Correspondence

TO THE EDITOR, British Journal of Venereal Diseases

T cell proliferative responses to Chlamydia trachomatis antigen in vitro in patients with a history of gynaecological chlamydial infection

Sir,
The importance of Chlamydia trachomatis in genital tract infections is well established.1-4 The humoral immune responses during chlamydial infections have been examined with several serological techniques,5 and in vitro cellular immune responses to chlamydial antigens in man have been studied with different assays.6-8 We report on the immune response after local and more severe chlamydial infections.

One group of patients consisted of 10 women who had had acute salpingitis 1-2 years before, probably caused by C trachomatis. The women had had a positive chlamydial culture from the cervix and a fourfold or more rise in chlamydial IgG antibody titre during the disease. The other group consisted of 10 women who 1-2 months earlier had had chlamydial cervicitis, with only lower genital tract symptoms.

T cells and non-T cells were prepared from defibrinated blood, as described previously.8 A suspension of partially purified C trachomatis subtype LGV-2 was used as antigen throughout the study.8 An enzyme linked immunosorbent assay (ELISA) was used to show chlamydial IgG antibodies.5 A titre >1/8 was defined as a positive result. The T cell proliferative responses to chlamydial antigen were tested in U-bottomed wells using 2 x 10^4 antigen pulsed non-T cells and 5 x 10^4 T cells. After culture for five days the incorporation of 3H-thymidine was assessed by liquid scintillation counting.

As shown in the table, the proliferative T cell responses and the antibody titres to the chlamydial antigen were similar whether the women had had a serious or a minor chlamydial infection. Substantial variations were, however, observed in both patient groups. Our data suggest that T cell mediated immune responses are triggered to a quite high level after only local infections (such as cervicitis), and in general the response is no higher after a more invasive infection (such as salpingitis).

To further explore the role of T cell mediated immune responses, further studies should investigate T cell mediated cytotoxicity against cells infected with C trachomatis.

Yours faithfully,

E Ovquist*
M Onsrud* K Skaug†

*The Tissue Typing Laboratory, Rikshospitalet, The National Hospital, Oslo 1, Norway.
†The Microbiological Laboratory, Ullevaal Hospital, Oslo 1, Norway.

References


TO THE EDITOR, British Journal of Venereal Diseases

Treatment of uncomplicated anogenital gonorrhoea with a single oral dose of 3 g amoxycillin combined with 250 mg clavulanic acid

Sir,
Recent reports from the United States, Japan, and Europe show an increase in the incidence of gonorrhoea.1,2 Although the incidence in the United Kingdom has been static over the past year, that of β-lactamase producing strains has shown an exponential rise.3 We report our observations on 110 patients with a diagnosis, based on smears and cultures or on cultures alone, of uncomplicated anogenital gonorrhoea. All patients were treated with a single oral dose of 3 g amoxycillin potentiated with 250 mg clavulanic acid (Augmentin). Patients who did not live in Bournemouth, were sensitive to penicillin, or pregnant were not included in the trial. In the final analysis those who did not attend for at least two follow up examinations were excluded.

TABLE I Bacteriologically positive diagnoses in men

<table>
<thead>
<tr>
<th>Site</th>
<th>Smear only</th>
<th>Culture/smear and culture</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urethra</td>
<td>6†</td>
<td>50</td>
<td>56</td>
</tr>
<tr>
<td>Rectum</td>
<td>0</td>
<td>17</td>
<td>17</td>
</tr>
<tr>
<td>Both</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>6</td>
<td>68</td>
<td>74</td>
</tr>
</tbody>
</table>

TABLE II Bacteriologically positive diagnoses in women

<table>
<thead>
<tr>
<th>Site</th>
<th>Smear only</th>
<th>Culture/smear and culture</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urethra</td>
<td>1</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Cervix</td>
<td>4</td>
<td>3</td>
<td>7</td>
</tr>
<tr>
<td>Both</td>
<td>1</td>
<td>21</td>
<td>22</td>
</tr>
<tr>
<td>Total</td>
<td>6</td>
<td>29</td>
<td>35</td>
</tr>
</tbody>
</table>

*At least one positive culture.
†Primary female consort of 4 gave positive cultures (treated in this clinic).
Correspondence

Of the 110 patients studied (75 men, 35 women), 109 (99.2%) were cured, with negative results to smears and cultures on at least two occasions after treatment (tables I and II). Side effects were mild and infrequent (3.6%). One treatment failure was identified among the 75 male patients. Neisseria gonorrhoeae was isolated on the ninth day at his second follow-up examination, although smear and culture had given negative results at his first checkup 48 hours after treatment. He emphatically denied re-exposure, but the culture isolate was sensitive to Augmentin and even to amoxycillin alone. Isolates from 88 patients were tested for drug sensitivity; 22 isolates having failed to grow on subculture. One β-lactamase producing strain of Neisseria gonorrhoeae was isolated from a Nigerian patient in whom treatment was successful although the isolate was resistant to amoxycillin alone (MIC 32 mg/l). One other isolate was resistant to amoxycillin (MIC 4 mg/l). The trial included a patient who had failed to respond to oral tinalplicillin 2 g and to intramuscular spectinomycin at a dosage of 4 g but was cured by Augmentin. As only a single β-lactamase producing strain was isolated in this trial it is not possible to generalise about the effectiveness of Augmentin against such strains. We think that this combination drug is an ideal second line single dose oral medication for use where other forms or oral preparations have failed, especially as the incidence of β-lactamase producing and other resistant strains is increasingly reported.1 4

Yours faithfully,

A H De Silva S A Q Bashi
R Basu Roy
Department of Genitourinary Medicine, Royal Victoria Hospital, Gloucester Road, Bournemouth BH7 6JF

References

TO THE EDITOR, British Journal of Venereal Diseases

A simple suggestion to distinguish between auxotypes of Neisseria gonorrhoeae

Sir,

Neisseria gonorrhoeae strains occur as natural auxotypes,1 2 and at present auxotyping is probably the only practical tool for studies on the epidemiology of gonorrhoea. The method is based on the growth of clinical isolates in a series of synthetic media with different nutrients (specific requirements) added or removed.3 4 A specific substrate requirement results from a block in its synthesis by the auxotrophic strain. However, the specific nutrient required by an auxotype may also serve as an energy source in vivo if the organism's primary energy source is limited at the site of infection.5 6 In vitro studies have shown that glutamate dehydrogenase can serve either catabolic or anabolic functions in the gonococcus depending on the availability of glucose.7 8

Previous data indicate that a proline requiring (pro−) auxotype in a synthetic medium4 requires about twice as much proline as a non-requiring (zero) auxotype by early log phase, and needs about four times as much proline for an equivalent level of growth by stationary phase (MA Chan and M Goldner, personal communication).

In view of these data, a pro− auxotype (HGH P5/79) and a zero auxotype (HGH Z5/79) were compared regarding the amount of carbon dioxide produced from the metabolism of proline and glucose. Classic Warburg respirometry9 and the simple barium hydroxide indicator system of Slifkin and Pouchet10 were adapted for the experiments. The organisms were cultured in GC broth with added supplement. Equivalent suspensions of washed log phase gonococci in buffered (pH 7-0) maintenance medium were then provided with a substrate for the metabolic studies. Warburg respirometry9 verified the accuracy and reproducibility of the barium hydroxide indicator system.10 The indicator system consisted of glass serum vials for the metabolic reactions and evacuated glass tubes (Vacutainer, Becton Dickinson, Rutherford, New Jersey, USA) containing a saturated solution of barium hydroxide. The gaseous contents of a serum vial were sampled by inserting an adapter needle to connect the vacutainer (containing barium hydroxide) with the reaction chamber for 10 minutes. The amount of barium carbonate precipitate formed was measured by its net absorbance (650 nm). (The net absorbance refers to the absorbance reading minus the combined endogenous and atmospheric readings for each determination). The contents of a particular vial were sampled only once in the course of the experiment.

The results indicated that the zero auxotype (HGH Z5/79) liberated approximately twice as much carbon dioxide as the pro− auxotype (HGH P5/79) when presented with glucose, while the pro− auxotype liberated appreciably more carbon dioxide when catabolising proline. Although the difference in glucose degradation by HGH P5/79 and HGH Z5/79 was considerable, we do not know whether this would differentiate between HGH Z5/79 and other non-zero auxotypes. As gonococci use two metabolic pathways in their metabolism of glucose,11 it is possible that varying amounts of carbon dioxide are generated during glucose metabolism by different gonococcal isolates. Warburg respirometry confirmed the observations made using the barium hydroxide indicator system for the two strains. The major change seen was that the metabolic pattern for proline as opposed to glucose is consistently reversed for the two auxotypes. Thus the pro− auxotype (HGH P5/79) could be distinguished from the zero auxotype (HGH Z5/79) by comparing the extent of proline and glucose catabolism. A simple way to trace the more common auxotypic markers, such as proline or arginine, may be by observing their use as an energy source.

Epidemiological studies of gonorrhoea could be helped by a simpler method of distinguishing between clinical isolates. The barium hydroxide indicator system may point to an inexpensive alternative method more amenable to wider use. Other strains should be processed in order to illustrate its applicability. Auxotypes with multiple requirements could also be tested.

We thank Dr Anne Hendry of Hamilton General Hospital, Hamilton, Ontario, for providing the auxotypes.

Yours faithfully, J A E Franczyk M Goldner
Department of Microbiology, University of Toronto, Toronto, Ontario MSS 1A8, Canada

References
Treatment of uncomplicated anogenital gonorrhoea with a single oral dose of 3 g amoxycillin combined with 250 mg clavulanic acid.

A H De Silva, S A Bashi and R Basu Roy

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