Changes in hematologic parameters associated with liver transplantation

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ABSTRACT

Objective: The changes in hematologic parameters of 40 orthotopic liver transplantation procedures for 37 patients performed between January 1994 through December 1995 at King Faisal Specialist Hospital and Research Centre, Liver Transplant Unit are retrospectively analyzed to determine the changes in hematologic parameters post-liver transplantation.

Methods: The following hematologic studies complete blood count, differentiate prothrombin time, partial thromboplastin time and fibrinogen level were retrospectively reviewed and statistically analyzed to evaluate the possible changes in hematologic parameters.

Results: The hemoglobin level and hematocrit ratio are corrected by the end of the first month post-transplantation. A fairly steady decline in platelet counts occurred throughout the operation and continued during the first week post operatively. A sharp rise of platelet counts is consistently noted at day 7 post orthotopic liver transplantation, reaching the normal range of 150-200 x 10^9/L by the 2nd week of transplantation. A sudden prolongation of the activated partial thromboplastin time occurred when the donor liver was reperfused. By the 2nd postoperative day, the activated partial thromboplastin time in most of the patients returned to normal. Unlike the activated partial thromboplastin time, the mean prothrombin time started to shorten by the 5th postoperative day.

Conclusion: Pronounced hemostatic abnormalities develop during orthotopic liver transplantation. The most prominent early effects of graft reperfusion on hemostasis, found in this study, were a drop in platelet counts. We also confirmed the observation that a sharp increase of platelet count is expected at the 7th post-operative day.

Keywords: Liver transplantation, hematologic parameters, coagulopathy, thrombocytopenia.


Hematological monitoring after liver transplantation is principally directed at the detection of blood loss, infection and bleeding diatheses. However, careful attention to the full blood count often yields valuable nuggets of information, ranging from the emergence of severe, albeit, unusual, complications like aplastic anemia and graft-versus-host disease, to diagnostic implications of the thrombocytopenia encountered much more commonly in the immediate post-transplant period. Orthotopic liver transplantation (OLT) is associated with serious bleeding problems, which often require the use of large amounts of blood products.1,5 Bleeding of surgical origin may be caused by disturbances of the hemostatic system, and blood loss is somewhat dependent on intraoperative deterioration of the hemostatic function.6,5 Despite the infusion of platelets, a decrease in platelet counts is one of the changes that are uniformly seen after recirculation of the donor liver.4,7 Although thrombocytopenia is a sign of disseminated intravascular coagulation (DIC), it is often seen as an isolated phenomenon. Many studies have examined the pathogenesis of the hemorrhagic diathesis in

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OLT. Results of several studies, however, are contradictory. The use of different operation techniques and management procedures and variations in assay methods made comparison of the results difficult. In the last few years, improvement in operative management, surgical techniques and improved graft preservation have played an important role in reducing the intraoperative blood loss. Although these factors have undoubtedly contributed to lowering intraoperative mortality, patients using more than 50 units of red blood cells are still not exceptional. Serious hemostatic disorders can still be found in patients undergoing OLT and they are often a diagnostic and therapeutic dilemma to many clinicians. The pre-existing hemostatic disorders in cirrhotic patients undergoing orthotopic liver transplantation, in combination with the specific intraoperative hemostatic changes, contribute largely to the complexity of hemostatic management of these patients. Dangerous defects seldom occur during most surgical procedures of the liver but they may become a threat during liver transplantation. Further investigations on the etiology of hemostatic deteriorations in orthotopic liver transplantation and application of recently developed specific and sensitive assays may give better insight into mechanisms underlying the hemostatic disorders in OLT, and provide a new theoretical basis for improvement in treatment.

**Methods.** Between January 1994 and December 1995, at King Faisal Specialist Hospital and Research Centre, 40 orthotopic liver transplantations were performed for 37 patients (23 males and 14 females) mainly suffering from liver cirrhosis secondary to viral hepatitis. The age ranged from 9 to 69 years (mean is 43 years). At harvest, the donor liver was flushed with University of Wisconsin (UW) solution, placed in a plastic bowl that contained clean solution, and then packed in ice. The mean ischemia time for the donor liver was 6/2 hours. Conventionally, liver transplantation procedures are divided into 3 stages. Stage one begins with the induction of anesthesia and ends with occlusion of the blood flow to the patient's liver. Stage 2 (anhepatic phase) continues until the donor liver is reperfused by the patient's circulating blood. In the current series, the anhepatic phase averaged 126 minutes. Stage 3 starts from the moment of re-circulation until closure of surgical incision. Removal of the diseased liver involves freeing it from adhesions and tying of multiple collateral vessels that may be present. The hepatic artery, portal vein, supra hepatic vena cava and infra hepatic vena cava are clamped. At this point, the diseased liver is removed and the recipient is anhepatic. The recipient is usually placed on venovenous bypass by cannulating the femoral vein and portal vein, and the blood is returned to the axillary vein by means of a centrifugal pump. The patient does not receive systemic anti-coagulation at any time during the liver transplantation procedure. The following hemostatic studies were performed: complete blood count (CBC) including platelet count, prothrombin time (PT), activated partial thromboplastin time (APTT) and fibrinogen level. In retrospect, the above mentioned coagulation values in addition to hematocrit (HCT), mean cell volume (MCV) and white blood cell count (WBC) differential were reviewed. The data was entered using Microsoft Excel, version 6.1 (PC-Compaq 486) and converted to JMP software, version 3.1 (Power Mac 7500/100). Various methods of descriptive statistics were used to analyze the data. Demographic characteristics and clinical features of the study group were described using frequency tables and univariate distribution parameters such as median and range.

**Results.** A total of 40 orthotopic liver transplantation procedures for 37 patients are retrospectively analyzed including 23 males and 14 females. The age ranged from 9 to 69 years with a mean of 43 years. The major indication for transplantation was liver cirrhosis secondary to hepatitis C. The mean values for hemoglobin, MCV, HCT, APTT, PT, plasma fibrinogen, platelet count, WBC, monocyte, lymphocyte, and neutrophil percentage at various times during and after liver transplantation up to one year are shown in Figures 1-6.

**Hemoglobin and red cell parameters.** The patients started with a borderline low normal hemoglobin level of 108 g/L and hematocrit ratio of 0.30, proceeding at the same level up to the end of the first month of transplantation then increasing to normal level of 120 g/L and hematocrit of more than 0.35 ratio (Figure 1). The MCV started with a high normal level (88 fl) at the time of transplantation, dropping very quickly by the 2nd day of transplantation into the average normal range of 82-84 fl and even dropping further to a level of 80 fl at the 2nd month post transplantation (Figure 2).

**Platelet counts.** The mean platelet counts were subnormal at the time of transplantation with an average of 100-120 x 10^9/L. A fairly steady decline in platelet counts occurred throughout the operation and continued during the 7-day period post operatively (Figure 3). The intraoperative reduction in platelet count occurred despite the infusion of a mean of approximately 10-15 units of platelets per patient. The mean value of platelet count during the 7 days post transplantation were at range of 65-79 x 10^9/L. At day 7 post OLT, a sharp rise of platelet count was consistently noted reaching the normal range of 150-200 x 10^9/L in the 2nd week of transplantation.
Figure 1 - Mean values for hemoglobin level and hematocrit ratio at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd month (M) through the end of the year.

Figure 4 - Mean values for activated partial thromboplastine time (APTT) and prothrombin time (PT) at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd month (M) through the end of the year.

Figure 2 - Mean values for mean cell volume (MCV) at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd (M) through the end of the year.

Figure 5 - Mean values for white blood cell count (WBC) at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd (M) through the end of the year.

Figure 3 - Mean values for platelet counts at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd month (M) through the end of the year.

Figure 6 - Mean values for neutrophils, polymorphs, lymphocytes, and monocyte percentage at pre-liver transplantation (Pre), day (D) 1 to day 7, week 2 (W) to week 4, and from the 2nd month (M) through the end of the year.
Activated partial thromboplastin time. A sudden and dramatic prolongation of the APTT occurred when the donor liver was reperfused. By the 2nd postoperative day, the APTT in most of the patients was normal. However, a small number of patients returned to normal by the 3rd day (Figure 4). To what extent the shortening of the APTT can be attributed to the surgical procedure or, alternatively, to the use of cryoprecipitate or fresh frozen plasma is difficult to assess.

Prothrombin time. As expected, the mean prothrombin time was prolonged. Unlike the APTT that, on the average, became normal by the 2nd postoperative day, the mean PT started to shorten by the 5th postoperative day (Figure 4). This finding suggests a factor VII abnormality, but specific assays for factor VII were not done in most of these cases and the information in some of the performed studies is not supportive of this conclusion.

Fibrinogen. The trend of declining fibrinogen values during transplantation and prompt recovery postoperatively parallels a pattern of fibrinogen values in patients undergoing any surgical procedure. Unfortunately, fibrinogen assay was not performed for most of the patients and by the end of the 2nd month, the test was not requested in most of the patients.

White blood cell count and differential. A moderate increase of the white blood cell count was noticed, reaching a maximum at day 4 and 5 posttransplantation and declining to normal values by the end of the first month (Figure 5). The high WBC was paralleled by an increase in the mature neutrophil polymorphs percentage and a relative decline of the lymphocytes and monocytes percent (Figure 6).

Discussion. OLT has become an accepted treatment for patients with chronic end-stage liver disease, but the procedure can be associated with massive intraoperative blood loss and considerable perioperative mortality and morbidity. Surgical bleeding is frequently complicated by severe coagulation disorders, which occur, especially during the anhepatic phase and after reperfusion of the graft. Increased fibrinolysis probably plays an important role during bleeding in OLT. Disseminated intravascular coagulation (DIC) has been implicated, but improvements in organ procurement and the preservation techniques seem to have reduced the intraoperative signs of DIC, which were frequently reported in the past. Abnormalities in hemostasis occur during the anhepatic period because of insufficient clearance of activated hemostatic and fibrinolytic factors. After reperfusion of the graft, hemostatic abnormalities become more severe, probably because of the prior ischemic damage to the graft. Pronounced hemostatic abnormalities developed during each of 40 consecutive OLT procedures, an outcome that is not surprising for those patients. All patients had sufficiently serious liver disease before the operation, and most of the plasmatic coagulation factors are derived from the liver. During any surgical procedure, coagulation changes occur. These alterations include reduction of coagulation factors and elevation of fibrinolytic split products, akin to the findings in DIC. Liver transplantation is a major surgical procedure actually consisting of 2 operations, hepatectomy and a subsequent period (about 2 hours) of anhepatic state followed by transplantation of a new liver. The new liver is washed, freed of blood after removal from the donor and was then preserved in UW solution for about 6 hours before being attached to the patient's circulation. As a result of the implantation of a cold liver, the patient's body temperature decreases to about 33°C (occasionally, one or 2 degrees lower). An additional factor that may affect the hemostatic system is the veno-venous bypass equipment that carries lower torso blood back to the heart during the anhepatic phase. Because no heparin or other systemic anti-coagulant is used, some platelets might conceivably be trapped in the pump, and certain coagulation factors might become activated on the walls of the pump. Theoretically, DIC may contribute to bleeding complications by 2 different mechanisms. First, consumption of coagulation factors, due to intravascular fibrin formation, which may lead to extremely low plasma levels of these coagulation factors. Second, intravascular thrombin formation during DIC may cause secondary activation of the fibrinolytic system. Both processes have been considered to be major causes of hemorrhage during the anhepatic period of OLT. In some experimental and clinical studies, a simultaneous reduction of platelet count, fibrinogen, clotting factors II, V, VII, VIII, and X and anti-thrombin III have been observed and interpreted at the time of DIC. Lack of hepatic clearance of activated coagulation factors and lack of synthesis of coagulation factors and inhibitors during the anhepatic period have been mentioned as factors provoking DIC. However, in other studies no clear signs of DIC were found and the role of DIC has been disputed. Although some authors have reported a beneficial effect of intravenous heparin, others could not influence the abnormalities by administration of heparin. Currently, the use of heparin is generally avoided as even small amounts of heparin may lead to severe hemorrhage. In recent reports of large series of OLT, only minor changes in coagulation profile were described during the anhepatic period. These observations do not support the theory of DIC in this period. The more stringent scheme for transfusion of plasma products

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that is currently followed by most anesthesiologists is regarded as an important factor in maintaining appropriate plasma levels of coagulation factors. Increased fibrinolysis, as measured by shortened whole blood clot lysis time, euglobulin clot lysis time,5,11,12 and by thrombelastography,5,13,14 however, has been found during the anhepatic period in many studies. Several authors have speculated that increased fibrinolytic activity is an important factor in the origin of hemorrhage.7,20,26 In general, the assessment of whether increased fibrinolytic activity is due to a primary process or secondary to DIC is hampered by difficulties in differentiation by the laboratory tests.20 Recently, specific enzyme immunoassays have been developed for the separate determination of fibrinogen degradation products in plasma.28 These assays appeared to be promising tools for studies on discrepancies in the equilibrium of the coagulation and fibrinolysis.28,29 Although these assays have not been used in orthotopic liver transplantation yet, they may give a better understanding of whether the observed fibrinolytic activity is of primary origin or secondary to DIC. Liver graft recipients tend to have reduced circulating platelet counts prior to transplantation. In patients with chronic liver disease as the majority of our patients, this is attributed to hypersplenism resulting from portal hypertension. Thrombocytopenia is also observed in acute liver failure. In one series the platelet count was less than 100 x 10^3/μl in 7 of 8 patients transplanted for viral or drug-induced acute liver failure.20 Our patients started with an average count of 108 x 10^3/μl and the circulating platelet count is further reduced during and after transplantation. This decrease is initially most apparent after reperfusion of the graft and the nadir is reached between 3 and 5 days after transplantation. A sharp increase in the platelet count is noted on day 6 post transplantation and afterward. The postulated theories for these changes include increased platelet consumption, hemodilution, immunologic reactions or sequestration of platelets in the reperfused liver graft.2 Since signs of DIC, characterized by simultaneous decrease in coagulation factors and the presence of fibrin degradation products are not always found in liver transplantation, the role of platelet sequestration in the liver graft has been the subject of speculation and further research.3,12 The fall in platelet count is probably not caused by hemodilution since no significant differences were found in hematocrit values of the corresponding blood sample. Activation of platelets by the artificial surfaces of the veno-venous bypass machines is also unlikely to play a role, as an identical drop in platelet count was found after orthotopic and heterotopic liver transplantation, in the latter, no veno-venous bypass is used.3,12 Specific accumulation of platelets in the newly grafted liver has been suggested by platelet labeling study.31 Further studies provided additional evidence that a systemic decrease in platelet count is due to a process of platelet accumulation in the liver graft since a sharp drop in platelet count was found in the venous outflow of the graft after reperfusion. The increased white blood cell count with increased mature neutrophil percentage in the first week of transplantation is most likely attributed to surgical manipulation of blood vessels, and the release of margined neutrophils to the central pool usually seen after most major surgical procedures.

In conclusion, our results are in concordance with the few previously reported studies. However, further detailed studies are mandated to determine the mechanisms of thrombocytopenia and hemostatic defects, associated with liver transplantation.

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