Food additive-induced urticaria: studies of mediator release during provocation tests

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Since Juhlin [1] studied 330 cases of recurrent urticaria by means of challenge tests and attributed a third to food additive-induced reactions, there has been considerable controversy over the prevalence and, indeed, the existence of reactions of this kind [2,3]. Claims that specific antibodies are involved [4] remain unconfirmed [5] and it is possible that, as in aspirin-induced urticaria [6,7] the inflammatory mediators which are released during an urticarial reaction are triggered by a pharmacological and not an immunological mechanism [8].

In studying food additive reactions by means of challenge tests, urticaria has the advantage of providing a clear clinical endpoint. Since the mechanism remains unclear, it seemed appropriate to measure the release of inflammatory mediators such as histamine and prostaglandins, and to see whether the observed changes in mediator levels correlated with objective clinical effects.

Patients were recruited over a period of 18 months from three clinical centres that were already collaborating in other studies of food additives, based at High Wycombe, the Brompton and Guy’s Hospitals. Of 49 patients with urticaria, 24 had symptoms which appeared to remit on an additive-free diet. These 24 patients were then submitted to double-blind challenge tests with capsules containing various food additives. When the codes were subsequently broken, 15 had failed to react. Four reacted to aspirin (without having presented with a history of aspirin intolerance), two reacted to sodium benzoate, and only three were found to have given positive responses to at least two separate challenges to azo colours, with negative responses after placebo. This article is concerned with the subsequent investigation of these three patients, of whom two showed a response to rechallenge in the ward while the third failed to do so.

Methods
Preliminary studies

The diagnosis of food additive-induced urticaria was initially considered in patients with chronic urticaria whose symptoms disappeared or improved substantially on an additive-free diet. Where possible (as in all our three subjects) medication was withdrawn, and the response to a series of double-blind, placebo-controlled, food additive challenges was assessed. Patients were asked to take one of a series of capsules each breakfast time and to record symptoms daily on a diary card. Each series of 10 capsules consisted of five containing mixtures of additives interspersed with placebo capsules, put together in ‘low-dose’ and also in ‘high-dose’ packs.

The diary cards were analysed and the occurrence of moderate or severe urticaria after either high dose, or both low and high dose capsule challenge was recorded as a positive response. The code was then broken. In patients with severe urticaria a report of mild symptoms on up to two out of 10 placebo challenge days was not necessarily regarded as invalidating the test. Subjects who gave positive responses were, however, rechallenged with single substance capsules in a further series of tests and the diary cards analysed as before.

Inpatient study protocol

After giving clear-cut positive responses in both series of tests, three subjects were admitted to an investigation ward either at Guy’s or at High Wycombe Hospital for up to five days, each subject having stopped all medication and been symptom-free for at least 3–4 weeks. The patient’s use of an additive-free diet was confirmed and a low additive, low histamine diet prescribed, together with a liberal fluid intake, so that baseline measurements could be made in the absence of variations in urinary histamine

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Fig. 1. (J.L.) Five consecutive challenge days with tartrazine, sunset yellow, amaranth, carmoisine and azo mix given at regular time periods (↑). Clinical symptoms include urticaria (U), itch (I), erythema (E) and headache (Hd). Mediator measurements included plasma histamine (Hist), urinary 1-methyl histamine (1MH), histamine (H), prostaglandins 6-keto F₁α (6-keto F₁α), thromboxane B₂ (TXB₂) and prostaglandin E₂ (PGE₂). Creatinine = Creat.
Table 1. Patient data.

| Patient | Age (years) | Sex | Atopy | Duration of urticaria (years) | Other problems      |
|---------|-------------|-----|-------|-------------------------------|---------------------|
| J.L.    | 36          | F   | Y     | 20                            | Contact sensitivity to nickel |
| K.F.    | 40          | M   | N     | 5                             | Dermatographism     |
| A.P.    | 17          | M   | Y     | 5                             | Asthma              |

Histamine and creatinine measurements

Blood and urine samples were collected and assayed for histamine and urinary metabolites according to the method described earlier [10]. Urinary prostaglandin E2, 6-keto F1α, and thromboxane B2 were assayed using a competitive radioimmunoassay kit (NGN DuPont plc) following extraction and concentration on sep-pak (Walters plc) with methyl formate elution. All measurements were expressed as quantity of product/mg creatinine to correct for volume of urine voided. Creatinine was measured spectrophotometrically by the Jaffé reaction [11].

Bronchial provocation tests

Bronchial reactivity to inhaled histamine acid phosphate was determined by the method of Cockcroft [12] employing a Wright nebuliser calibrated to 0.12 ml/min.

Results

Case 1. J.L., a woman of 36, reacted on outpatient challenge both to mixed (high dose) azo dyes and to single-substance capsules containing sunset yellow (5.0 mg), tartrazine (0.5 mg) and carmoisine (50 mg), but not to amaranth (50 mg). In addition, mild urticaria (grade 1 out of 3) was observed on two of the 10 placebo days during these challenges. After a symptom-free interval of four weeks she was admitted to the ward. On each study day, she was challenged with incremental doses of either tartrazine, sunset yellow, amaranth or carmoisine, and clinical reactions were observed to all the four challenge compounds within 4–6 hours of capsule ingestion (Fig. 1).

Marked urticaria was seen with tartrazine and carmoisine, and the ingestion of sunset yellow and amaranth was followed by erythema and pruritus. Significant rises of plasma histamine paralleled the clinical symptoms.

There was a very good correlation between measurement of urinary histamine, its primary metabolite, and all three of the prostaglandins that were assayed. No changes were observed throughout the study in any of the other parameters measured, including the concentration of histamine needed to provoke bronchoconstriction. The PC20 was persistently in excess of 16 mg/ml.

Case 2. K.F., a man of 40, had been free of urticaria for several weeks on an azo colour-free diet when studied and had stopped all medication. On day 1 he received tartrazine in incremental doses of 0.5, 5.0, 50 and 150 mg. No urticaria developed although, unusually for him, sweating occurred after 5 mg and itching after 50 mg. Subsequent plasma assays showed a gradual increase in circulating plasma histamine from 0.1 to 1.0 ng/ml throughout the day. There was good agreement between measurements of urinary histamine and methyl histamine which paralleled the sustained rise in plasma histamine (Fig. 2). No significant change in bronchial provocation was noted at the end of the day (PC20 = 8 mg/ml).

On the second challenge day, mixed capsules of amaranth, sunset yellow and carmoisine were given containing 0.5 mg, 5 mg and then 50 mg of each at two-hourly intervals. Headache, sweating and pallor developed within two hours of taking the highest dose (50 mg) capsules, and this was found to coincide with a moderate rise in plasma histamine. There was no correlation between plasma and urinary histamine, nor were changes in lung function found after a histamine provocation test.

On day 3 he received, sequentially, 50 mg each of carmoisine, sunset yellow, amaranth and tartrazine. By 18.00 h he had no clinical symptoms and his response to a histamine challenge was unchanged. He was then discharged but, three hours after the last additive capsule, developed urticaria at home which progressed to angio-neurotic oedema, requiring antihistamine treatment with terfenadine. Plasma histamine showed rises after each additive and rose in the pre-discharge sample. He maintained his urine collection throughout the night; urinary 1-methyl histamine rose but subsided to baseline by 08.00 h the next morning.

Case 3. A.P. was an atopic asthmatic with exacerbations of urticaria after challenge with azo dyes on an outpatient basis. As an inpatient, he failed to develop any symptoms during single-blind challenges with 0.5, 5, 50 or 150 mg of either tartrazine, amaranth, carmoisine or sunset yellow. Furthermore, when the ingestion of azo colours was followed by strenuous exercise, this did not provoke his urticaria. After three study days he was discharged without a recurrence of symptoms.

Discussion

While it is generally accepted that reactions to sulphites, nitrites and monosodium glutamate can occasionally
occur [8], many of the symptoms that have been reported to occur after challenge with food colourants or other additives have been difficult to reproduce in controlled conditions. This has raised numerous questions about the validity of the initial diagnosis and it is clear that the public perception of additive-provoked reactions greatly exaggerates its prevalence [13]. Even when the initial diagnosis has itself been made by means of the double-blind challenge method, subsequent events may show it to be a transient phenomenon [14].

Because of these many doubts, it is important to document further cases in which there is a clear relationship between the ingestion of food additives and an objectively confirmed clinical reaction. We believe that our cases 1 and 2 fall into that category, although case 2 showed a substantially diminished response, compared with previous challenge tests. This variability in the clinical response requires explanation, and it remains to be established whether modifying factors play a part—such as the histamine content of the diet, the permeability of the bowel, or the essentially sedentary circumstances of inpatient studies. It is, for example, known that food-induced anaphylaxis can be strongly potentiated by exercise [15,16].

We believe that it is now established that adverse reactions to tartrazine and other colourants can occur.

Fig. 2. (a) (K.F.) Single challenge day with tartrazine doses of 0.5, 5, 50, 150 mg for urinary histamine and 1-methyl histamine showing periods of sweating and itching. (b) Plasma histamine levels measured on the same day.
We have no data to explain why they should be so variable in their expression, but this and the lack of abnormal immunological findings make an immunological explanation unlikely, and our evidence on mediator release would fit equally well with a pharmacological response. We repeatedly noted a time delay between the rise in the urinary excretion of histamine, its 1-methyl metabolite and cyclo-oxygenase products on the one hand and the rise in plasma histamine. Whether this implies a release of mediators from the gut or other tissues remains totally unknown.

Finally, we may need to evaluate further the use of mediator measurements as an aid to diagnosis. In case 2, the release of mediator was insufficient to cause objective clinical changes, and it remains to be seen whether measurements of these and other mediators can provide a reliable indicator of an adverse reaction, whether clinical or subclinical. The strict criteria needed to establish the validity of a clinical syndrome are not necessarily practical for clinical purposes and if test methods could be devised to demonstrate reproducible subclinical changes, this could be of considerable diagnostic value. Therefore there should be further study of mediator release assays as a potential adjunct to diagnostic challenge tests.

This article records part of a multicentre study of food additive intolerance commissioned by the Ministry of Agriculture, Fisheries and Food.

References
1. Juhlin, L. (1981). British Journal of Dermatology, 104, 369.
2. Stevenson, D. D., Simon, R. A., Lumry, W. R. and Mathison, D. A. (1986) Journal of Allergy and Clinical Immunology, 78, 182.
3. Gibson, A. and Clancy, R. (1980). Clinical Allergy, 10, 699.
4. Welisky, N. and Heiner, D. C. (1980). Clinical Allergy, 10, 375.
5. Report of a working group on adverse reactions to ingested additives, 111/556 81-EN (1981). Brussels: Commission of the European Communities.
6. Asad, S. I., Youlten, L. J. F. and Lessof, M. H. (1983). Clinical Allergy, 13, 459.
7. Asad, S. I., Kemeny, D. M., Youlten, L. J. F., Frankland, A. W. and Lessof, M. H. (1984). British Medical Journal, 288, 745.
8. Lessof, M. H. (1987). Journal of Royal College of Physicians of London, 21, 237.
9. Granerus, G. (1968). Scandinavian Journal of Clinical and Laboratory Investigation, 22 (suppl), 104.
10. Murdoch, R. D., Pollock, I. and Naeem, S. (1987). Journal of Royal College of Physicians of London, 21, 257.
11. Wooton, I. D. P. (1986). In Microanalysis in medical biochemistry (ed E. D. Wooton) p 67. Edinburgh: Churchill Livingstone.
12. Cockcroft, D. W., Killian, D. N., Mellon, J. J. A. and Hargreave, F. G. (1977). Clinical Allergy, 7, 235.
13. Young, E., Patel, S., Stoneham, M., Rona, R. and Wilkinson, J. D. (1987). Journal of the Royal College of Physicians of London, 21, 241.
14. Pollock, I. and Warner, J. O. (1987). Journal of Royal College of Physicians of London, 21, 246.
15. Maulitz, R. M., Pratt, D. S. and Schocket, A. L. (1979). Journal of Allergy and Clinical Immunology, 63, 633.
16. Kidd III, J. M., Cohen, S. H., Sosman, A. J. and Fink, J. N. (1983). Journal of Allergy and Clinical Immunology, 71, 407.