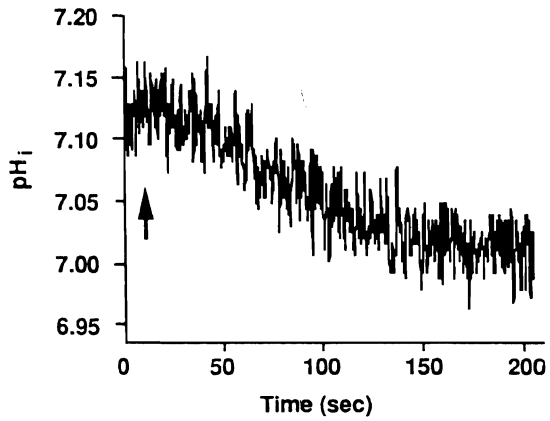

Effect of sodium bicarbonate (12 mmol/l, at arrow) on cytoplasmic pH (pH_i) in bovine aortic endothelial cells (AG4762, National Institute of Aging cell repository, passage 11) loaded with BCECF, trypsinised and suspended in stirred HEPES buffered Tyrode's solution at room temperature.

surface of the cell membrane. Despite this, we have observed qualitatively similar responses in suspensions of bovine aortic endothelial cells (figure), so the effect is not peculiar to platelets. Dr Iles and Dr Beech have very interesting data demonstrating increased pH_i in liver caused by a 15 min intravenous infusion of molar sodium bicarbonate in the ketoacidotic rat. It will be particularly interesting when the resolving power of this method is increased to permit measurement of the effect of bicarbonate on pH_i in cardiac muscle in animal models of cardiac arrest.

Professor Cohen and Professor Woods in their 1987 chapter[1] on treatment of acid-base disturbances presented a balanced account of the arguments, and we are sorry that they feel that our statement that they advocated the use of bicarbonate misrepresented them. They did write that hypertonic bicarbonate is standard therapy for the acidosis of cardiac arrest, but also pointed out that such therapy is not of unequivocal benefit. There are several precedents of treatments once used routinely in severely ill patients on grounds that they appeared reasonable but which have subsequently been proved to be positively harmful. Although our studies showing reduction in pH_i in response to bicarbonate in vitro are not conclusive evidence that such a deleterious effect occurs during bicarbonate therapy, we feel that the findings shift the burden of proof towards those that favour such treatment: perhaps the time is right to plan controlled trials of bicarbonate therapy despite the formidable difficulties that Cohen and Woods rightly identify.

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Factor VIII:C inhibitor associated with monoclonal-antibody-purified FVIII concentrate

SIR,—Several comments need to be made to put in perspective the letter by Dr Kessler and Dr Sachse (June 9, p 1403) concerning factor VIII:C inhibitor formation.

The actual rate of inhibitor formation in patients with severe haemophilia A, followed from childhood to adulthood, is unknown. The estimated rates of 5–15% are derived from cross-sectional rather than longitudinal studies and include patients with all degrees of severity. In one large "natural history" study, 29% of patients had inhibitor after 30 years of age, and at a time when only crude preparations of factor VIII:C were available.[1] Most inhibitors developed in children. It has been estimated that if severely affected (factor VIII:C < 1%) children were followed for 20 or 30 years then an inhibitor might develop in 30% or more (L. Aledort, personal communication). The cited rate of inhibitor formation in previously unexposed patients (7 of 39 or 18%) is not extraordinary or unexpected.[2]

The Malmö procedure for induction of immune tolerance has been successfully applied to many patients, but the actual response rate of patients is unknown.[3] Of the 11 patients treated by Nilsson et al, 2 (18%) did not respond. There is no way to know the significance of "the failure of the immune tolerance induction" in Kessler and Sachse's patient. The speculation that the manufacture of monoclonal-antibody-derived factor VIII:C could be associated with subtle denaturation of a native factor VIII:C protein, and creation of a neoantigen, is without factual support. The article referenced, with respect to alterations in the factor VIII:C gene, actually provides data that argue against this mechanism. Extensive animal testing during the preclinical laboratory evaluation of 'Monoclate' failed to reveal any evidence of neoantigen formation.[4]

It is premature to suggest that use of ultra-pure factor VIII:C products is related to an increased rate of inhibitor development, or to speculate on their failure to suppress inhibitors.

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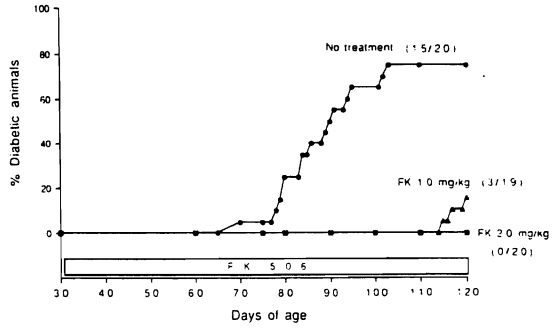
Prevention of spontaneous diabetes in BB rats with FK 506

SIR,—Cyclosporin can mitigate or prevent autoimmune BB rat diabetes,[1-3] and can induce remissions of human type I diabetes if instituted early after diagnosis and in adequate doses.[4-6] The potent new immunosuppressive agent, FK 506, which has had extensive clinical trials in transplantation,[7] suppresses T-cell activation in the same general way as cyclosporin does, by inhibiting the transcription of mRNA for early phase lymphokines such as interleukin-2, interleukin-3, and γ-interferon.[8] We have examined the influence of daily enteral tap water (group 1, n = 20), 1 mg/kg FK 506 (group 2, n = 19), or 2 mg/kg FK 506 (group 3, n = 20) on the incidence of spontaneous diabetes in our BB rat colony. The 59 animals were obtained from seven consecutive litters, randomly deployed to the three sex-matched groups, raised under controlled lighting, and allowed tap water and regular rat food (Purina 'Chow 5001') ad libitum. Treatment was between 30 and 120 days of age. The FK 506 (Fujisawa Pharmaceutical, Osaka) was dispersed with hydroxypropylmethylcellulose, diluted in tap water by sonication, and administered by gastric intubation. Onset of diabetes was diagnosed by glycosuria (tested thrice weekly) and confirmed by non-fasted blood sugar above 200 mg/dl from tail vein samples for 3 consecutive days.



Onset and incidence of diabetes during FK 506 treatment.

FATE AFTER 120 DAYS OF RATS TREATED WITH FK 506 FROM DAYS 30 TO 120

Outcome	Group 1 (water)	Group 2 (1·0 mg/kg)	Group 3 (2·0 mg/kg)
Survival ⩾120 days without diabetes	5/20	16/19	20/20
Killed at 120 days	2 of 5	6 of 16	6 of 20
Animals remaining beyond 120 days	3	10	14
Survival ⩾150 days without diabetes	3	8*	14
Killed at 150 days	3 of 3	2 of 8	2 of 14
Animals remaining beyond 150 days	..	6	12
Survival 165–195 days without diabetes	..	6	9†

*Other 2 animals became diabetic, at 132 and 147 days.
†Other 3 animals used for breeding and became diabetic while pregnant at 170, 175, and 180 days.

During the 90-day treatment period, diabetes developed in 15, 3, and none of the tap water, low-dose FK 506, and high-dose FK 506 groups, respectively (figure). Rats with diabetes died within a few days but there was no mortality or inhibition of weight gain among the non-diabetic rats. In these protected animals, blood glucose levels, measured every 2 weeks, and hepatic and renal function tests were always normal.

9 FK 506-treated rats (3 from group 2, 6 from group 3) had intraperitoneal glucose tolerance tests (2 g/kg) on the last day of treatment, and pancreas insulin content was measured by radioimmunoassay in 11 of the group 2 and 3 non-diabetic animals which were killed. The results were normal, and not different from those in 6 untreated Wistar-Furth (non-BB) rats. On histopathological examination, the pancreases of the non-diabetic FK 506-treated rats had little or no evidence of the insulitis and periductular mononuclear inflammation which was characteristic of the non-treated diabetic animals.

As with cyclosporin[3] the protective effect of FK 506 often outlasted the time of active treatment (table). Glucose intolerance, which has been described in BB rats under cyclosporin treatment,[9] was not observed with the doses of FK 506 used in this study. These findings support the possibility of clinical trials of immune intervention for recent onset diabetes.

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Amaurosis fugax and antiphospholipid antibodies

SIR,—Dr Booy (June 23, p 1538) describes a case of amaurosis fugax in a young woman and discusses whether a carotid angiogram should be done. The far less invasive procedure of phospholipid (aPL) antibody detection may be appropriate. The antiphospholipid antibody syndrome (APS), whose clinical features include thrombosis, thrombocytopenia, and recurrent miscarriages,[1] affects mainly young women. The solid phase cardiolipin antibody test and the lupus anticoagulant test are regarded as useful diagnostic tools.[2] Although a close pathogenetic link between autoantibodies and neurological diseases is still speculative, the clinical association of phospholipid antibodies and neurological disorders is no longer in dispute. Primary APS (without signs of definite connective-tissue disease) has been reported in association with focal cerebral ischaemia, migraine, transient global amnesia, chorea, myelopathy, and ocular ischaemia.[3,4] These antibodies have been described in systemic lupus erythematosus (SLE) in children, the highest levels occurring during neurological events,[5] and in children without any underlying pathological condition (primary APS).[6]

During the past two years we have studied fifteen female patients with primary APS. Amaurosis fugax is the only symptom in one patient; another patient presents with amaurosis fugax during recurrent episodes of migraine; a third has a central retinal artery thrombosis with permanent visual impairment. Asherson et al[7] pointed out that cardiolipin antibodies represent a risk factor for occlusive ocular vascular disease in SLE. Several of our SLE patients with cardiolipin antibodies have migraine with or without visual disturbances. We agree with Booy that carotid angiography is not indicated in young patients with amaurosis fugax, but we suggest that phospholipid antibodies be looked for.

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Acute viral hepatitis B in children: lack of chronicity

SIR,—Chronic hepatitis B virus infection had been shown to evolve from acute viral hepatitis B (AVHB) in 5–10% of cases.[1] However, recent studies in adults have shown that chronic infection develops after AVHB in only 1–3% of patients.[2,3] To evaluate the rate of progression to chronic infection after AVHB in children of less than 15 years old we followed prospectively 154 patients classified as having AVHB on hospital admission. All the children were positive for hepatitis B surface antigen (HBsAg) and/or IgM anti-HBs on admission, and were followed for between 3 and 24 months after admission. Diagnosis of acute viral hepatitis was made from clinical, biochemical, and serological features. Enzyme immunoassay (EIA) was used to detect HBsAg (Abbott) and anti-HBV antibodies (Institute of Organic Synthesis, Riga, USSR; Behring), anti-hepatitis A virus (HAV) antibody (Abbott), and anti-hepatitis D