Attemp to modify klebsiella carriage in ankylosing spondylitic patients by diet: correlation of klebsiella carriage with disease activity

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SUMMARY Patients with ankylosing spondylitis were asked to follow a ‘klebsiella exclusion diet’ for 5 months of a 10-month study. The same percentage of faecal samples were positive for klebsiella whether the patients were on or off the experimental diet. The diet also failed to influence variability of klebsiella serotypes. We found no correlation between acquisition of klebsiella and deterioration of disease symptoms, as recorded by the patients. Furthermore, carriage of klebsiella did not correlate with any of the following parameters of disease activity measured in the outpatient clinic: morning stiffness, pain measured on a visual analogue scale, analgesic consumption, ESR, total serum IgA. We found no evidence, therefore, that faecal klebsiella is involved in disease exacerbations of ankylosing spondylitis.

The suggestion by Ebringer and co-workers¹ that carriage of Klebsiella aerogenes in the bowel is linked with disease activity in ankylosing spondylitis has sparked off considerable controversy. Warren and Brewerton² claim that no such relationship exists. Our first study³ suggested a link between klebsiella and both acute anterior uveitis and peripheral synovitis, but found no relationship between spinal disease activity and faecal klebsiella carriage. In a second, long-term, prospective study we tentatively reported the relationship between the acquisition of klebsiella and increasing spinal disease activity or peripheral synovitis, but there were too few patients with acute anterior uveitis from whom to draw any definite conclusions about this aspect of the disease.⁴

Klebsiella in the bowel is derived mainly from ingested food, particularly salads and cold meats.⁵ This study was set up to determine whether it is possible to eliminate klebsiella from the bowel by the exclusion of these foods. We also wished to re-examine the relationship between faecal klebsiella and disease activity in ankylosing spondylitis.

Patients and methods

Thirty patients (24 men and 6 women) with ankylosing spondylitis fulfilling New York criteria⁶ were recruited. These patients had a mean age of 43 years (range 25–64 years) and a mean duration of ankylosing spondylitis of 13 years (range 3–26 years). The patients were divided randomly into 2 groups. The first group received the exclusion diet (see Appendix) for the first 5 months of the 10-month study, and then changed back to their normal diet for 5 months. The second group received the exclusion diet for the second half of the study. All patients were assessed at 10-weekly intervals in the outpatient clinic by the following measurements: morning stiffness, pain score on a 10 cm visual analogue scale, anterior and lateral spinal and cervical flexion, chest expansion, patient’s overall assessment of ability to work, the erythrocyte sedimentation rate (ESR, Westergren) and total serum immunoglobulins were also recorded.

In addition patients were asked to record on diary cards morning stiffness, analgesic requirements, and any eye soreness (if present) at weekly intervals throughout the study. By means of these cards the patients were then subdivided into 3 categories of disease activity as follows:

1. Active: 1 hour or more of morning stiffness and requiring more than 100 mg of indomethacin or equivalent per 24 hours.

2. Inactive: less than 15 minutes’ morning stiffness and requiring less than 50 mg of indomethacin or equivalent per 24 hours.
Correlation of klebsiella carriage with disease activity

(3) Probably active: between the above 2 categories.

The patients sent faecal samples for determination of klebsiella carriage at fortnightly intervals throughout the study.

Klebsiella were isolated by a method adapted from Cooke et al. Faeces were subcultured on to MacConkey-inositol-carbenicillin (MIC) agar and in to MIC broth formulated as for agar but without the agar. The broth was incubated for 48 hours at 37°C before subculture to an MIC plate. The plates were incubated overnight at 37°C.

Klebsiella strains from patients whose faeces carried this organism at least once both on and off the diet were serotyped by standard capsular techniques.

Results

Nine patients withdrew from the study after less than 7 months; 8 of these patients withdrew because of difficulty in adhering to the diet and the ninth patient moved out of the area. Results from these patients have been excluded from the analysis.

Influence of diet on klebsiella carriage

Fourteen of the 21 patients analysed carried klebsiella at some time while on their normal diet and 17 while on the experimental diet. With each sample being taken as an individual event, 41 out of 123 (33%) yielded klebsiella on a normal diet and 50 out of 149 (34%) yielded klebsiella on the experimental diet. Thus the experimental diet had no effect on klebsiella carriage rates.

Capsular serotyping of the klebsiella strains isolated from 13 patients showed that the introduction of new serotypes occurred even when the patients were on the experimental diet. The 4 examples shown in Table 1 are typical of the changes found.

Relationship between klebsiella carriage and disease activity

Fluctuations in disease activity during the 10-month study period tended to be small. With the patients' own records being used to define disease activity, deteriorations ('inactive' to 'probably active' or 'probably active' to 'active') were reported only on 21 occasions out of a possible 206. Nine patients reported no clinical change throughout the 10-month study. Clinical deterioration was accompanied in only 4 instances by a change in faecal culture from negative to positive for klebsiella. On 22 other occasions when faecal culture changed from negative to positive the disease activity remained the same, and on one further occasion the disease activity actually diminished (Table 2).

Table 1 Influence of diet on variability of klebsiella serotypes

<table>
<thead>
<tr>
<th>Patient</th>
<th>Diet</th>
<th>Exclusion</th>
<th>Normal</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>62 - 52 52 -- 46</td>
<td>31 -- 6 17 -- NT</td>
<td>6 46</td>
</tr>
<tr>
<td>2</td>
<td>NT -- 43 -- 18 43 9</td>
<td>NT 20</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>-- 21 13 -- 46 46 -- 8</td>
<td>24</td>
<td>46</td>
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<tr>
<td>4</td>
<td>-- 52 -- 8 3</td>
<td>3 3 52 -- 8 --</td>
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</tbody>
</table>

Numbers refer to standard capsular serotypes.
NT = nontypable.
- = No klebsiella isolated.

We were thus unable to reproduce earlier results from this department suggesting that deterioration in clinical state was significantly associated with the appearance of klebsiella in the stool.

The 10-weekly assessments performed in the outpatient clinic also showed very little fluctuation in disease activity in individual patients, though there was considerable variation in the range of disease activity within the group as a whole. Figs. 1-4 illustrate the relationships between morning stiffness (Fig. 1), visual analogue pain score (Fig. 2), ESR (Fig. 3), serum IgA (Fig. 4), and klebsiella carriage, but show no significant trends. Patients with the highest pain score, morning stiffness, ESR, and serum IgA did not show the highest percentages of stools yielding klebsiella. Patients with the most variation in disease activity measured by these 4 parameters might have been expected to show the greatest variability in klebsiella carriage—that is, 50% of samples positive and 50% of samples negative for klebsiella—but this was not seen.

Assessments of anterior and lateral spinal and cervical flexion, chest expansion, and the patient's general assessment of pain and ability to work showed no significant changes over the 10-month period. There were also no attacks of uveitis requiring specific therapy, and too few instances of peripheral
Discussion

As a means of eliminating klebsiella and reducing disease exacerbations in ankylosing spondylitis this study has not been successful. There are a number of reasons why this approach may have failed to modify klebsiella carriage. There may have been poor compliance with the diet, as several patients commented that it was difficult to adhere to during summer months. Minor sources of klebsiella in food may not have been excluded, and environmental sources, particularly other parts of the bowel or body, are always a possible origin of faecal recolonisation.

An alternative method of reducing or eliminating klebsiella in the bowel would be the administration of appropriate antibiotics. However, organisms may fail to be eliminated by standard doses of antibiotics, as such standard doses are based on adequate serum concentrations. Antibiotic levels in the small and large bowels are usually considerably lower. Even if adequate antibiotic levels were achieved, long-term administration of the antibiotic might result in the emergence of resistant strains rather than complete elimination of the organism.*

The definition of disease activity in ankylosing spondylitis has caused considerable problems in previous work. Some authors have chosen to use an ESR of 15 mm/h to divide their patients into those with active and inactive spondylitis, an arbitrary division which has been widely criticised. We believe that dependence on a single parameter to monitor disease activity is unreliable and have therefore chosen to use a variety of clinical and laboratory measurements. We are confident that disease exacerbations were

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* For explanation see Figs. 1-4.
reliably detected by this approach. However, despite this approach we were unable to detect a relationship between any of these measurements and acquisition of klebsiella, acquisition of new klebsiella capsular serotypes, or overall carriage rates (prevalence) of klebsiella.

High yield rates for klebsiella (33% of samples positive) matched those of previous studies, and indicate that our faecal culture methods are sensitive and reliable. However, there are several problems inherent in studies using faecal culture as a means of detecting klebsiella. Firstly, such methods may fail to detect important changes in klebsiella carriage occurring elsewhere in the bowel, such as in the ascending colon. Secondly, proliferation of small, usually undetectable, numbers of bacteria resident in the bowel may be falsely interpreted as acquisition of klebsiella or acquisition of a new strain of klebsiella.

The idea that a bowel factor such as klebsiella bacterial antigens can trigger disease activity in ankylosing spondylitis remains attractive. Ankylosing spondylitis has numerous clinical similarities with Reiter's disease and other members of the seronegative spondylarthritides group. It also shares with Reiter's disease a strong association with the genetic marker HLA B27, and there is good evidence that Reiter's disease can follow certain enteric infections, notably shigella, salmonella, and campylobacter. It is therefore tempting to think that bowel flora may also play a part in the pathogenesis of ankylosing spondylitis.

We must conclude from this study that faecal klebsiella carriage bears no relationship to disease activity in ankylosing spondylitis. However, our results do not exclude a role for bacteria acting elsewhere in the bowel. We believe that examination of faecal flora is not an appropriate method for elucidating such a role.

APPENDIX

Exclusion diet

This diet was designed to exclude foods which are the most likely vehicles of klebsiella carriage.

INSTRUCTIONS TO PATIENTS

1. All hot food may be eaten. 'Hot' food served in canteens and restaurants may not be really hot, and, generally, eating food not prepared at home should be avoided whenever possible.
2. Salads, cold meat (except tinned) and all ready-prepared cold foods should be avoided.
3. Home prepared cold puddings may be eaten immediately after preparation, or later if refrigerated.
4. Raw fruit may be eaten after being peeled.
5. All tinned food may be eaten immediately after opening the tin.
6. Pasteurised, sterilised, or UHT milk should be used. If cold milk is drunk alone, this should be taken from a freshly opened bottle.
7. Cheese, yoghourt, eggs, pickles, sauces, bread, toffee, chocolate, biscuits, and cakes other than those which contain cream or custards are satisfactory.

References