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Distinguishing lymphoma and small cell anaplastic carcinoma of the thyroid by immunocytochemistry

I read with interest the report by Burt *et al*[1] regarding the problem of differential diagnosis between lymphoma and small cell anaplastic carcinoma of the thyroid. A similar study has been performed in our department in Leicester on a smaller number of cases (19). In addition to thyroglobulin and epithelial membrane antigen antisera, our study also included a cytokeratin antibody (CAM 5·2). It was found that this antibody was more sensitive for detecting epithelial malignancies than epithelial membrane antigen. A combination of common leucocyte antigen and CAM 5·2 resolved the differential diagnosis in all but two of the cases. The use of this or a related cytokeratin antibody, together with common leucocyte antigen is thus suggested for this diagnostic problem. Epithelial membrane antigen, apart from reduced sensitivity compared with cytokeratin, suffers from the additional disadvantage of being reactive in a proportion of lymphomas.[2]

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T lymphocyte numbers and serum E rosette inhibitory substance

It is well established that T lymphocyte numbers are diminished in certain disease conditions. These include protein calorie malnutrition[1 2] plasmodium falciparum infection,[3 4] measles infection,[5 6] and systemic lupus erythematosus.[7] Others are HBs Ag positive chronic hepatitis,[8] rheumatoid arthritis,[9] and cancer.[10] Sera from most of these patients inhibit both in vitro phytohaemagglutinin transformation of lymphocytes,[5 11] and E rosette formation by normal human lymphocytes.[12–17]

There is circumstantial evidence to suggest that the presence of E rosette inhibitory substance (probably an immune complex) could be partly responsible for the diminished number of E rosettes that are recorded in these patients. Treatment with levamisole considerably increases the number of E rosette forming T lymphocytes in vivo and in vitro in several disease conditions, in which patients commonly possess circulating immune complexes and low E rosetting lymphocytes.[10 18–20] We also observed recently that, in common with children who had protein calorie malnutrition, children with malaria or measles infections had increased titres of circulating immune complexes, serum E rosette inhibitory substance(s), and diminished numbers of circulating E rosettes.[16]

It is therefore necessary for workers carrying out studies of lymphocyte subpopulations to be cautious in interpreting findings of diminished E rosettes. The percentage of E rosettes observed in such cases may not represent the total circulating E rosette numbers present. From our experience a test for the presence of E rosette inhibitory substance(s) in such patients is useful, as there may be some circulating T lymphocytes that are not capable of forming E rosettes in vitro, probably as a result of the previous binding of their surface receptors to inhibitory substances in vivo.[14]

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Detection of *Chlamydia trachomatis* by enzyme immunoassay, immunofluorescence, and cell culture

Mumtaz *et al*[1] presented their evaluation of a commercial enzyme immunoassay (Abbott Laboratories) for detecting *Chlamydia trachomatis* in urethral and cervical specimens. The hospital and laboratory in which the work was performed has a long and well established research interest in *C trachomatis*. We present our experience with this enzyme immunoassay in a district general hospital that does not have such an established interest but wishes to provide a rapid and reliable service for the diagnosis of *C trachomatis* infection. We also simultaneously tested many of our patients using a third technique.

We evaluated 83 cervical specimens by enzyme immunoassay and a McCoy cell culture technique that was essentially similar to that described by Mumtaz *et al*[1] except that cell monolayers were stained with Giemsa. Cell cultures were not passaged. Specimens were taken from women on their first visit to the clinic of genito-urinary medicine, irrespective of their reason for attendance. Fifty five of these patients were also tested by direct immunofluorescence using a fluorescein labelled genus specific monoclonal antibody (Boots-Celltech Diagnostics). Specimens were considered to be positive if 10 or more fluorescing elementary bodies were seen. In each case the swab for enzyme immunoassay was taken before the swab for cell culture. If immunofluorescence was being performed the swab for culture was used to prepare a slide before it was placed in transport medium.

Comparison of cell culture, enzyme immunoassay, and immunofluorescence for detecting C trachomatis in cervical samples

	Cell culture	
Enzyme immunoassay	*Positive*	*Negative*
Positive	17*	3†
Negative	3‡	60§

* 11 tested by immunofluorescence: 10 positive;
† Two tested by immunofluorescence: two positive;
‡ One tested by immunofluorescence: one positive;
§ 41 tested by immunofluorescence: 41 negative.

The Table shows the results. *C trachomatis* was isolated from 20 (24%) samples. Seventeen of these were positive by enzyme immunoassay. Of the three samples negative by enzyme immunoassay, one was positive by immunofluorescence. Two of the three cases that were negative by cell culture but positive by enzyme immunoassay were tested by immunofluorescence, and both were positive. None of the three patients whose samples were negative by cell culture had been treated with antibiotics in the few months before sampling.

Although the number of specimens evaluated was small, our results were similar to those of previous studies.[1–4] In addition, the results obtained indicate that specimens positive by enzyme immunoassay but negative by cell culture are not necessarily false positives but may represent the loss of viability of *C trachomatis* during transport. Our findings also raise some doubt about the use of cell culture as the "gold standard" and the value of defining a "specificity" (the number of healthy subjects with a negative test result divided by the total number of healthy subjects)[5] for antigen detection assays.

If the enzyme immunoassay test is compared only with cell culture its specificity in our evaluation was 95% (60/63). Two of the three discrepant results were positive by immunofluorescence, and the third was not tested by this technique. If these results are taken to indicate that these two specimens were positive (and the third specimen disregarded) the specificity of enzyme immunoassay may be considered to be 100% (60/60) in our small series. Similarly, the sensitivity of the enzyme immunoassay improves from 85% (17/20) to 86% (19/22) compared with a sensitivity for cell culture of 91% (20/22). Therefore, sensitivity and specificity figures must be interpreted with caution when the reference test is known to have a sensitivity of less than 100%.

In conclusion, although under ideal conditions cell culture may be more sensitive than enzyme immunoassay, in routine diagnostic use this is probably balanced by the failure to isolate *C trachomatis* from several infected patients. The use of such an enzyme immunoassay has resulted in a considerable improvement in our service, as we no longer encounter the problems of maintaining a cell line of required sensitivity for the reliable isolation of *C trachomatis*.

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Comparison of methods for detecting *Chlamydia trachomatis*

Dr Ridgway and others reply as follows: Morgan-Capner *et al* raise the possibility that apparent false positive results with the new chlamydial antigen detection methods may reflect deficiencies in the cell culture