

Plasma Levels of Leukotriene B4 During Hepatic Allograft Rejection

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At the present time, rejection is the most frequent cause of graft dysfunction in liver transplantation. Differential diagnosis between this and other possible causes of dysfunction—preservation injury, vascular, biliary, viral—may well be difficult, as the clinical and analytical findings are often similar; moreover, no markers specific to rejection are available, and histological studies are necessary for a definitive diagnosis. For this reason, markers indicating activity within the immune system need to be established so as to provide a more specific means of distinguishing rejection from other causes of graft dysfunction.

The immune response to an allograft is complex, and the intricate mechanisms regulating it are still not entirely understood. Nevertheless, several specialists have drawn connections among changes in the lymphocyte subpopulations,^{1,2} rises in the interleukin-2 levels, expression of the interleukin-2 receptor,^{3,4} and alteration in the expression of antigens belonging to class II in the greater complex of histocompatibility, with rejection of the allograft.⁵

Leukotriene B4 (LTB4) is a derivative of the metabolism of arachidonic acid via 5-lipoxygenase, whose in vitro behaviour is to encourage rejection by favoring leukocyte aggregation, proliferation of T lymphocytes, interleukin-1 and -2 secretion, and the development of "natural killer" cell subpopulations.⁶⁻⁸ This study examines the role of LTB4 in mediating the immune response to the hepatic allograft in order to assess its usefulness in early diagnosis of rejection.

MATERIALS AND METHODS

In this study we performed orthotopic liver transplant on five mongrel dogs weighing between 20 and 30 kg. We followed the technique described by Starzl et al,⁹⁻¹¹ with some modifications. For preservation, Euro-Collins solution was used at 4°C, the cold ischemia time being 87 ± 21 minutes.

In the anhepatic phase, spontaneous femoro-porto-jugular by-pass was performed. After the portal venous system had been revascularized, end-to-end arterial anastomosis was performed between the celiac axis of the donor and recipient. In two out of five animals this anastomosis was performed end to side between the hepatic artery of the donor and the exit of the celiac axis in the recipient. Biliary reconstruction was carried out by means of cholecystoduodenostomy. In all cases, there was autotransfusion of a unit of blood extracted a week before the operation. None of the animals were given immunosuppressives. Percutaneous liver biopsy was performed systematically on the fourth postoperative day.

Blood samples were taken at the following stages during the study: one week before surgery (basal), after endotracheal intubation before laparotomy, 8 hours after revascularization of the graft through the portal veins, and every day for the rest of the postoperative period. At each stage, the levels of the following were noted: glutamic-oxaloacetic transaminase (GOT), glutamic-pyruvic transaminase (GPT), alkaline phosphatase, gamma-glutamic transpeptidase (gamma-GT), bilirubin, and LTB₄.

Levels of LTB₄ were calculated by radioimmunoassay. Antibody anti-LTB₄ was obtained from Advanced Magnetic Inc. (Cambridge, Mass).

The results were expressed as mean \pm SEM. The mean of the five basal dogs were used as control parameters. Observations were compared using Student's t test for paired samples. The level of significance was set at $P < .05$.

RESULTS

The five animals survived for an average of 6.6 days (range 4 to 8). In all cases, anatomical and pathological examinations revealed the presence of major rejection. In two animals there were clear anatomical signs of rejection on the fourth day. Both animals survived fewer than 6 days.

LTB₄ levels in the five dogs showed a variable development pattern. Nonetheless, our findings showed a tendency for the mean levels of this parameter to increase as the postoperative period progressed (Fig 1). A significant difference exists between the base level and the reading at 24 ($P < .001$), 48 ($P < .001$), and 72 ($P < .01$) hours before decease. The other parameters investigated are enumerated in Table 1.

DISCUSSION

This study analyses for the first time the behaviour (in vivo) of LTB₄ in a model of hepatic rejection without immunosuppression. The findings described here confirm the pro-rejection character attributed to this substance by in vitro studies.⁶⁻⁸ This is a new step toward gaining an understanding of the intricate mechanisms of immunological regulation. Our work confirms a progressive rise in LTB₄ levels as rejection becomes established.

This parameter rises earlier than the other parameters generally used to evaluate rejection (Table 1), which means that it could be used as a specific and early marker for rejection.

The inhibition of the synthesis of this compound by blocking the metabolism of arachidonic acid via 5-lipoxygenase has been the subject of previous studies,¹²⁻¹⁴ and may well offer new therapeutic alternatives in the prevention and treatment of rejection.

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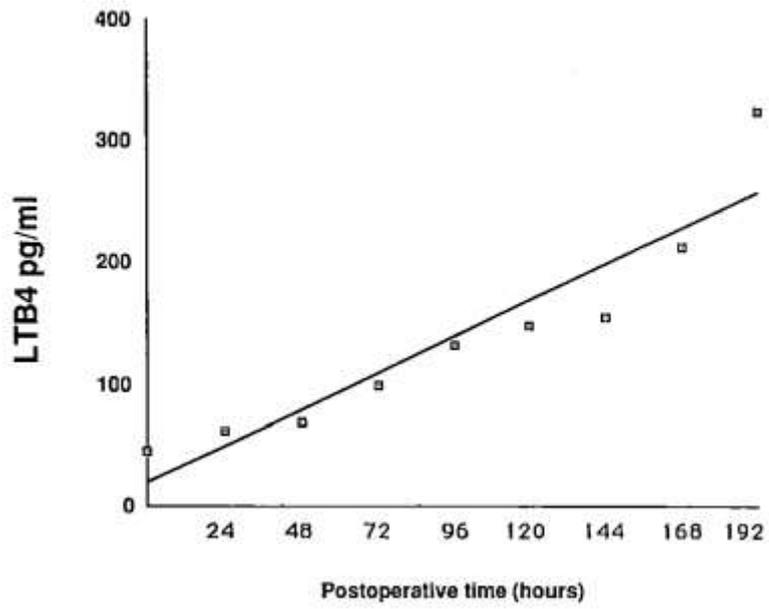


Figure 1. Mean trend of LTB4

Table 1. Mean Values of Hepatic Function Parameters				
	Basal	Day 1	Day 4	Day 7
LTB4	45.4	81.9	131.5	212.7
GOT	14	1722	106	493
Alkaline P	54	79	825	6671
Bilirubin	0.18	0.50	1.31	1.61