Cytotoxic Agents in Childhood Glomerulonephritis

There is a substantial body of evidence that many forms of nephritis are mediated by immunopathogenetic mechanisms (Lewis and Couser, 1971). It has, therefore, seemed appropriate to attempt to modify the natural history of these diseases with agents known to alter the immune response (Cameron, 1971). These drugs are rarely simple in their effects (Berenbaum, 1967) and the rationale for their use in renal disease is slender. Nevertheless, gratifying results have been obtained in some circumstances, and in this review the evidence of effect of two such drugs—azathioprine and cyclophosphamide—will be examined.

In analysing the data, all anecdotal material must be treated with considerable reserve, for the natural history of nephritis in childhood is variable, and, until recently (Cameron, 1972), poorly documented. It is clear, for example, that some of the earlier claims of effect of azathioprine in steroid resistant nephrotic syndrome (White, Cameron, and Trounce, 1966) were in those diseases such as Henoch-Schönlein purpura where spontaneous improvement often occurs independent of therapy (Meadow *et al.*, 1972).

'Minimal Change' Lesion

Induction of remission. There is one effect of cytotoxic agents that is so consistent that it may be accepted without confirmation by controlled trial, and that is the ability of cyclophosphamide to induce a remission in children with the nephrotic syndrome and minimal histological abnormality on light microscopy of their renal biopsy specimens (Moncrieff *et al.*, 1969). It would now be ethically impossible to establish this point rigorously, for it would require an untreated control group from whom a known effective therapy, i.e. corticosteroids, was withheld. Incidentally, this effect of corticosteroids themselves has also not been formally demonstrated, though here again it is so obvious as to render trials unnecessary and unethical.

The incidence of cyclophosphamide-induced remission in the first episode of nephrotic syndrome in children unselected by renal histology has been

compared to that of prednisolone by Tsao and Yeung (1971). Fifteen children were treated with cyclophosphamide 5 mg/kg per day until leucopenia ($WBC < 4,000/mm^3$) had been induced; the cyclophosphamide dosage was then modified to maintain this leucopenia (usually about 3 mg/kg per day) for at least 6 months. 16 children received prednisolone 2 mg/kg per day until remission and were then maintained at a lower dose for 6 months or more. All but one of the cyclophosphamide-treated and all but 2 of the prednisolone-treated children went into remission, but two interesting differences were observed: first, the median time to remission on cyclophosphamide was 3 months, significantly longer than with prednisolone (1 month), and second, no relapses occurred after cessation of therapy in the 14 patients successfully treated with cyclophosphamide in contrast to 6 of the 14 patients who remitted on prednisolone.

The median time to remission is unusually long in Tsao and Yeung's children in Hong Kong: in our experience (T. M. Barratt, 1971, unpublished data) and others (Moncrieff *et al.*, 1969) the median time to remission is similar with cyclophosphamide and prednisolone. The reason for this difference is not clear, but the possibility of geographical variation in the nephrotic syndrome must be considered. The results are, nevertheless, impressive, but the dose of cyclophosphamide used was large, and as at least half the nephrotic children can be managed quite satisfactorily with corticosteroids alone (Arneil and Lam, 1966) it does not seem reasonable to use cyclophosphamide as the primary drug in the treatment of childhood nephrotic syndrome.

There is a small group of nephrotic children who do not respond to corticosteroid therapy as conventionally administered, yet have highly selective proteinuria and minimal histological abnormality. These children appear to respond to cyclophosphamide as satisfactorily as their steroid sensitive fellows (Moncrieff *et al.*, 1969).

Both azathioprine and 6-mercaptopurine, to which azathioprine is metabolized *in vivo*, appear to

be able to induce remission in corticosteroid dependent nephrotic children: Pachioli and Genova (1971) reported that all of 15 children treated with azathioprine and 39 children treated with 6-mercaptopurine remitted, but this effect is not consistent (T. M. Barratt, 1971, unpublished data).

Stability of remission. The stability of the remission induced by cyclophosphamide is striking, and has been formally demonstrated by Barratt and Soothill (1970). Steroid sensitive frequent relapsing nephrotic children were entered into this trial in remission on a maintenance dose of prednisolone, and were randomly allocated either to prednisolone withdrawal over an 8-week period, or to cyclophosphamide (3 mg/kg per day) and maintenance prednisolone for 8 weeks, followed by prednisolone withdrawal over a further 8-week period. 11 of 15 patients allocated to prednisolone alone had relapsed during a predetermined assessment period of 8 weeks after prednisolone withdrawal, in contrast to 3 of 15 cyclophosphamide treated children. At 1 year the corresponding figures were 13 and 5 relapses respectively.

Cyclophosphamide was administered to these patients during a steroid maintained remission when they had no overt manifestations of the disease, yet it clearly modified the underlying disease process in that it diminished the probability of relapse upon subsequent steroid withdrawal. This intriguing effect suggests that cyclophosphamide may act at a more fundamental level of the disease process.

Life table analysis (Cutler and Ederer, 1958) of steroid sensitive frequent relapsing nephrotic children shows that $75 \pm 7\%$ (SE) remain free from relapse one year after an 8-week course of cyclophosphamide at 3 mg/kg per day (T. M. Barratt, 1971, unpublished data). At 2 years $61 \pm 8\%$ are in remission. Some workers (Moncrieff *et al.*, 1969) have deliberately attempted to induce a leucopenia with cyclophosphamide, and Cameron (1971) has reported a similar analysis of patients treated in this way: $78 \pm 8\%$ were free from relapse at one year and $51 \pm 8\%$ at two years of follow-up. Thus there does not appear to be any particular advantage in inducing a leucopenia during therapy, and in the patients treated with cyclophosphamide 3 mg/kg per day for 8 weeks, there was no difference in the total white count or lymphocyte count at the end of therapy between the children who relapsed and those who remained in remission (T. M. Barratt, 1971, unpublished data). The choice of dose and duration of therapy in these series was arbitrary;

it is quite possible that much less cyclophosphamide would be sufficient, and trials are currently in progress to investigate this possibility.

It is by no means certain that cyclophosphamide is acting in its role as immunosuppressive agent in this disease, and it is indeed paradoxical that the most convincing evidence of its effect is in that variety of glomerular disease in which the evidence of immunopathogenesis is incomplete and circumstantial (Soothill, 1971).

The role of azathioprine in preventing relapse of steroid sensitive nephrotic syndrome has been examined by the International Co-operative Study of Kidney Disease in Children (Abramowicz *et al.*, 1970) by a less sensitive technique. The incidence of relapse in 18 patients given azathioprine (60 mg/m² per day) and intermittent prednisolone (40 mg/m² per day, 3 days out of 7) was compared to that in an equal-sized group given prednisolone and placebo. Five of the azathioprine treated children relapsed in a 6-month period in contrast to 8 of the control group; the difference is not significant. However, as the relapse rate on intermittent prednisolone is already quite low, only a large azathioprine effect would have been detected. The failure to detect an effect with azathioprine in contrast to the demonstrated effect of cyclophosphamide should not be construed as evidence of the superiority of cyclophosphamide, for the two drugs have not been directly compared, nor even tested in a similar manner.

Other Varieties of Glomerular Disease

Similar clinical patterns may result from many different forms of renal disease, each of which may have a different natural history and susceptibility to therapy. Extrapolation of assumptions of natural history or treatment response from one variety of renal disease to another, are, of course, extremely insecure.

In spite of the fact that cyclophosphamide has been used in the treatment of renal disease for 9 years (Coldbeck, 1963), no controlled study of its use in glomerular diseases in children other than nephrotic syndrome with minimal histological change has been published. This surprising deficiency is currently being rectified by the International Co-operative Study of Kidney Disease in Children, but anecdotal evidence suggests that if there is an effect, it must be small. There is some evidence that cyclophosphamide ameliorates lupus nephritis (Cameron *et al.*, 1970; Steinberg *et al.*, 1971), which is interesting in view of the marked protection against nephritis that is conferred by cyclophosphamide on New Zealand B/W mice who

tend to get a lupus-like illness (Russell and Hicks, 1968).

Azathioprine, by contrast, has been the subject of two important clinical trials. In the International Study Group trial (Abramowicz *et al.*, 1970), it was given to children with steroid resistant nephrotic syndrome for 90 days in a dosage of 60 mg/m² per day together with intermittent prednisolone (40 mg/m² per day, 3 days out of 7): of 16 patients 2 lost their proteinuria, 2 were improved, and the remainder unchanged. However, identical results were obtained with placebo, emphasizing the need for control studies.

The Medical Research Council (1971) contrasted azathioprine (2.5 mg/kg per day) and prednisolone (20 mg/day in adults, 0.5 mg/kg per day in children) with no treatment in children and adults with various forms of glomerular disease excluding minimal change. The authors concluded: 'There was no evidence of benefit from the treatment group as a whole; and the mortality was in fact higher in the treated group.' This conclusion may, however, be too pessimistic, for analysis of the data reveals that the excess of deaths was confined to the adult group. In the children treated for more than six months the creatinine clearance (92 ± 10 (SE) ml/min) was significantly higher than in the control group (63 ± 10 (SE) ml/min); this, after all, is the goal of treatment of glomerular disease.

Toxicity

Toxicity of these agents has been reviewed by Cameron (1971). Using cyclophosphamide in the dosage of 3 mg/kg per day for 8 weeks, overt toxicity is slight: under 5% of children have significant hair loss or neutropenia (T. M. Barratt, 1971, unpublished data). However, the potential toxicity of these agents is great: among the most alarming are facilitation of tumour growth, teratogenicity, gonadal effects, and long-term genetic damage. One important study is that of Berry (1971) who administered small doses of cyclophosphamide to prepubertal rats and observed a significant reduction in fertility of treated females, due to preimplantation losses. There must, therefore, remain some anxiety about the use of these agents in children with non-fatal disease; their use should be confined to individuals who cannot be satisfactorily managed by other means and the smallest effective dose should be determined for each agent, and particular care should be taken not to use more drug than necessary for each individual. Their use should be confined to clinical situations in which an effect has been rigorously demonstrated,

or to carefully controlled clinical trials in a restricted number of centres with adequate facilities for the assessment of renal histology and function; it is particularly important that clinical information on the effect of these toxic agents in the relatively uncommon disease situations described here should not be squandered by their indiscriminate use in inadequately studied patients.

The excess of deaths in the adults treated with azathioprine in the Medical Research Council was not due to complications of immunosuppressive agents, but to advancing renal failure: a similar effect has been observed by A. Adeniyi (personal communication, 1971) in the treatment of malarial nephrotic syndrome in Nigerian children. These observations raise the possibility that in some circumstances immunosuppressive agents may be actively harmful and augment the nephritic process. It is to be hoped that future understanding of the immunopathology of renal disease will lead to a more secure rationale for the use of these drugs in children with glomerulonephritis.

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MARTIN BARRATT

Department of Immunology, Institute
of Child Health, 30 Guilford Street, London W.C.1

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