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HLA compatibility and different features of liver allograft rejection

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Abstract. The influence of human leukocyte antigen (HLA) on acute liver allograft rejection was investigated in 48 adult patients. The diagnosis of rejection was always based on the full triad of histological findings, clinical signs, and the required antirejection treatment. Sixty-two percent of the patients closely observed for 6 months postoperatively revealed acute rejection within the first 3 weeks, mostly on days 7-11. HLA compatibility was not observed to have any significant influence on the incidence of acute rejection. However, different histological and clinical features were revealed in conjunction with DR compatibility. Patients without DR compatibility showed a type of rejection with fever and increase of bilirubin, frequently associated with cholestasis and cholangitis, which sometimes persisted for weeks. Patients with 1 DR compatibility showed a predominant increase of transaminases, which was never associated with cholangitis. The conjunction of different DR compatibilities and various clinical signs may indicate possible pathways from immunological assault to the clinical appearance of acute rejection. A knowledge of a patient's individual compatibility and an expectation of certain rejection patterns may lead to earlier and more reliable diagnosis and treatment.

Key words: HLA compatibility - Rejection - Liver transplantation.

The influence of human leukocyte antigen (HLA) has been proven in clinical kidney transplantation. In contrast, HLA has failed to show any effect on liver transplantation. In spite of the large number of patients investigated recently [9], no beneficial or reasonable effect of HLA compatibility has been demonstrated on the long-term outcome after liver transplantation. However, long-term results are influenced by many factors, HLA being only one of these. Therefore, HLA compatibility may have some effect on liver transplantation, which cannot be shown by statistics concerning overall results, but can be important for some features of the postoperative course. As acute rejection is still one of the major problems of the early postoperative course, it is the aim of this study to investigate the relationship between the different HLA compatibilities and acute allograft rejection phenomena.

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Patients and methods

Eighty-one adult patients received a liver graft between 1 January 1986 and 31 July 1987 consecutively. Liver grafts were harvested according to our standard procedure [4], and orthotopic liver transplantation was performed using the bypass procedure during the anhepatic phase in all patients [12]. Postoperatively blood chemistry, including hematologic parameters, bilirubin and cholestatic enzymes, clotting factors, transaminases and bile volume, were monitored daily and if clinically required, several times. Liver biopsy specimens were taken before and after harvesting, 1-2 h after reperfusion, and routinely on postoperative days 3, 7, 14, 21, 28; if clinically required, e.g., suspicion of acute rejection or other disturbances, further specimens were taken. Standard immunosuppression consisted of cyclosporin A and low-dose steroids. Cyclosporin A dosages were adjusted to blood levels (500-700 ng/ml, whole-blood RIA, polyspecific). In patients with poor graft function or in critical condition, individualized immunosuppression with different drugs including antilymphocytic sera have been used [11].

A diagnosis of acute rejection was based on both histological and clinical findings. Some cases with very slight changes in these parameters, not requiring treatment were considered nonrejection cases. Thus, for establishing a diagnosis of acute rejection, the following full triad of parameters was necessary: (a) histological findings, (b) clinical findings, and (c) treatment required.

Table 1. Histological "Hannover" classification of acute liver allograft rejection

Severity grade	A-0	A-0-1	A-1	A-2	A-3
General characteristics	No evidence of rejection	Consistent with rejection, but nondiagnostic	Mild acute rejection	Moderate acute rejection	Severe acute rejection
Infiltrates	No	Slight	Slight	More pronounced	Marked
- Characteristic		Mixed	Mononuclear; predominantly lymphocytic; partially mixed	Mixed; predominantly mononuclear	Mixed; predominantly mononuclear
- Location		Portal	Portal and less parenchymal	Portal and less parenchymal	Portal and parenchymal
Parenchyma	No	No	Degenerative changes up to necrosis	Degenerative changes	Pronounced degenerative changes partly bridging necrosis
- Retrogressive changes (in % of all hepatocytes)			(less than 10%)	Focal nonbridging necrosis (10%-30%)	(more than 30%)
Endothelialitis	No	No	+	+	+
- Location			Portal or/and central	Portal and central	Portal and central
Bile duct damage	No	Less than 50%	More than 50%	More than 50%	More than 50%

Histological signs of acute rejection were classified into five different grades of severity according to the Hannover classification [5-7]; see Table 1. A diagnosis of cholangitis was established according to general histopathological criteria [17]. No cases of purulent cholangitis were observed in this study.

Clinical findings indicating acute rejection were considered to be an increase in transaminases and GLDH, bilirubin, cholestatic enzymes, the occurrence of fever, and deterioration of the patient's general condition.

Initial treatment of acute rejection always consisted of bolus application of methylprednisolone, 500 mg/day - mostly for several days [10]. If the initial treatment failed, the therapeutic strategy was changed to polyclonal or monoclonal antibodies. The first day of treatment was defined as the onset of rejection.

Patients were closely observed for at least 6 months postoperatively or until postoperative death. Ten of the 81 patients had to be excluded from this study: 3 patients showed irreversible initial nonfunction and died because a second graft was not available in time; in 2 patients severe hemorrhage caused intraoperative death (1 patient) due to non-viable donor organ and 1 patient due to multiple previous operations; in 2 patients vascular thrombosis and in 3 patients severe infection led to death in the first few postoperative days. In order to differentiate the early postoperative risk for acute rejection, the first 3 postoperative weeks were investigated separately. In 48 patients, complete HLA determination of the donor and recipient, including a relevant DR typing, was available.

Table 2. Incidence of acute rejection after liver transplantation

No. of patients	71	42
Follow-up period	3 weeks	6 months
Acute rejection	46	26
No rejection	25	16
Relative incidence of acute rejection (%)	65	62

Table 3. Incidence of acute rejection in patients with different HLA compatibilities

Locus of compatibility	A or B	DR	A or B	DR	A or B	DR	A or B	DR
Number of compatibilities	0	0	1-2	0	0	1	1-2	1
Patients	13		21		7		7	
Rejection	9		14		6		5	
No rejection	4		7		1		2	

Results

The frequency of acute rejection was 62% in all patients observed for 6 months postoperatively (see Table 2). In the larger group of patients followed for 3 weeks, the incidence of acute rejection was 65%. In all patients, the first rejection episode took place within the first 3 postoperative weeks, mostly on days 7-11.

Complete typing data for HLA-A, HLA-B and HLA-DR antigens were available for 34 patients with early acute rejection and for 14 patients without rejection during the first 6 months after liver transplantation. Thirteen of the transplanted patients had no compatibility either in class I or in class II antigens; 21 had 1 or 2 compatibilities in the A or B locus; 14 patients had 1 DR compatibility

Table 4. HLA compatibility and histological features of acute rejection

HLA compatibility	Severity grade			
	A_{0-1}	A_1	A_2	Cholangitis
No	1	5	3	2
1-2 A or B	2	8	4	5
1 DR	1	4	1	0
1-2 A or B and 1 DR	0	2	3	0

Table 5. HLA compatibility and clinical features of acute rejection

HLA compatibility	Clinical type		
	I	II	I+II
No	0	5	4
1-2 A or B	1	9	4
1 DR	5	0	1
1-2 A or B and 1 DR	2	1	2

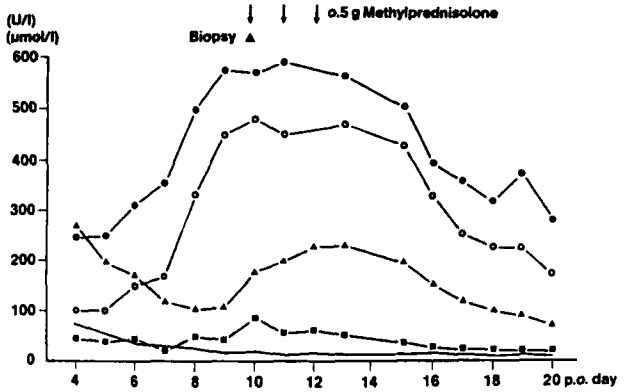

Fig. 1. Course of a clinical type-I rejection with predominant increase in transaminases, in this case associated by increase in cholestatic enzymes, but without fever or rise in bilirubin. ■ GOT, ▲ GPT, ● AP, O γ-GT, — bilirubin

with or without compatibility in the A or B locus. None of the patients showed complete DR compatibility or more than 2 compatibilities in the A or B locus. The data are given in detail in Table 3. The frequency of acute rejection is similar to the frequency of rejection-free courses in the corresponding groups and no significant influence of HLA compatibility on the incidence of acute rejection could be shown.

The histological feature of acute rejection is demonstrated in Table 4. The frequency of the different grades of severity is similar in all four groups of different HLA compatibilities, with A_1 as the

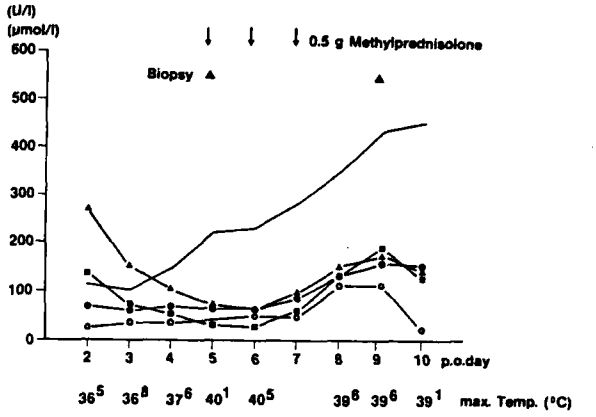

Fig. 2. Course of clinical type-II rejection with predominant increase in bilirubin and fever. ■ GOT, ▲ GPT, ● AP, O γ-GT, — bilirubin

most frequent severity grade, followed by A_2. None of the patients showed A_3 in the first biopsy specimen, indicating rejection phenomena. On the contrary, cholangitis frequently occurs in patients without any DR identity and is not seen in patients with 1 DR compatibility.

Investigation of the clinical features of acute rejection revealed two typical different types of clinical signs: type I, with a predominant increase in transaminases, sometimes combined with cholestasis; type II, with a predominant increase in bilirubin and temperature, frequently combined with cholestasis, which sometimes persisted for weeks.

Figures 1 and 2 show a typical example of each type. Table 5 demonstrates the distribution of these types in the four compatibility groups. Whereas the frequency of clinical types I and II is similar in patients with and without one or two compatibilities in class I antigens, a significant difference ($\chi^2 = 15.65$; statistical error $\alpha < 0.001$) is obvious between patients with and without one compatibility in class II antigens. Patients with 1 DR compatibility nearly always show clinical type-I rejection. In 3 cases with mixed clinical type I and II, the increase in transaminases was the predominant part. Patients without compatibility in antigen class-II antigens showed type II or mixed type I and II with type II as the predominant part. Table 6 gives detailed data on the patients according to their class II compatibility. Four of 11 patients with 1 DR compatibility (36.4%; 15.8-66.4%)[1] underwent further rejection episodes after an initial recovery. All recurrent rejections revealed the same clinical type as during the first rejection. In 5 patients (45.5%; 22.5-70.0%)[1] chronic or early irreversible rejection

[1] Confidence interval, calculated with a statistical error of 5% [16]

Table 6. Histological and clinical features of acute rejection in patients with 1 DR compatibility and with no DR compatibility

Patient	Severity grade	Clinical type	Combined cholangitis	Further course after first rejection
a Patients with 1 DR compatibility				
E.S.	A_1	I	–	Recurrent rejection, type I
M.M.	A_{0-1}	I	–	Acute rejection, type I
H.H.	A_2	II	–	→ Retransplantation
W.I.	A_1	I	–	No rejection
K.R.	A_1	I+II	–	Recurrent rejection, type I+II $\xrightarrow{ATG}$ retransplantation
E.L.	A_2	I	–	→ Retransplantation
W.U.	A_2	I	–	Irreversible rejection
B.D.	A_1	I	–	Recurrent rejection, type I
H.W.	A_1	I+II	–	No rejection
K.P.	A_2	I+II	–	Chronic rejection $\xrightarrow{ATG}$ retransplantation
P.L.	A_1	I	–	Recurrent rejection, type I
b Patients with no DR compatibility				
F.H.	A_1	II	–	No rejection
B.I.	A_2	II	+	Recurrent rejection, type II
R.M.	A_2	II	–	No rejection
T.A.	A_1	II	+	Recurrent rejection, type II + cholangitis
L.M.	A_1	II $\xrightarrow{later}$ II+I	–	Severe infection → death
E.H.	A_2	II	–	→ Retransplantation
J.J.	A_{0-1}	I+II	–	No rejection
K.S.	A_1	I+II	+	Recurrent rejection
G.S.	A_2	II	+	Recurrent rejection
K.D.	A_1	II	–	No rejection
L.G.	A_2	II	–	Death (arterial problem)
Z.K.	A_2	II	+	No rejection
V.M.	A_2	I+II	–	No rejection
H.M.	A_1	I+II	–	No rejection
P.A.	A_{0-1}	I+II	–	Chronic rejection → retransplantation
W.S.	A_1	I+II	+	Recurrent rejection, type II
R.U.	A_1	II	–	Recurrent rejection, type II
S.H.	A_1	II	–	No rejection
J.A.	A_1	II $\xrightarrow{later}$ I+II	+	Irreversible rejection
K.R.	A_1	II $\xrightarrow{later}$ I+II	–	ATG → irreversible rejection
E.P.	A_1	I+II	–	Persisting rejection → OKT3 → sepsis → death
M.C.	A_{0-1}	I+II	–	No rejection; cholestasis
R.K.	A_1	I	–	No rejection

developed, requiring retransplantation or leading to death. Only 2 patients had no further rejection episodes - either acute or chronic. Six of 23 patients without DR compatibility (26.1%; 12.8-46.6%)[1] had further rejection episodes, all of the same clinical type as during the first rejection. Four patients (17.4%; 7.2-36.8%)[1] showed irreversible rejection and a retransplantation was necessary.

Discussion

Results in liver grafting depend on many different factors. Donor assessment, harvesting techniques and preservation, as well as the circumstances of transplantation and postoperative surgical and non-surgical problems, all influence the clinical courses and the results. Several factors are of well-known relevance; the importance of others is as yet unclear. If one parameter is the most decisive towards causing the death of the patient (e.g., surgical complications or irreversible initial non-function of the graft), the importance of this parameter is obvious. In general, however, both the clinical courses and results are influenced by many factors. HLA compatibility is only one of these factors, and its importance may be obscured by other, more spectacular parameters. In particular, when looking at the long-term outcome without taking the postoperative clinical course of each investigated group

into consideration, the influence of HLA compatibility may not be detectable.

We therefore studied the individual postoperative courses, paying special attention to the rejection phenomena, particularly the types, grades of severity, and recurrence episodes. Within the first three weeks, 65% of the patients underwent acute rejection. To avoid the errors obtained when using the short observation period (e.g., the appearance too frequently of rejection-free courses) all patients surviving for 6 months or more were followed up during this time period. In this smaller group the rejection incidence was 62%. These incidence rates are very similar to the experience of other transplantation centers [8, 9, 10, 13] and indicate that the patient groups investigated are representative regarding acute rejection phenomena under the present immunosuppressive protocols.

This study did not reveal any relationship between the incidence of acute rejection and HLA compatibility, which correlates with other findings [1, 2, 14] and does not agree with the suggestion that HLA compatibility may reduce the incidence of acute rejection [9]. Of course, our results may be due to the small number of patients investigated and, in addition, to the fact that no more than two compatibilities in class I antigens and one compatibility in class II antigens were investigated. It could be that different rejection rates will be detected in larger or more compatible groups.

On the contrary, the acute-rejection feature shows quite obvious differences between the HLA compatibility groups in both histological and clinical findings. Whereas the frequency of the different grades of histological severity grades is similarly distributed in all compatibility groups, the phenomenon of cholangitis only occurs in patients who are completely DR-mismatched.

In contrast to class-II antigens, which also seem to play a major role in clinical features class-I antigens had a similar incidence in the different clinical types. The 1 DR compatibility group nearly always shows signs of clinical type I, whereas patients without DR compatibility show type II (mostly exclusively), which is sometimes combined with a minor expression of type I.

Both histological and clinical findings establish two certain patterns of acute rejection, depending on the DR compatibility: in patients with 1 DR compatibility, clinical type I is the typical feature, with a predominant increase in transaminases and sometimes with signs of cholestasis, but never combined with cholangitis. On the other hand, clinical type II is typical of acute rejection in patients without DR compatibility: fever, elevation of bilirubin and severe cholestasis, frequently combined with cholangitis, both of which sometimes persist for weeks. The dependence of the two types on DR compatibility is statistically significant, although total DR compatibility could not be tested.

As Table 6 shows, several patients had recurrent rejection episodes after successful treatment of the initial acute rejection. In all cases, the pattern of rejection was exactly the same as that of the first one in an individual patient. Therefore, rejection types can be considered to be independent of other factors, e.g., the toxic effects of Cy-A.

It is suggested that the de novo expression of MHC antigens after liver transplantation, particularly DR expression on the bile duct epithelium, will play a major role and may be the reason for the different patterns of rejection demonstrated in this study; this has been demonstrated even in clinically undisturbed courses as early as the 3rd postoperative day [15]. Furthermore, especially strong DR-expression on bile duct epithelium was found in conjunction with cholangitis [15]. Thus, complete DR incompatibility may provoke the pattern of rejection with severe cholestasis and cholangitis and, consecutively, increase of bilirubin in serum.

The frequency of recurrent, irreversible, acute and chronic rejection requiring retransplantation is demonstrated in Table 6. These data suggest that the prognosis for patients with 1 DR compatibility is worse in comparison to the patients that are completely DR mismatched. Although this contracting effect corresponds to the findings of other groups concerning the occurrence of vanishing bile duct syndrome [3] and the long-term outcome [9], we consider our findings due to the relatively small number of patients investigated for a long time period.

Thus, three conclusions concerning the pathophysiological and clinical aspects can be drawn:

1. As yet, no clear-cut differences in the frequency of rejection and the outcome of patients can be demonstrated after liver grafting according to the different histocompatibility grades. HLA compatibility is therefore not necessarily required for liver transplantation.

2. The differences in the type of rejection phenomena according to histocompatibility differences in class-II antigens indicate that histocompatibility may be clinically relevant in liver grafting. In particular, knowledge of the individual compatibility of a transplanted patient and the expectation of a certain pattern of acute rejection may improve early diagnosis and treatment, especially in cases of recurrent rejection.

3. The conjunction of different DR compatibilities and various clinical signs of rejection may indicate potential pathways from immunological assault to the clinical appearance of acute rejection.

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