so called primary-non function (PNF) and the delayed-non-function (DNF). The incidence of the former is relatively small, being nowadays generally below 5%1. Yet, the occurrence of DNF is much more frequent, as it may develop in up to 10-50% of patients and is strictly related to the number of risk factors which identify marginal livers. According to current literature and the most recent guidelines, including those of the Italian Association for the Study of the Liver (A.I.S.F.), marginal livers are defined by the presence of at least one of the following risk factors: (1) a donor aged > 50 years; (2) a donor with hemodynamic instability or with a residence time in ICU greater than 5 days; (3) a donor with hypersodiemia; (4) a donor with HCV or HBV infection; and (5) a donor with macrovesicular steatosis present in > 25% of hepatocytes. The presence of steatosis involving less than 25% of hepatocytes is not considered sufficient to identify a marginal donor, although it may be associated with some risk of early or late graft failure. The reason is that the steatotic liver is characterized by a decreased tolerance to ischemia/reperfusion. It has been observed that the accumulation of fat in the hepatocytes and the increased cell volume cause an impairment of liver microcirculation. Steatosis is associated with decreased capability of ATP production and storage, with increased lipid peroxidation, and with increased release of tumor necrosis factor-α which is believed to be responsible of the lung damage possibly occurring after transplant. The assessment of the type and extent of steatosis requires liver biopsy, not usually indicated in healthy individual. In the transplant setting a precise assessment of steatosis is the prominent reason for performing a liver biopsy of the donor liver.

**Key Words:**
Steatosis, Liver transplantation, Graft, Biopsy.

**Introduction**

Early graft dysfunction represents an important concern in human liver transplantation, which may significantly affect morbidity and mortality. Early graft failure includes the
sis involving less than 25% of hepatocytes is not considered per se to be sufficient to identify a marginal donor, although it is clear that it may be associated with some risk of early or late graft failure. On the other hand, it is also clear that marginality is a concept in continuous evolution which will be likely implemented in the near future, as soon as long-term outcome data of transplants stratified for an increasing number of baseline data will become available. In addition, it should be considered that in a non negligible number of current donors two or more risk factors may coexist, which may result in an exaggerated chance of graft failures. In this regard, it is important to remind that the number of aged donors has dramatically increased in the last 10 years, due to the tremendous scarcity of donors compared to the actual needs. Donors older than 60 years currently represent more than 30% in Italy, compared to less than 10% in the early nineties. Current figures also show that up to 50-60% of potential donors may have at least two risk factors. This may have an important impact on transplant outcome, as it as has been estimated that the percent of patients developing DNF increases dramatically in the presence of multiple risk factors for marginality.

Apart from aged donors, the presence of some degree of steatosis in the donor is currently the most frequent cause for marginality. Indeed, graft with massive steatosis (>60%) should not be transplanted as their use determines a high risk of death. Even the use of livers with mild or moderate degrees of steatosis is associated with impaired early graft function in the first 3 postoperative days, as indicated by a greater peak of AST, little bile production and an increased risk of bleeding, and, more importantly, with decreased 2-year survival. The presence of pathological amount of fat in the liver favours the onset and/or worsens the severity of ischemic-preservation/reperfusion injury, which is known be associated with a significantly greater chance of PNF and DNF.

The reasons why the steatotic liver is characterized by a decreased tolerance to ischemia/reperfusion are manifold. First, the accumulation of fat in the cytoplasm of hepatocytes is associated with an increase in cell volume which causes an impairment of liver microcirculation. This has been observed in both human fatty donor livers and in experimental models of hepatic steatosis. The fatty infiltration is inversely related to the blood flow of the liver, eventually resulting in partial or complete obstruction of the hepatic sinusoidal space, thus favouring ischemic damage. On the other hand, steatosis is associated with decreased capability of ATP production and storage and with increased lipid peroxidation because of reduced antioxidant defenses and/or augmented production of ROS. In addition, steatotic livers have an increased release of tumor necrosis factor-α due to Kupffer cell dysfunction, which is believed to be responsible of the lung damage possibly occurring after transplant; they also display abnormal leucocyte adhesion and necrosis of endothelial cells, both further concurring to worsen the impairment of microcirculation.

It is interesting that recent experimental studies in the rat have shown the capability of ischemic preconditioning to improve the tolerance of fatty liver to hepatic ischemic-reperfusion injury. The use of ischemic preconditioning, which consists in determining brief periods of vascular occlusion prior to inducing ischemia/reperfusion, has already been shown to confer protection from tissue damage in normal rat liver, as well after hepatic resection of human liver, particularly in association with steatosis.

Fatty liver is a highly frequent condition in the general adult population, where is usually associated with lack of symptoms and a benign clinical course. Yet, a precise assessment of the type and extent of steatosis clearly requires histologic examination of liver tissue. This is not usually indicated in the healthy individual. In the transplant setting, however, a precise quantitative assessment of steatosis is the prominent reason for performing a liver biopsy of the donor liver. Unfortunately, current imaging methods are inaccurate in the quantitation of liver steatosis and do not distinguish between the microvesicular and the macrovesicular types. Recent studies, mostly performed in the setting of living donor liver transplantation, have shown that ultrasonography, TC scan and MR all display a reasonably good specificity for the diagnosis of steatosis, but also have an unacceptably low sensitivity compared to histology, with the only exception of cases of massive steato-
sion\textsuperscript{12}. All the imaging methods appear therefore to be inadequate for the proper assessment of mild-to-moderate degrees of steatosis, those which in practice are more frequent. Hence, liver biopsy currently remains the gold standard for the diagnosis and quantitation of steatosis\textsuperscript{13} and is considered to be mandatory in special settings\textsuperscript{14}. The Dionysos study, an Italian population based ultrasonographic study has shown that steatosis is present at ultrasonography in 58\% of adults\textsuperscript{15}. This figure appears to be much lower (18\%) in subjects who are alcohol abstainers and lean (with a Body Mass Index – BMI – lower than 25), but increases to 46\% in heavy drinkers (> 60 g of alcohol daily) and to 76\% in obese subjects. By extrapolating these numbers to the pool of potential donors, it appears that the vast majority of them are likely to exhibit at least some degree of steatosis. Therefore a liver biopsy to precisely assess whether the liver is suitable for grafting is mandatory in all donors with a history of moderate or severe alcohol assumption and with a BMI greater than 25. The same applies to donors who have an overt diabetes or the so-called “metabolic syndrome”, another condition often associated with steatosis due to the underlying insulin resistance. Liver biopsy is also recommended in donors without these risk factors, as steatosis may indeed be present in up to 20-30\% of this category, being under-diagnosed because of insensitivity of ultrasonography\textsuperscript{9}. This issue is important as even relatively low grades of macrovesicular steatosis (15\% of hepatocytes) have been reported to negatively impact on long-term outcome of transplant recipients when are associated with other parameters of marginality\textsuperscript{5}.

In conclusion, given the high prevalence of steatosis in the adult population, execution of a liver biopsy of the donor liver is generally recommended for the proper selection of the graft and the optimisation of the donor-recipient matching. This recommendation is hampered by the knowledge that other risk factors for marginality are common and often coexist with steatosis. This strategy will allow a better assessment of the long-term outcome and safety of the extended use of marginal grafts, which is expected to further increase in the near future. It will also hopefully allow to identify better donor-recipient matching criteria, based on evidence-based clinical and histological data.

References