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Orjinal Araştırma / Original Article

Parameters That May Indicate Early Postoperative Rejections in Patients with Liver Transplantation

Karaciğer Nakli Yapılan Hastalarda Postoperatif Erken Dönemdeki Rejeksiyonların Göstergesi Olabilecek Parametreler

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Abstract

Aim: We aimed to investigate the importance of inflammatory parameters in determining acute rejection in the early post-operative period in liver transplant patients.

Material and Method: When rejection was developed and after pulse steroid therapy, Preoperative, Hemoglobin, Neutrophil / Lymphocyte rate, AST / Lymphocyte rate, AST / Neutrophil rate, ALT / Lymphocyte rate, ALT / Neutrophil rate, CRP / Albumin rate, Glucose, tacrolimus and GRWR levels of patients who developed rejection in the early period after liver transplantation performed Yeditepe University Hospital in 2020 were measured. Preoperative and discharge values of the group without rejection were also evaluated.

Results: Acute rejection developed in the first one month in the early postoperative period at eight (23.5%) of 34 patients who underwent liver transplantation. It was observed that there were significant differences in terms of changes in ALT and AST values at different stages of treatment in clients who developed rejection (p=0.01). The preoperative albumin value of the patients who developed rejection was significantly lower than the patients who did not develop rejection (p=0.040). The difference among pre-transplantation CRP values was significant (p=0.035). In the multiple analyzes performed, the ratio of neutrophil/lymphocyte (p=0.026), AST/lymphocyte (p=0.003), ALT/lymphocyte (p<0.001) and ALT/neutrophil (p=0.003) at the rejection stage according to the pre-transplantation period was significant.

Conclusion: Acute rejection can occur days after transplantation and may lead to graft failure. The importance of parameters that supports the correct diagnosis in treatment is increasing day by day.

Keywords: Liver transplantation, rejection, immunosuppression

Öz

Amaç: Karaciğer nakli yapılan hastalardaki postoperatif erken dönemde gelişen akut rejeksiyonların belirlenmesinde enflamatuar parametrelerin önemini araştırmayı amaçladık.

Gereç ve Yöntem: 2020 yılında Yeditepe Üniversitesi Hastanesinde yapılan karaciğer nakilleri sonrası erken dönemde rejeksiyon gelişen hastaların, preoperatif, rejeksiyon geliştiğinde ve pulse steroid tedavi sonrası taburculuğundaki hemoglobin, Nötrofil/Lenfosit oranı, AST/Lenfosit oranı, AST/Nötrofil oranı, ALT/Lenfosit oranı, ALT/Nötrofil oranı , CRP/Albumin oranı, Glukoz, tacrolimus ve GRWR düzeylerine bakıldı. Rejeksiyon gelişmeyen grubun da preoperatif ve taburcu edildiğindeki değerlerine bakıldı.

Bulgular: Karaciğer nakli yapılan 34 hastadan sekizinde (%23,5) postoperatif erken dönemde ilk bir ayda akut rejeksiyon gelişti. Rejeksiyon gelişen hastalarda ALT ve AST değerlerinin tedavinin farklı aşamalarındaki değişimleri açışından anlamlı farklılıkların olduğu görüldü (p<0.005). Pre-transplantasyon ve taburculuk öncesi ölçülen hemoglobin, trombosit, nötrofil, lenfosit, ALT, AST ve serum glukoz değerleri arasında anlamlı farklılık tespit edilmedi (p>0.05). Rejeksiyon gelişen hastaların preoperatif albümin değeri, rejeksiyon gelişmeyen hastalardan anlamlı olarak daha düşüktü (p=0.040). Pre-transplantasyon CRP değerleri arasındaki fark anlamlı idi (p=0.035). Yapılan çoklu analizlerde, pre-transplantasyon dönemine göre rejeksiyon aşamasında nötrofil/lenfosit (p=0.026), AST/lenfosit (p=0.003), ALT/lenfosit (p<0.001) ve ALT/nötrofil (p=0.003) oranlarında anlamlı idi.

Sonuç: Akut rejeksiyon transplantasyondan günler sonra ortaya çıkabilen ve greft kaybı ile sonuçlanabilen bir tablodur. Tedavisinde doğru tanıyı destekleyecek parametrelerin önemi her geçen gün daha da artmaktadır.

Anahtar Kelime: Karaciğer transplantasyonu, rejeksiyon, immünsupresyon

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INTRODUCTION

Transplantation is the only treatment method for end-stage organ failure such as liver, heart, lungs, and pancreas. The method of the follow-up and treatment of these patients requires a multidisciplinary approach among related patients. One of the most important problems after liver transplantationis that an acute rejection picture occurs days later. T lymphocytes that respond to alloantigenes such as MHC molecules in vascular endothelial and parenchymal cells cause the development of an acute reaction. Activated T lymphocytes kill the graft cells directly by destroying them or by activating the inflammatory cells that cause necrosis and secreting cytokines. The effects of T lymphocytes in acute rejection can be reduced by immunosuppressive therapy. Therefore, early diagnosis and treatment of rejection is very significant.[1,2]

In our study, we aimed to investigate the importance of inflammatory parameters and their contribution to the early diagnosis and treatment process in patients who developed acute rejection after liver transplantation.

MATERIAL AND METHOD

The records of 34 patients who underwent liver transplantation between January and December 2020 in the Yeditepe University Organ Transplant Clinic were retrospectively analyzed after receiving the approval of the Ethics Committee of the Yeditepe University Faculty of Medicine (Clinical Research Ethics Committee Decision No: 1391). Signatures of all patients included in the study were obtained on the informed consent form. According to the results obtained, the patients were divided into two groups. The first group (Group I) included patients who developed acute rejection, while the second group (Group II) included clients who did not develop acute rejection. Hemoglobin, Neutrophil / Lymphocyte rate, AST/Lymphocyte rate, AST/Neutrophil rate, ALT/Lymphocyte rate, ALT/Neutrophil rate and CRP/Albumin ratios, glucose, tacrolimus levels were compared by statistical methods among the groups when were patients discharged preoperatively, rejection developed, and after pulse steroid therapy.

Statistical Analysis

In summarizing the data obtained from the study, descriptive statistics were tabulated as average±standard deviation or median, minimum and maximum, depending on the distribution for numerical variables. Categorical variables were summarized as numbers and percentages. The normality of numerical variables was controlled by Shapiro-Wilk, Kolmogorov-Smirnov and Anderson-Darling tests. In comparing two independent groups; Mann Whitney U test was used in cases where numerical variables did not show normal distribution. In comparing the differences between categorical variables according to groups, Pearson Chi-Square was used in 2×2 tables with expected cells of five and above, Fisher's ExactTest was used in tables with expected cells below five, while Fisher Freeman Haltontest was used in R×C tables where expected cells were below five. The Wilcoxon test was used in numerical variables that did not show normal distribution to evaluate the differences between tacrolimus (ng/mL) rejection and pre-discharge measurements. Friedman Test was used to evaluate the statistical differences between pre-transplantation, rejection and pre-discharge measurements. Durbin-Conover test was used to detect differences between measurements.

Statistical analyzes were made by "Jamovi project (2020), Jamovi (Version 1.6.16.0) [Computer Software] (Retrieved from https:// www.jamovi.org) and JASP (Version 0.14.1.0) (Retrieved from https: // jasp- stats.org) programs and the significance level was taken into account as 0.05 (p-value) in statistical analysis.

RESULTS

The average age of the patients (n=34) included in the study was 51.9±12.1 years. Male patients constituted the majority (61.8%) and median BMI (body mass index) was calculated as 26.9 kg/m². The groups were similar in terms of age, gender distribution and BMI (**Table 1**).

Table 1. Demographic characteristics of the patients in the groups

	All patients (n=34)	Patients who developed rejection (n=8)	Patients who did not developed rejection (n=26)	p
Age (year)	51.9±12.1 54 [22-72]	55.1±7.9 54.5 [45-68]	50.9±13.1 54 [22-72]	0,563*
Gender				
Male	21 (61.8)	5 (62.5)	16 (61.5)	0.999**
Female	13 (38.2)	3 (37.5)	10 (38.5)	
Height (cm)	165 [145-187]	163.5 [157-186]	166.5 [145-187]	0,858*
Weight (kg)	72 [46-104]	74 [60-100]	72 [46-104]	0,563*
BMI (kg/m²)	26.9 [17.7-37.7]	27.4 [24-32]	26.7 [17.7-37.7]	0,647*

Descriptive statistics were tabulated as average±standard deviation or median, minimum and maximum, depending on the distribution for numerical variables. Categorical variables were summarized as numbers and percentages..
*. Mann-Whitney U test used. **. Pearson Chi-Square, Fisher's Exact or Fisher Freeman Halton test used. BMI: Body mass index

As a result of the statistical evaluations, significant differences were observed in ALT and AST values in patients who developed rejection (Group I) in terms of changes at different stages of treatment ($p<0.001$ and $p=0.010$). In multiple analyzes, ALT and AST values at the rejection stage were significantly higher than pre-transplantation values. The average ALT 25 IU/mL and 44.5 IU/L AST values before transplantation, increased to average values that 190.5 IU / mL and 186.5 IU / mL at the rejection stage ($p <0.001$ and $p=0.001$). Significant decreases were observed in ALT and AST values at the discharge process compared to the rejection process($p <0.001$ and $p=0.003$). There were no significant differences in terms of other variables in terms of pre-discharge, rejection process and pre-transplantation change. Neutrophil and lymphocyte counts, ALT and CRP values before discharge were significantly higher in patients who did not develop rejection (Group II) compared to pre-transplantation values ($p<0.05$).

The preoperative albumin value of the patients with rejection (Group I) was significantly lower than the patients who did not developed rejection (Group II) (median value 3.2 g/dL etc. 4 g/dL, p=0.040).

While pre-transplantation CRP value was 11 mg/dL in patients with rejection (Group I), it was measured as 3.5 mg/dL in patients who did not developed rejection (Group II). The difference between them was significant (p=0.035). Neutrophil / lymphocyte, AST/lymphocyte, AST/neutrophil, ALT/lymphocyte, and ALT/neutrophil rates in patients with rejection (Group I) were found to show significant changes at different stages of the treatment process (p<0.005) (**Table 2**). In multiple analyzes, the rejection process compared to the pre-transplantation period when significant increase was present in neutrophil / lymphocyte (p=0.026), AST/lymphocyte (p=0.003), ALT/lymphocyte (p<0.001) and ALT/neutrophil (p=0.003) rates, significant decreases were found in AST / lymphocyte (p=0.001), AST/neutrophil (p<0.001), and ALT/lymphocyte (p=0.003) rates in the pre-discharge period compared to the rejection process (**Table 2**).

CRP/albumin and thrombocyte / lymphocyte rates did not show significant changes at different stages of treatment in patients with rejection (Group I) (p>0.05). In patients who did not developed rejection (Table II), ALT/lymphocyte and CRP/albumin rates were found to be significantly higher in the pre-discharge period compared to the values in the pre-transplant period (p=0.023 and p=0.016). It was observed that the AST/Neutrophil rate decreased before discharge (p=0.001).

When tacrolimus level was taken into account in patients with rejection (Group I) and who did not developed rejection (Group II), pre-discharge values were similar (p=0.714).

Table 2. Comparison of the treatment process of patients with and without rejection in terms of proportional laboratory values in the pre-transplantation, rejection and pre-discharge stages

	Patients who developed rejection (n=8)	Patients who did not developed rejection (n=26)	p***
Neutrophil / Lymphocyte rate			
Pre-transplantation	3.8 [1.8-10]	3.1 [1.3-9.3]	0.307
Rejection	8.2 [2.6-73.8]	[-]	
Discharge value	7.9 [3.5-38.2]	4.3 [1-15.3]	0.008
p	0.030*	0.521**	
AST/Lymphocyte rate			
Pre-transplantation	49.2 [16.5-257.6]	52.2 [12.3-112.1]	0.705
Rejection	194.8 [55.2-2187.5]	[-]	
Discharge value	41.2 [31.9-671.4]	31 [8.2-151.2]	0.053
p	0.010*	0.078**	
AST/Neutrophil rate			
Pre-transplantation	14 [3-82.5]	16.5 [3.1-54.3]	0.765
Rejection	20.1 [10.5-88.2]	[-]	
Discharge value	7 [1-33.8]	8 [1.3-28]	0.796
p	0.011*	0.001**	
ALT/Lymphocyte rate			
Pre-transplantation	28 [6.3-220.8]	44.9 [13-86.4]	0.618
Rejection	434.9 [108.4-2525]	[-]	
Discharge value	151.5 [80.9-1414.3]	50.1 [14.4-288.4]	0.002
p	0.002*	0.004**	
ALT/Neutrophil rate			
Pre-transplantation	10.1 [1.1-70.7]	11.9 [4.2-33]	0.253
Rejection	45.2 [20.6-110]	[-]	
Discharge value	29.5 [3.2-71.2]	13.9 [2.7-95.6]	0.177
p	0.021*	0.424**	
CRP/Albumin rate			
Pre-transplantation	3.2 [0.7-9.6]	1.2 [0.2-19.5]	0.253
Rejection	4.7 [1.3-30.5]	[-]	
Discharge value	3.8 [0.4-19.7]	3.8 [0.5-17.9]	0.984
p	0.197*	0.014**	
Platelet/Lymphocyte rate			
Pre-transplantation	69419.7 [34285.7-475949.4]	134440.7 [21311.5– 414457.8]	0.205
Rejection	195844.4 [76562.5-562500]	[-]	
Discharge value	158367.7 [121276.6-634482.8]	102274.9 [28571.4-389090.9]	0.031
p	0.223*	0.501**	
Tacrolimus (ng/mL)			
Pre-transplantation	8.7 [6.2-11.9]	[-]	
Discharge value	10.2 [3.1-12.6]	11,2 [3.7-18.9]	0.714
p	0.675**	N-a-N	

Descriptive statistics were tabulated as average ± standard deviation or median, minimum and maximum, depending on the distribution for numerical variables. *. Friedman Test used. **. Wilcoxon test used. ***. Mann-Whitney U test used. ALT: alanine aminotransferase, AST: aspartate aminotransferase, CRP: C-reactive protein, N-a-N: Not-A-Number

DISCUSSION

In liver transplantation, rejection is a common complication, especially in the first 12 months, and is associated with increased morbidity and mortality . The number of acute rejection attacks, histological severity, and low drug levels are seen as risk factors for graft loss.[3] Recently, many studies have been conducted on markers that play a role in the inflammatory process to predict postoperative events.[4] Similarly, there are studies on genomic markers alternative to invasive liver biopsy associated with many risks for diagnosis in acute cellular rejection.[5]

In a study evaluating the effect of hematocrit on the blood tacrolimus level, a significant positive correlation was found between the hematocrit rate and the tacrolimus rate. The hematocrit has a significant effect on the tacrolimus level. It is important to consider hematocrit levels in better dose adjustment for patients.[6,7] In a study in which hematopoietic stem cell transplantation was used and tacrolimus was used for graft-versus-host disease prophylaxis, it was reported that changes in blood tacrolimus concentration were significantly associated with hemoglobin levels, but not with changes in white blood cell and platelet count.[8] Hemoglobin values in our patients were 9.5 g/dL [8.4-11.6] (P=0.419). However, we believe that the hematocrit must be at a sufficient level to arrive an effective blood tacrolimus level.

Acute rejection is generally reported in patients with tacrolimus blood concentrations below 10 ng/mL.[9] In another study, a statistically significant (p=0.046) relationship was found between increasing tacrolimus blood concentrations in a 7-day period and a decrease in the risk of acute rejection. [10] In the experimental liver transplant rat model, it has been shown that tacrolimus has important effects on the acute rejection table on the 7th day.[11] In a study conducted, it was reported that the decrease in chronic rejection rates in many centers may be associated with effective immunosuppression therapies.[12] In our patients, we aimed to reach a drug level of 10 ng/ml in the blood within an average of 7 days by starting tacrolimus at a low dose of 0.5 mg on the postoperative day and giving it in increasing daily doses. When acute rejection developed, the average tacrolimus levels of the patients were 8.7 ng/mL [6.2-11.9] and (P=0.675).

It has been shown in national cohortt studies that the incidence of acute rejection is significantly lower in patients with liver transplantation with hepatocellular carcinoma compared to with benign end-stage liver disease. In the same study, parameters such as neutrophil/lymphocyte rate, monocyte/lymphocyte rate, thrombocyte/lymphocyte rate, aspartate aminotransferase/lymphocyte rate, C-reactive protein/albumin rate and fibrinogen level were examined.[13] In our study, neutrophil/lymphocyte (p=0.026) and AST/lymphocyte (p=0.003) were significant in those who developed rejection. However, CRP/albumin and thrombocyte/lymphocyte rates were not significant at different stages of treatment (p>0.05).

Feng et al; did not see a significant difference in associating low and normal graft-to-recipient weight rate (GRWR) with perioperative mortality, biliary complications, postoperative bleeding, and the risk of acute rejection.[14] No significant statistical relationship was found in our study, either.

We see that the limitations of this study are the lack of histopathological verification and the small sample size to support the diagnosis when we think that rejection has developed. We think that the diagnosis and treatment approach in acute rejection should be in the light of clinical, radiological, immunological and pathological data.

CONCLUSION

Liver transplantation is the most effective treatment for those with end-stage liver disease. However, acute rejection is still a major source of concern. The mechanisms underlying acute rejection remain uncertain. More research is needed on biochemical parameters that may be indicative of acute rejection.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study permit was obtained from Yeditepe University Clinical Research and Ethical Committee, 17/02/2021 No: 1391

Informed Consent: Written consent was obtained from all patients who participated in the study and their relatives.

Referee Evaluation Process: Externally peer-reviewed.

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