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SHORT REPORTS

Haemolytic Anaemia due to Penicillin

A case of penicillin-induced haemolytic anaemia is described. Unusual features were that the penicillin was given in the usual therapeutic dose and only IgM antibodies were shown.

Case Report

A 22-year-old woman was admitted after a car accident in which she sustained a compound fracture of her frontal bone. She was given prophylactic antibiotics: an immediate dose of 600 mg of benzyl penicillin intramuscularly followed by 300 mg of benzyl penicillin q.d.s. for 24 hours, then 500 mg of phenoxymethyl penicillin four times a day. Eight days after admission her haemoglobin had fallen from 13.7 g/dl on admission to 10.7 g/dl, her reticulocyte count was 4.4% and a direct Coombs test was positive. The serum haptoglobins were 0.13 g/l. The result of the Schumm's test was negative. Three days after this her haemoglobin had fallen to 7.8 g/dl with a reticulocyte count of 12.5%. The next day haemoglobin was 6.8 g/dl. At this time her serum contained an antibody which reacted against her red cells when they had been sensitized with penicillin, and against penicillin-sensitized pooled red cells. There was no reaction with unsensitized red cells. The conclusion was that haemolytic anaemia was being caused by penicillin. The penicillin was stopped and eight days later her haemoglobin had risen to 11.3 g/dl, the reticulocyte count was 15%, and the Coombs test was only weakly positive.

The temperature at which maximum activity occurred was between 25°C and 30°C. The antibody was tested for activity against red cells sensitized by the different penicillins and by cephaloridine: benzyl penicillin B.P. gave a titre of 1/128, phenoxymethyl penicillin gave 1/256, and cephaloridine B.P. gave no titre. Assay of the immunoglobulin fractions showed that the IgG and IgA levels were normal but that the IgM level was increased. The serum was exposed to 2-mercaptoethanol for two hours and then dialysed overnight. The treated serum did not react with penicillin-sensitized red cells in saline at 20°C or by the indirect Coombs technique at 37°C. The inhibition of activity by 2-mercaptoethanol is peculiar to IgM.

Discussion

Since haemolytic anaemia due to penicillin allergy was first described by Ley *et al.*¹ fewer than 20 cases have been reported. Nearly all of these were in patients with subacute bacterial endocarditis who had been treated with high doses of intravenous penicillin. There is some discussion about the roles of the IgG and IgM fractions of the immunoglobulins in the pathogenesis of this condition. Levine and Redmond² found that only patients with a high IgG titre were positive on Coombs test. White *et al.*³ thought that a strongly positive Coombs test result due to IgG was an important diagnostic feature. Bird *et al.*⁴ have recently described a case similar to ours; their patient was a 3-year-old boy who was treated with 125 mg of phenoxymethyl penicillin four times a day for 48 hours for an upper respiratory tract infection. He developed a haemolytic anaemia which regressed rapidly when the penicillin was stopped. They showed that the antibody was in the IgM class. It has been thought that the hapten responsible is a part of the molecule common to penicillins and cephaloridine, but as shown above the antibody in this case did not react against cephaloridine. This case confirms that penicillin can cause a haemolytic anaemia in adults at doses customarily used for minor infections. It is interesting to speculate how many patients who

develop an anaemia while on penicillin are suffering from this type of haemolytic anaemia.

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Rhesus Sensitization Associated with I.U.D. in Pregnancy

Finn first put forward the idea that it might be possible to prevent immunization of Rh-negative mothers by giving them antibody to destroy Rh-positive fetal cells.¹ Others subsequently suggested the use of anti D-globulin as such an antibody; and, by its use, Clarke showed that fetal red cells were removed from the maternal circulation and their antigenicity reduced.²

It is well known that a diseased or disturbed placenta increases the risk of maternal/fetal transfusion and of sensitization. Numerous reports have been published of fetal erythrocytes appearing in the maternal circulation after surgical intervention in pregnancies at or near term, as a result of termination of early pregnancy, or following intranatal procedures such as external cephalic version. Toxaemia of pregnancy, with its resultant placental infarcts of uncertain age, is recognized as an obstetric determinant of Rhesus sensitization. The immunizing effects of spontaneous or induced abortions in cases of fetal/maternal blood group incompatibility was recognized by Levine.³ And Weiner discussed the sufficiency of a microtransfusion of 0.05 ml of fetal cells in sensitizing the mother.⁴ The following case illustrates the sensitizing effect of microtransfusion.

Case Report

In 1965 a 20-year-old married woman had a normal spontaneous delivery in hospital, at 33 weeks' gestation, of a healthy infant weighing 1930 g. Her blood group is O Rhesus D negative, genotype cde/cde.

In 1967 she had an uneventful domiciliary delivery at 39 weeks of a healthy 3630 g infant. In 1971 she spontaneously aborted and underwent an evacuation of uterus at 10 weeks by dates. Rhesus antibodies were not noted on regular routine screening in any of her pregnancies.

On 30 January 1974 when eight weeks pregnant by dates, the pregnancy was terminated by vacuum extraction. She was given anti-D globulin as a routine prophylactic procedure. On 19 February she was fitted with a Dalkon shield, an intrauterine contraceptive device, but became pregnant again. She had a positive Prognosticon test on 17 May, suggesting an expected date of confinement around January 1975. During this pregnancy her blood was tested for autoimmune antibodies on 17 May 1974 when six weeks by dates, and on 22 November at 33 weeks: none were found.

A few days before her expected date of confinement she went into spontaneous labour. Within two hours the intrauterine device was extruded spontaneously, there being little if any associated bleeding. Shortly before delivery the membranes were artificially ruptured, routine delivery following within 15 minutes. The total duration of labour had been four-and-a-half hours. The baby weighed 3350 g. Routine postnatal cord and maternal blood samples were submitted for investigation.

On spontaneous delivery of the placenta a thin layer of adherent retro-placental clot extended segmentally from the placental margin to its centre. The intrauterine device, which was extruded prior to membrane rupture, and without blood loss, must have lain adjacent to the placental edge and been responsible for the retroplacental bleeding.

Examination of the maternal delivery sample of blood showed "an immune antibody, very much like anti-D." It was present in only a weak titre which it was thought could well have formed in the time available between the sample submitted in November and the delivery one. A subsequent sample was noted 10 weeks after delivery to have an identical titre of anti-D by automated quantitative testing. Cord sampling at delivery suggested the infant's blood group to be O Rhesus positive.

Conclusion

Intrauterine contraceptive devices, and in particular the Dalkon shield, have been associated with a number of other complications of pregnancy.⁵ In this case it is possible that the intrauterine contraceptive device, causing retroplacental damage in late pregnancy, was responsible for sufficient fetomaternal haemorrhage to cause maternal rhesus sensitization.

I am indebted to Dr. W. Wagstaff, Director of the Sheffield Regional Transfusion Centre, for his opinion and advice.

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Pseudomonas Infection Associated with Contamination of Wick-type Air Freshener

The importance of *Pseudomonas aeruginosa* in the neonatal nursery is well recognized.¹⁻⁴ During a routine bacteriological survey in a maternity hospital four babies in a special-care neonatal unit were carrying a strain (pyocine type 5)⁵ that had not been isolated previously in the hospital (see table). The wick-type air freshener was suspected when an attendant was seen to change a baby's napkin and immediately raise the wick of the bottle before washing her hands. Subsequently another person lowered the wick before handling the infant. Strains of the same pyocine type were isolated from air-freshener bottles in the infected babies' cubicles. All strains were of the same phage type (31/44/68/F8/109/119X/1214) and serotype (6). Several bottles in the nursery yielded Gram-negative organisms other than *Ps. aeruginosa*.

The air freshener was intended as a deodorant and not as a disinfectant. It contained 0.1% formaldehyde to maintain sterility. When the wick was permanently raised the fluid normally evaporated completely in about four weeks. Empty bottles were replenished from a stock bottle of the proprietary fluid. Possibly, as the fluid vaporized, the strength of the residual formaldehyde solution would fall to a level which was no longer bactericidal. If a partially empty bottle was

Babies Colonized by *Ps. aeruginosa* in Intensive Care Nursery

Case No.	<i>Pseudomonas</i> first Isolated	Diagnosis in Patient Colonized	Sites Colonized
1	25/7/73	Apnoeic attacks, pneumonia	Nose, throat, urine, rectum
2	3/8/73	Respiratory distress syndrome	Nose, throat, rectum
3	13/8/73	Respiratory distress syndrome	Nose, faeces
4	20/8/73	Birth asphyxia after breech delivery	Nose, throat, umbilicus, rectum

then refilled from stock the fluid would contain less than 0.1% formaldehyde.

Studies on Air Fresheners

The bactericidal activity of both aqueous formaldehyde solution and unused air-freshener fluid was determined against the organisms isolated from the ward bottles and a standard strain of *Ps. aeruginosa* NCTC 10490. Organisms were inoculated into doubling dilution series prepared in nutrient broth, and after overnight incubation subcultures were made to agar plates. Surprisingly, though the standard strain was killed by a formaldehyde concentration of 0.009% it required a concentration of 0.037% to kill the air-freshener strains even after these had been repeatedly subcultured. Whereas the standard organism was killed by the air-freshener fluid at 1/16 concentration, a concentration of 1/4 was needed to kill the air-freshener strains, equivalent to 0.025% formaldehyde content.

Eight bottles of air freshener from a specially prepared batch without colouring agent were placed in a warm room with the wicks fully extended, and the volumes and formaldehyde concentrations measured weekly. Formaldehyde concentrations were determined by adding Schiff's leucobasic fuchsin to the fluid and measuring the colour developed in an E.E.L. densitometer. The average formaldehyde concentration fell to the critical level of 0.025% after three weeks. Organisms inoculated directly into the residual fluid after four weeks' exposure could still be recovered on subculture but organisms inoculated on to the top of the wick were not recovered from the fluid, possibly because the wick was too dry.

Our findings prompted a further survey in a general hospital. *Ps. aeruginosa* was isolated from several air fresheners from a respiratory unit where many patients were colonized by this organism. In a later survey *Ps. aeruginosa* together with other Gram-negative bacilli were isolated from three out of 35 bottles located in wards throughout the hospital, mainly in sluices and toilets.

Comment

Though unproved, the bottles may have been directly implicated in cross-infection and they should not be used in intensive care units and similar places. The practice of topping-up existing bottles from a stock solution should be discouraged since the resulting weakened mixture may eventually support the growth of organisms. Since an increase in the formaldehyde concentration proved irritant we recommend the use of safe non-volatile disinfectants. The possibility that organisms may develop resistance to formaldehyde solutions was not further investigated.

We thank Dr. M. T. Parker, Central Public Health Laboratory, Colindale, for typing the strains and the manufacturer of the air-freshener bottles for his helpful co-operation in these studies.

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