PAPERS AND SHORT REPORTS

Blood cyclosporin concentrations and renal allograft dysfunction

D W HOLT, J T MARSDEN, A JOHNSTON, M BEWICK, D H TAUBE

Abstract

Forty nine renal allograft recipients taking oral cyclosporin suffered 76 episodes of renal dysfunction within six months of transplantation. These episodes were diagnosed as graft rejection or cyclosporin induced nephrotoxicity on the basis of histological findings in allograft biopsy specimens and the response to treatment. Mean predose blood cyclosporin concentrations measured by radioimmunoassay during the week before the onset of renal dysfunction were significantly higher when the cause was cyclosporin toxicity rather than graft rejection (392 v 741 nmol/l (471 v 891 ng/ml)). During this period there was a significant association between both the frequency of measurements above 666 nmol/l (800 ng/ml) and the diagnosis of toxicity and the frequency of measurements below 333 nmol/l (400 ng/ml) and the diagnosis of allograft rejection. Cyclosporin measurements made at the time of biopsy and reference to the highest or lowest concentrations measured during the week preceding biopsy were of less value in distinguishing between the two groups.

Despite lacking specificity for the parent compound, the radioimmunoassay used produced results which were of clinical value in optimising cyclosporin treatment.

Introduction

Cyclosporin is a cyclic undecapeptide of fungal origin, widely used as an immunosuppressant after transplant surgery. The main

adverse effect of cyclosporin is nephrotoxicity, which after renal transplantation may be difficult to distinguish from graft rejection. There is no consensus on the relation between cyclosporin concentrations in blood, plasma, or serum and the therapeutic or toxic effects of the drug. Interpretation of these data is complicated by methodological variables, including the choice of both the analytical technique and the sample matrix, as well as the clinical differentiation of allograft rejection from cyclosporin induced nephrotoxicity.¹

Currently two methods are available for measuring cyclosporin concentrations—high performance liquid chromatography² and radioimmunoassay.³ Most data have been produced with a radioimmunoassay kit manufactured by Sandoz Products Ltd. Values obtained using high performance liquid chromatography are substantially lower than those produced by the radioimmunoassay kit, since the antibody it uses cross reacts with several metabolites of cyclosporin.⁴

Renal allograft rejection may be differentiated from cyclosporin induced nephrotoxicity by histological criteria.^{5,7} A previous study, which defined these histological features,⁵ concluded that measurements of cyclosporin in blood samples collected at the time of allograft biopsy were of limited value; there was considerable overlap of blood concentrations between patients suffering nephrotoxicity and those with allograft rejection. The study did not, however, use predose blood concentrations, nor did it assess the cyclosporin concentrations to which the kidney had been exposed before the recognition of renal dysfunction. These latter measurements may be more relevant, since if there is a relation between cyclosporin concentrations and the clinical effects there is likely to be a temporal separation between the clinical manifestation of rejection or toxicity and the drug concentrations which precipitated the event. This study investigates the relation between the cause of renal graft dysfunction, using the previously published histological criteria, and trends in predose blood cyclosporin concentrations during the week before the episode of renal dysfunction using the Sandoz radioimmunoassay kit to measure the drug.

Patients and methods

Forty nine recipients of cadaveric renal allografts were studied (37 men, 12 women; mean age 46 (SD 14) years, range 19-69). Thirty two of the patients were given their first allograft, 13 their second, two their third, and two their fourth. Preoperatively patients received 1 g methylprednisolone intravenously and one oral dose of cyclosporin 8 mg/kg. Postoperatively they received oral cyclosporin twice daily, 16 mg/kg/day for three days and

Poisons Unit, Guy's Hospital, London SE1 9RT

D W HOLT, PHD, MRCPATH, principal biochemist

Department of Chemical Pathology, King's College Hospital, London SE5

J T MARSDEN, BSC, PHD, research biochemist

Department of Clinical Pharmacology, St Bartholomew's Hospital, London EC1

A JOHNSTON, BSC, head, analytical unit

Renal Unit, Dulwich Hospital, London SE22

M BEWICK, MCH, FRCS, consultant surgeon

D H TAUBE, BM, MRCP, consultant physician

Correspondence to: Dr Holt.

10 mg/kg/day for the next four days. Subsequently the cyclosporin dose was adjusted to maintain predose blood concentrations of the drug within a target range of 333-666 nmol/l (400-800 ng/ml). Prednisolone dose was maintained at 20 mg/day for one month, reducing to 15 mg/day for the next month and 10 mg/day thereafter.

All patients suffered either primary non-function or at least one episode of allograft dysfunction during the six months after transplantation. Biopsy samples of the allografts were obtained during these episodes and the diagnosis of allograft rejection or cyclosporin induced nephrotoxicity based on their histological appearance and the response to treatment. All biopsy samples were taken after an ultrasound examination of the allograft to exclude ureteric obstruction and to locate the kidney precisely.

Mean blood concentrations of cyclosporin (nmol/l) during seven days before biopsy in patients diagnosed as suffering episodes of graft rejection or cyclosporin induced nephrotoxicity.

Conversion: SI to traditional units—Cyclosporin: 1 nmol/l $\approx$ 1.2 ng/ml.

The histological features associated with allograft dysfunction due to rejection⁵ included a diffuse interstitial mononuclear cell infiltrate, interstitial oedema and haemorrhage, and an arterial wall mononuclear cell infiltrate. Graft rejection was treated with 3 daily 1 g boluses of methylprednisolone given intravenously and either an increase or no change in cyclosporin dosage. Patients with severe rejection received a course of antithymocyte globulin.

The diagnosis of cyclosporin induced nephrotoxicity was a histological one of exclusion; in the absence of the features of rejection nephrotoxicity was diagnosed and the cyclosporin dose reduced. If blood concentrations of cyclosporin were less than 333 nmol/l (400 ng/ml) the diagnosis was made only when other causes of allograft dysfunction, such as renal artery stenosis, had been excluded. In both groups of patients the diagnosis was sustained only if renal dysfunction was reversed during the 10 days after initiation of treatment for either rejection or toxicity.

Blood samples for cyclosporin measurement were drawn before the morning dose of cyclosporin; results were usually available within two days. Blood samples were collected daily during inpatient care and three times a week for the first three months after transplantation, reducing to weekly by six months. Retrospectively, blood concentrations of cyclosporin during the week before biopsy were assessed with respect to the cause of renal dysfunction.

Whole blood concentrations of cyclosporin were measured by radioimmunoassay (Sandoz). The within assay reproducibility (coefficient of variation) of the method at 416 nmol/l (500 ng/ml) was 5% (n=10), while the between assay value at this concentration was 8% (n=25); the limit of accurate measurement was 54 nmol/l (65 ng/ml). The laboratory was a participant in the United Kingdom cyclosporin quality assessment scheme.⁸

Results

Thirty six episodes of renal dysfunction were diagnosed as graft rejection and 40 as cyclosporin induced nephrotoxicity; the diagnosis was confirmed in all instances by the response to treatment. During the study period seven

of the 49 allografts were lost as a result of irreversible rejection and two patients died with functioning grafts. Overall the median time between transplantation and biopsy of the graft was two weeks. Rejection episodes occurred later (median five weeks) compared with a median of two weeks for the nephrotoxic episodes. A median of five blood samples (range two to seven) were collected during the week before biopsy.

The mean cyclosporin concentration during the seven days before biopsy in those patients suffering graft rejection was 392 (SD 206) nmol/l (471 (248) ng/ml) compared with 741 (305) nmol/l (891 (366) ng/ml) in the nephrotoxic group ($p < 0.0001$; Mann-Whitney U test). While the concentrations in these two groups overlapped considerably (figure), there was a significant association between average concentrations in excess of 666 nmol/l (800 ng/ml) and the incidence of nephrotoxicity—24 out of 40 episodes compared with only two out of 36 rejection episodes ($p < 0.0001$; χ^2 test). For both groups the mean cyclosporin concentrations on the day of biopsy were almost identical, the mean concentrations for the seven days before biopsy being 421 (SD 244) v 748 (394) nmol/l (506 (293) v 899 (473) ng/ml). The groups were less clearly separated, however, with 20 of the 40 episodes of nephrotoxicity and five of the 36 episodes of rejection being associated with concentrations exceeding 666 nmol/l (800 ng/ml; $p < 0.002$). Similarly, though the highest and lowest concentrations recorded during the seven days before biopsy were substantially different (table), the spread of results was very wide, 28 of the 40 nephrotoxic episodes and 11 of the 36 rejection episodes being associated with at least one concentration exceeding 666 nmol/l.

Highest and lowest blood cyclosporin concentrations (nmol/l) recorded during week before allograft biopsy in relation to diagnosis of renal dysfunction

	Rejection		Nephrotoxicity	
	Mean (SD)	Range	Mean (SD)	Range
Highest	554 (305)	141-1664	1017 (458)	228-1664
Lowest	275 (157)	50-805	457 (248)	122-1321

Conversion: SI to traditional units—Cyclosporin: 1 nmol/l $\approx$ 1.2 ng/ml.

During the seven days before biopsy there was a significant association between both the number of cyclosporin measurements above 666 nmol/l and the diagnosis of nephrotoxicity and the number of measurements below 333 nmol/l (400 ng/ml) and the diagnosis of graft rejection ($p < 0.002$; χ^2 test). In the seven patients who lost their allografts because of irreversible rejection the mean cyclosporin concentration during the seven days before the initial diagnosis of rejection was 292 (SD 163) nmol/l (351 (196) ng/ml). Only one of these patients had no concentrations lower than 333 nmol/l in this period, while in the remainder 22 of 24 measurements were below this value.

Discussion

The interpretation of blood or plasma cyclosporin concentrations is complicated by the methodological problems associated with their measurement. Despite its lack of specificity, a relation between radioimmunoassay measurements of cyclosporin and clinical events has been reported.

Our findings suggest an association between mean predose blood cyclosporin concentrations in the week before an episode of renal dysfunction and the diagnosis of graft rejection or cyclosporin induced nephrotoxicity; concentrations outside the range of 333-666 nmol/l (400-800 ng/ml) were of particular relevance. Mean concentrations above 666 nmol/l were associated with rejection in only two of 36 patients, while samples collected only on the day of biopsy, or reference to the highest or lowest recorded concentrations in the week before biopsy, gave a less clear cut separation of the two groups. There was an overlap between the concentrations associated with nephrotoxicity and rejection but several variables, such as prolonged warm and cold ischaemia times and machine perfusion time, may make allografts more susceptible to cyclosporin nephrotoxicity.¹² Such factors may, in part, explain the large number of nephrotoxic episodes associated with comparatively low mean cyclosporin concentrations.

There are considerable between subject differences in the pharmacokinetics of cyclosporin, affecting such important variables as systemic bioavailability, and several clinically important drug

interactions have been identified.¹³ These variables contribute to the poor correlation between dose and blood cyclosporin concentration,¹¹ making knowledge of the daily dose a poor predictor of the blood cyclosporin concentration. In our study between patient variation of cyclosporin concentrations was very large, with many measurements falling outside the target range. It is noteworthy, however, that half of the episodes of toxicity occurred within the first two weeks of transplantation, during which there was little opportunity to alter the cyclosporin dose in the light of blood concentration measurements, since a fixed dosage schedule was used for the first seven days. Possibly a more flexible dosage schedule, in association with a more rapid provision of blood cyclosporin results, would help to reduce the incidence of toxicity during this period.

Our data suggest that frequent measurements of the drug, especially during the first few weeks after transplantation, are a useful aid in planning alterations of dosage with the aim of avoiding the high mean concentrations associated with nephrotoxicity while ensuring concentrations consistent with the suppression of graft rejection; infrequent measurements, or samples collected only at the time of renal dysfunction, would be of less value. We conclude that, despite its lack of specificity for the parent compound, the radioimmunoassay used in this study produced results which were of clinical value, exposure to average predose blood concentrations outside the range of 333-666 nmol/l (400-800 ng/ml) being associated with an enhanced risk of graft rejection or nephrotoxicity.

We thank Drs F Dische and V Parsons for advice and encouragement. The financial support of Sandoz Products Ltd, UK, is also acknowledged.

References

- Holt DW. Clinical interpretation of cyclosporin measurements. In: Morris PJ, Tilney NL, eds. *Progress in transplantation*. Vol 3. Edinburgh: Churchill Livingstone, 1986:32-54.
- Carruthers SG, Freeman DJ, Koegler JC, et al. Simplified liquid chromatographic analysis for cyclosporin A, and comparison with radioimmunoassay. *Clin Chem* 1983;29:180-3.
- Donatch P, Abisch E, Homberger M, et al. A radioimmunoassay to measure cyclosporin A in plasma and serum samples. *J Immunoassay* 1981;2:19-32.
- Robinson WT, Schran HF, Barry EP. Methods to measure cyclosporin levels—high pressure liquid chromatography, radioimmunoassay and correlation. *Transplant Proc* 1983;15:2402-8.
- Taube DH, Gwyn Williams D, Hartley B, et al. Differentiation between allograft rejection and cyclosporin nephrotoxicity in renal-transplant recipients. *Lancet* 1985;ii:171-4.
- Farnsworth A, Hall MB, Ng ABP, et al. Renal biopsy morphology in renal transplantation. *Am J Surg Pathol* 1984;8:243-52.
- Sibley RK, Rynasiewicz J, Ferguson RM, et al. Morphology of cyclosporine nephrotoxicity and acute rejection in patients immunosuppressed with cyclosporine and prednisolone. *Surgery* 1983;94:225-34.
- Johnston A, Marsden JT, Holt DW. The United Kingdom cyclosporin quality assessment scheme. *Ther Drug Monit* 1986;8:200-4.
- Irshik E, Tilg H, Niederwieser D, Gastl G, Huber C, Margreiter R. Cyclosporin blood levels do correlate with clinical complications. *Lancet* 1984;ii:692-3.
- Yee GC, Kennedy MS, Deeg HJ, Leonard TM, Thomas ED, Storb R. Cyclosporin-associated renal dysfunction in marrow transplant recipients. *Transplant Proc* 1985;17(suppl 1):196-201.
- Rogerson ME, Marsden JT, Reid KE, Bewick M, Holt DW. Cyclosporin blood concentrations in the management of renal transplant recipients. *Transplantation* 1986;42:276-8.
- Canadian Multicentre Transplant Study Group. A randomised trial of cyclosporine in cadaveric renal transplantation. *N Engl J Med* 1983;309:809-15.
- Ptacekinski RJ, Venkataramanan R, Burckart GJ. Clinical pharmacokinetics of cyclosporin. *Clin Pharmacokinet* 1986;11:107-32.

(Accepted 28 July 1986)

Prolonged pregnancy: the management debate

LINDA CARDOZO, JOHN FYSH, J MALCOLM PEARCE

Abstract

A prospective trial was conducted to compare the effects of conservative management of prolonged pregnancy (conservative group) with routine induction of labour at 42 weeks' gestation (active group) in otherwise uncomplicated pregnancies. Of the 402 pregnancies studied, 207 (51%) were allocated to conservative management and 195 (49%) were allocated to have labour induced. The groups were well matched for age, parity, and smoking habits. One hundred and sixty six (80%) of the patients in the conservative group went into spontaneous labour. Of the remainder, two underwent elective caesarean section, 19 had labour induced because of clinical concern, and the remaining 20 had labour induced at the patient's own request. One hundred and twenty five (64%) of the patients in the planned active group underwent induction of labour. Of the remaining 70, 49 went into spontaneous labour and 21 (11%) asked that they should not have labour induced.

Comparison of the two groups showed no difference in the length of the first stage of labour but a trend towards an increased need for intervention for fetal distress ($p < 0.06$) in the active group. There were no differences in the length of the second stage, the need for intervention, or the mode of delivery. In terms

of Apgar scores the neonatal outcome was not significantly different between the two groups, but a greater proportion of the babies (15% v 8%) in the active group required intubation. Umbilical cord venous pH estimated in the last 183 consecutive deliveries in the study showed a significantly lower mean value in the active group ($p < 0.05$). There was no difference in birth weight between the two groups.

Two deaths occurred in the study. There was a stillbirth in the conservative group at 292 days after massive abruption, and one neonatal death in the active group owing to multiple congenital abnormalities.

The outcome for mother and baby in patients from both groups who went into spontaneous labour was generally good. The outcome for patients for whom conservative management was planned but induction became necessary was no different from that of patients who underwent planned induction at term. Thus from our results we can find no evidence to support the view that women with normal prolonged pregnancy should undergo routine induction of labour at 42 weeks' gestation.

Introduction

Now all other animals bring the time of pregnancy to an end in a uniform way; in other words, one single term of pregnancy is defined for each of them. But in the case of mankind alone of all animals the times are diverse; for pregnancy may be of 7 months' duration or of 8 months or of 9 and still more commonly of 10 (lunar) months, whilst some women go even into the eleventh month.

ARISTOTLE, *Works*. Vol II

As long ago as 399 BC Aristotle appreciated that the gestation period for human pregnancy varied considerably and that prolonged pregnancy was not uncommon. Postmaturity is now one of the commonest indications for inducing labour in many hospitals in

Departments of Obstetrics and Neonatology, King's College Hospital, London SE5

LINDA CARDOZO, MD, MRCOG, senior registrar

JOHN FYSH, MRCP, DCH, lecturer

J MALCOLM PEARCE, FRCS, MRCOG, research fellow

Correspondence to: Mr J M Pearce, Department of Obstetrics and Gynaecology, St George's Hospital Medical School, London SW17 0RE.