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Notes on Some Immunosuppressive Agents and Their Use in Human Liver Transplant Recipients

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In this presentation, three aspects of immunosuppressive treatment will be dealt with: first, recent experience with the use of cyclophosphamide in whole organ graft recipients; second, some interrelationships between the major immunosuppressive drugs and the liver; and finally, the immunosuppression used recently for hepatic transplant recipients.

General Experience with Cyclophosphamide

Cyclophosphamide has long been known to be an extremely effective immunosuppressant in rodents. In humans, the drug has been used extensively for cancer chemotherapy and for the treatment of autoimmune disorders. Several years ago cyclophosphamide was introduced by Santos and his associates into the field of clinical bone marrow grafting. There are probably the reasons why cyclophosphamide has not been applied to clinical whole organ transplantation until recently. First,

a few trials in human kidney transplantation almost a decade ago were complicated by severe drug toxicity. Second, cyclophosphamide was found not to prolong renal homograft survival in dogs. Although the latter findings were due to a species peculiarity, they were apparently extrapolated to the human situation.

For 16 months, cyclophosphamide has been used to replace azathioprine in a series of whole organ graft recipients in Denver [7]. Prednisone and antilymphocyte globulin (ALG) has been administered as previously described. The daily dose of cyclophosphamide tolerated without undue toxicity has been 0.5–1 mg/kg or approximately half of that usually tolerated of azathioprine. Using such doses, bone marrow depression was observed about as frequently as with the old triple drug regimen containing azathioprine. Other side effects from cyclophosphamide were rare and consisted mainly of gastrointestinal symptoms.

In renal homograft recipients, the results with cyclophosphamide have been very similar to those achieved previously with azathioprine. With a follow-up of 4 to 16 months, the kidney survival among 39 recipients of related grafts is 82% and the patient survival is 90%. In a group of 36 recipients of non-related cadaveric kidneys,

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73% have functioning grafts and 78% of the patients are alive. Approximately half of the latter patients were retransplanted for the second to fifth time, and in these cases, the kidney survival was only 59%. In contrast, the patients undergoing primary cadaveric renal transplantation have a kidney survival exceeding 80%.

Kidney recipients have been treated with cyclophosphamide rather than azathioprine for as long as 1½ years. However, it is our present practice to use cyclophosphamide for approximately 2 months and then switch to azathioprine. This will be described in the last section of this article.

Immunosuppression and the Liver

Hepatotoxicity is obviously an undesirable feature of any drug to be used in liver transplant recipients. Furthermore, if the activation or the degradation of the drug occurs mainly in the liver, this will have special implications in such patients.

Azathioprine may be hepatotoxic in humans. In controlled studies in animals, the usual signs of toxicity are rises in serum transaminases and the alkaline phosphatase [8], although in one study an increase in bilirubin was also noted [1]. In humans, azathioprine is by no means as hepatotoxic as in dogs. Yet, there have been a number of cases in which azathioprine may have played a role in causing liver injury after renal transplantation [9, 10]. Administration of the main metabolite of azathioprine, 6-mercaptopurine, has also been shown to cause liver damage, as for instance, when used in leukemic patients [4].

Although cyclophosphamide, given in very high doses, has been found to cause liver damage in rodents, patients have been treated with the drug for several years without evidence of significant liver toxicity [5]. In the Denver kidney transplant

recipients, the incidence of liver dysfunction was similar whether azathioprine or cyclophosphamide was used [7], but the numerous factors involved make an evaluation of the effect of the individual drugs difficult.

Corticosteroids, including prednisone, may cause hepatomegaly and fatty degeneration of the liver in animals, but the incidence of this complication in humans under prolonged treatment is difficult to determine. There is no evidence that ALG has a significant hepatotoxic effect [2].

The reverse question concerning the effect of the liver on the metabolism of immunosuppressive drugs, might also be of consequence in hepatic transplant recipients. The way in which this factor could influence the daily doses of azathioprine in both dogs and man has been speculated upon [6, 9]; the same considerations apply to cyclophosphamide. Cyclophosphamide becomes an active immunosuppressive agent first after an *in vivo* activation process in which the liver plays an important role [3]. Consequently, the drug might become less activated and thus decreasingly immunosuppressive if the liver function deteriorates. To date, there is no direct evidence from experience with hepatic transplantation that this occurs, but the possibility will have to be borne in mind in future cases.

Alternatively, since the degradation and detoxification of azathioprine, cyclophosphamide, and prednisone occurs mainly in the liver, it could be suggested that the effect of the drugs would become enhanced with liver dysfunction. In fact, some evidence has been adduced that renal transplant recipients require reduced immunosuppression, particularly of azathioprine, when afflicted with severe liver dysfunction. However, the practical implications of the described interactions between the im-

Immunosuppressive drugs and the liver for the care of the hepatic transplant recipient remain obscure.

Present Practices of Immunosuppression

Immunosuppression in hepatic transplant recipients has been founded on experience accumulated in large series of renal transplant patients. Before 1966, treatment was with azathioprine and prednisone and no long term survivals were obtained.

In the first triple drug regimen introduced in 1966, azathioprine was given continuously in the maximum dose tolerated without leukopenia, the prednisone dose was adjusted according to the vigor of rejection, and ALG was given in a standard dose for the first 2-4 postoperative months. Intravenous Actinomycin C and local graft irradiation were occasionally used as supplemental immunosuppressive measures during homograft rejection.

In many cases, the course of ALG was prolonged to several months or even years. In addition, the practice was introduced of giving large doses of intravenous corticosteroids, first prednisone and later methylprednisone, for rejection or sometimes prophylactically. With this program, one year survival of 25% to 30% was achieved from 1967 to 1970.

In the last 16 months, azathioprine has been replaced with cyclophosphamide during the initial postoperative treatment. In the

first liver transplant patients on this regimen, the cyclophosphamide was continued for 9 months. More recently, the protocol has been adjusted in that cyclophosphamide is now given only for the first 1 to 2 months, following which a change is made to azathioprine. The rationale for such a successive use of two cytotoxic agents lies in their dissimilarity of structure and of action. Cyclophosphamide is an alkylating agent and azathioprine is a purine analogue. The two drugs are thought to act at different stages of the cell cycle and might thus have a reinforcing effect when given serially. The consecutive use of drugs in order to achieve "longitudinal synergism" in contrast to the synergism of simultaneously administered agents is common in cancer chemotherapy. With this new practice, we have limited ALG to the first few postoperative months.

A total of 15 recipients of orthotopic hepatic grafts have now been treated with a drug regimen including cyclophosphamide. Six of the patients are alive and well with a follow-up of 2-16 months.

Summary

Recent experience with the use of cyclophosphamide in whole organ graft recipients is described. The interrelationship between the major immunosuppressive drugs and the liver is discussed. Recent practice for clinical immunosuppression is outlined.

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