratory, Swansea; Nevill Hall Hospital, Abergavenny; Llanelli General Hospital.

False positive results in the Toxoreagent test for Toxoplasma gondii in immunocompetent patients

The latex agglutination test (Toxoreagent, Eiken Chemical Company) is used in many laboratories to screen serum samples for antibodies to Toxoplasma gondii and has been claimed to give results comparable with those of the dye test when assessed qualitatively.1 The dye test is recognised as the serological reference test for toxoplasmosis. As a toxoplasma reference centre we are aware of positive results being reported in the latex agglutination test which cannot be confirmed by the dye test. It has also been reported that false latex agglutination test results may be associated with a rise in cytomegalovirus (CMV) specific IgM in heart transplant recipients.2

We looked for false positive latex agglutination test results in sera from 552 non-immunocompromised patients with various clinical symptoms considered to be compatible with toxoplasmosis but no evidence that the immune system was depressed. Fifty four samples were referred as latex agglutination test positive but were negative when run in the dye test (titre < 8). The latex agglutination test reaction was confirmed in only five (9.3%) sera, while two (22.2%) sera reacted with unsensitised latex. A further 498 sera were tested in the latex agglutination test only after they had been shown to be dye test negative. Two of these samples gave a false positive latex agglutination test result, an incidence of 0.4%, although two further sera reacted with unsensitised latex. In total, serum from seven patients gave false positive latex agglutination test results, with titres of 32 to 64. Of these sera, four (57.1%) reacted in the CMV-IgM ELISA.3 None of the serum samples reacting in the CMV IgM ELISA had T. gondii specific IgM when tested by ELISA.4

These data indicate that the false positive results previously found in heart transplant patients with acute CMV infection can also be found in other non-immunocompromised patients. In transplant recipients the reaction seems to be IgM mediated but not associated directly with CMV specific IgM nor rheumatoid factor.5

More than 49 of 54 (90%) of the sera submitted as being latex positive (= > 32) but found to be dye test negative were negative when retested in the latex agglutination test at the reference centre. The data suggest that there is some variation between laboratories in the way that this assay is performed and read. Adsorption with unsensitised latex will reduce but not remove the

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problem, although unsensitised latex is not provided in the kit. Positive latex results should be confirmed in the dye test and current CMV infection excluded in those patients whose serum samples give false positive results in the latex agglutination test. This is important as clinical manifestations of both CMV and toxoplasmosis have many similar features.

Further work to identify the antigens recognised by sera giving false positive reactions is in hand, using the Western blot technique.

AH BALFOUR
ES PRESTAGE
I NOEL
Regional Public Laboratory,
Bridle Path, York Road,
Leeds LS15 7TR.

TG WREGHITT*  
*Clinical Microbiology and 
Public Health Laboratory, 
Addenbrooke's Hospital, 
Cambridge CB2 2QW.

References
1 Balfour AH, Fleck DG, Hughes HPA, Sharp D. 
Comparative study of three tests (dye test, 
indirect haemagglutination test, latex 
agglutination test) for the detection of 
antibodies to Toxoplasma gondii in human 
2 Wreggitt TG, Gray JJ, Balfour AH. Problems 
with serological diagnosis of Toxoplasma 
gondii infections in heart transplant 
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value of cytomegalovirus IgM antibody in 
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Harford JP, Fleck DG, Mythen M, Saunders RJ. 
Public Health Laboratory Service enzyme 
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Toxoplasma specific IgM antibody. J Clin 

Limited value of AgNOR enumeration in 
assessment of thyroid neoplasms

Argyrophilic nucleolar organising region 
associated proteins (AgNORs) have recently 
been shown to be of interest in a variety of 
different organs and disease states, including 
lymphomas,1 melanocytic lesions of the 
skin,2 and pleural mesothelioma.3 In these 
and other cases the enumeration of nucleolar 
structures has been shown to be of diagnostic 
value in differentiating benign from mali-
gnant disease, or in distinguishing between 
low and high grade malignancy. Accord-ingly, a study of AgNORs in thyroid tissue 
was undertaken to see if the technique could 
distinguish between benign and malignant 
lesions, particularly follicular neoplasms. 

Thirty three specimens were examined and 
these included anaplastic carcinoma (n = 4), 
follicular carcinoma (n = 4), papillary 
carcinoma (n = 6), follicular adenoma (n = 6), 
and nodular colloid goitre (n = 13). The 
usual one step silver colloid reaction4 was run 
at room temperature for 35 minutes. 
Intrac-
uuclear dots of silver deposit were counted in 
100 cells. Counting was difficult in papillary 
carcinomas because there were clear nuclei 
with often only one large intranuclear deposit, 
presumably corresponding to the 
nucleus. There was, however, consistency 
in all categories within cases.

The results are expressed in the figure.

Figure Scattergram showing mean number 
of AgNOR counts for each case in 
anaplastic, follicular, and papillary 
carcinoma and for adenoma and colloid 
goitre.

There was separation of AgNOR counts 
between anaplastic and both papillary and 
follicular carcinomas. The $\chi^2$ test was used to 
assess significance between the pooled means 
of anaplastic carcinoma and both follicular 
and papillary carcinomas (p < 0.05). A 
similar difference was found between ana-
plastic carcinoma and adenoma (p < 0.05) 
and a more significant difference (p < 0.02) 
between follicular and papillary carcinoma 
and also papillary carcinoma and colloid 
goitre. There was also a considerable overlap etweeen all carcinomas and colloid goitre.

No other pairings showed a significant 
difference, particularly follicular adenoma 
and carcinoma. These findings are not as 
clear cut as in other studies5 where a clear 
distinction was obtained between high and 
low grade lymphomas, benign and malig-
nant melanocytic lesions, and reactive 
mesothelium and mesotheliomas.

NORs are loops of ribosomal RNA and 
are therefore important in protein synthesis. 
It may be that follicular cells are in variable 
 stages of proliferation or protein synthetic 
activity in both benign and malignant 
conditions. Certainly in the study of other 
endocrine tissues such as breast and prostate 
by means of the AgNOR method, discrimination 
between benign and malignant tissue 
has been relatively disappointing. This may 
be, in part, the result of rRNA gene amplification 
in stimulated non-malignant endocrine 
cells, leading to increased gene copies and hence higher NOR numbers than in “resting” cells.6 AgNORs have therefore 
shown little value in differentiating between 
between benign and follicular follicular neoplasms in 
view of the “overlap” in numbers between 
these groups.

ER NORTON
J CROCKER
JULIE MCGOVERN
Department of Histopathology, 
East Birmingham Hospital, 
Bordesley Green East, 
Birmingham B9 5ST.

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region associated proteins in cutaneous 
melanocytic lesions: a quantitative study. J 
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tion of malignant from normal and reactive 
mesothelial cells using the argyrophil tech-
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Coulter Plus IV leucocyte volume analysis 
instrument: sensitivity of blast identification in 
peripheral blood

Previous reports evaluating the clinical 
usefulness of the three population differen-
tial have examined various diseases. We 
studied the sensitivity of the Coulter 
automated differential in identifying blast 
cells in peripheral blood samples.
Second International Conference on Gastrointestinal Cancer
August 27–September 1, 1989
Jerusalem, Israel
For further information please contact:
GIA Secretariat, PO Box 50006, 61500
Tel Aviv, Israel.

The symposium will be preceded by:
Clinical applications of a long-acting
somastostatin analogue on August 27 at
3.30 pm, The Jerusalem Hilton.

Fourth South-East-European Symposium of Paediatric Surgery
Graz, Austria, September 8–9, 1989
on normal and disturbed oesophageal
function in childhood
(Simultaneous translation)
Inquiries to: Professor Dr H Sauer,
Department of Paediatric Surgery,
the University of Graz, Heinrichstraße 31,
A-8010 Graz, Austria.

FUTURE IMLS SYMPOSIA

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<td>4 March 1989 Southampton</td>
<td>Conception to cradle—fetal and neonatal diagnosis and monitoring</td>
<td>Dr D McLellan, School of Pharmacy, Portsmouth Polytechnic, Park Building, King Henry I Street, Portsmouth PO11 2DZ.</td>
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<td>18 March 1989 Cambridge</td>
<td>Special coagulation review—educational seminar</td>
<td>Mr D M Reardon, Haematology Department, Addenbrooke’s Hospital, Hills Road, Cambridge CB2 2QH.</td>
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<tr>
<td>March 1989 Nottingham</td>
<td>Microbiology/virology</td>
<td>Mr A Pawley, Virology Department, PHL, Queens Medical Centre, University Hospital, Nottingham.</td>
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<td>8 April 1989 Sheffield</td>
<td>Haematology—in a lighter vein</td>
<td>Mr N R Porter, Haematology, Floor H, Royal Hallamshire Hospital, Glossop Road, Sheffield S10 2JF.</td>
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<td>26 April 1989 Liverpool</td>
<td>Laboratory developments in paediatric biochemistry</td>
<td>Mr D Kilshaw, Pathology Department, Arrow Park Hospital, Upton, Wirral, Merseyside L49 3PE.</td>
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2–9 September 1989 Coventry
IMLS Triennial Conference
Mr C G Smith, Pathology Department, Dudley Road Hospital, Summerfield, Birmingham B18 7QH

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<td>17 March 1990 Cambridge</td>
<td>Special red cell review—educational seminar</td>
<td>Mr D M Reardon, Haematology Department, Addenbrooke’s Hospital, Hills Road, Cambridge, CB2 2QH.</td>
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<td>30–31 March 1990 York</td>
<td>Microbiology/histology/cytology/immunology</td>
<td>Mr B Jones, Pathology Department, Royal Halifax Infirmary, Free School Lane, HalifaxHX1 2YP.</td>
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<td>April 1990 Durham</td>
<td>Blood Group Serology ’90</td>
<td>Miss D L Trattles, Blood Transfusion Laboratory, Department of Clinical Pathology, General Hospital, Ayresome Green Lane, Middlesbrough, Cleveland TS5 5AZ.</td>
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<td>13–16 September 1990 Sheffield</td>
<td>Microbe ’90</td>
<td>Mr C J P Brazier, Public Health Laboratory, Northern General Hospital, Herries Road, Sheffield S3 7AU.</td>
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<td>September 1990 Birmingham</td>
<td>Cellular pathology</td>
<td>Miss L Grossenvor, Histopathology Department, General Hospital, Steelhouse Lane, Birmingham B4 6NH.</td>
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<td>April 1991 Durham</td>
<td>Microbiology</td>
<td>Mr G P Hedley, Regional Transfusion Centre, Holland Drive, Barrack Road, Newcastle upon Tyne NE2 4NG.</td>
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<td>April 1991 Sheffield</td>
<td>Haematology—in a different vein</td>
<td>Mr N R Porter, Haematology, Floor H, Royal Hallamshire Hospital, Glossop Road, Sheffield S10 2JF.</td>
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<td>September 1991 Birmingham</td>
<td>Microbiology</td>
<td>Mr T Johnson, Microbiology Department, Selly Oak Hospital, Raddlebarn Road, Selly Oak, Birmingham B29 6JD.</td>
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