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Erythropoietic proteins and antibody-mediated pure red cell aplasia: a potential role for micelles

Sir,

In a recent edition of *NDT*, Locatelli *et al*. [1] provided a useful review of the chronology and potential causes of anti-erythropoietin antibody-mediated pure red cell aplasia (PRCA) with erythropoiesis-stimulating agents, together with current actions and recommendations. The authors note the potential causal link between a change in the formulation of Eprex® (epoetin alfa), where the stabilizer human serum albumin was replaced with polysorbate 80 and glycine, and the recent upsurge in PRCA cases associated with its use [2]. However, a mechanism for how this formulation change breaks the immune tolerance to erythropoietin was not given. In principle, two main mechanisms exist by which B cell tolerance to self antigens can be broken. One way of breaking tolerance is to present the self antigens in combination with a danger signal, such as denatured protein or endotoxins. The most potent way to induce antibodies to self antigens is to present the self antigens in highly structured arrays that resemble viral capsid-like structures. Several potential causes of antibody induction with Eprex have been put forward, including the presence of leached contaminants from the rubber stoppers of pre-filled syringes or the silicon oil used as a lubricant, which may act as 'danger signals'.

We have shown recently the presence of surfactant molecule aggregations (otherwise known as micelles) in the Eprex formulation, which are caused by the high concentration of polysorbate 80. We have also shown that epoetin alfa is associated with these micelles, which may lead to the formation of structures with repeated epoetin antigens. These, in turn, may be presented at the surface of the micelles and break B cell tolerance. We employed gel permeation chromatography (GPC) and enzyme-linked immunosorbent assay (ELISA) to determine whether samples of Eprex (epoetin alfa) and NeoRecormon® (epoetin beta) formulations contained micelles and how much epoetin was micelle-associated [3]. The critical micelle concentration (CMC) is the concentration of a surfactant at which an appreciable number of micelles are formed, and CMCs for polysorbates 20 and 80 have been calculated previously [4]. Notably, Eprex contains 0.03% (w/v) Tween (polysorbate) 80, around 20 times its CMC; in contrast, NeoRecormon contains 0.01% (w/v) Tween 20, only approximately 1.5 times its CMC.

GPC analysis confirmed that the Eprex formulation contained micelles of polysorbate 80, while no micelles were detected with NeoRecormon. Subsequent analysis of the GPC fractions by ELISA demonstrated that with the Eprex samples, small amounts of epoetin were co-eluted with the polysorbate 80 micelles. No such co-elution was seen with NeoRecormon. The findings suggest that not only does the Eprex formulation contain polysorbate 80 micelles, but also that epoetin alfa molecules are solubilized in or attached to these micelles.

Micelle-associated epoetin is, therefore, a potential risk factor for immunogenicity in anaemic patients, and further investigation utilizing animal models may provide additional relevant data.

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Managing refractory uraemic pericarditis with colchicine

Sir,

Pericarditis is a complication of end-stage renal disease (ESRD), still occurring in 20% of uraemic patients before and at the initiation of haemodialysis [1]. Multiple factors contribute to the appearance of uraemic pericarditis, which responds readily to treatment and has a good prognosis in the majority of cases. We present a patient with 'refractory uraemic pericarditis' who ultimately responded to colchicine. The use of colchicine in uraemic patients with pericarditis has not been reported previously.

A 48-year-old woman, suffering from ESRD due to autosomal dominant polycystic kidney disease, attended our clinic in August 2000. She manifested tachycardia and deep heart tones without fever, dyspnoea, thoracic pain or cough. Laboratory tests were compatible with ESRD. There was no leukocytosis. Chest X-ray, electrocardiogram and heart ultrasound revealed a large amount of pericardial effusion (anterior wall, 9 mm; posterior wall, 17 mm), diastolic dysfunction and hypertrophy of the left ventricle; the ejection fraction remained within a satisfactory range (65%). The patient was enrolled in daily 3 h haemodialysis sessions without anticoagulation. A week later, a new ultrasound exhibited persistent pericardial effusion (anterior wall, 7 mm; posterior wall, 15 mm). Moreover, after 3 weeks of intensive dialysis, a progression of the effusion was noticed (anterior wall, 12.4 mm; posterior wall, 21.1 mm), while the ejection fraction remained stable. The patient's condition was also