Incidental Cholangiocarcinoma is Associated with Poor Outcome in Patients Transplanted for Hepatocellular Carcinoma

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Abstract

Background: Hepatocellular carcinoma (HCC) is a common malignancy in Asia for which orthotopic liver transplantation (OLT) offers curative treatment in selected patients. Incidental HCC-cholangiocarcinoma (HCC-CLC) has been associated with poor OLT outcomes.

Aims: To examine the prevalence of incidental HCC-CLC in liver explants from patients undergoing liver transplantation for HCC in our center and to examine its association with post-transplant disease recurrence and mortality.

Methods: Medical records of all liver transplant patients in our center were reviewed. The presence of HCC-CLC in liver explants was reviewed by experienced pathologists. Factors associated with recurrence of HCC post OLT and post-transplant mortality were analyzed. Survival in patients with incidental HCC-CLC was compared against those without.

Results: A total of 54 transplants were performed during the study period, of which 24 were transplanted for HCC. Mean age was 57 ± 9 years and 87.5% were male. Incidental HCC-CLC was documented in the explants of 4 patients (16.7%). Of these, 2 developed HCC recurrence and 3 died within 24 months. Over a median follow-up of 36 months, 3(12.5%) developed HCC recurrence and 8(33%) died. Factors associated with HCC recurrence and death were number and size of HCC lesions, microvascular invasion and incidental HCC-CLC. Age and AFP level were associated with recurrence but not with mortality. Mean survival of patients with HCC-CLC was poorer compared to those without.

Conclusions: Incidental HCC-CLC is not uncommon in OLT for HCC and is associated with tumor recurrence and poor survival.

Keywords: Predictors; Mortality; Recurrence; Intrahepatic; Incidental; Cholangiocarcinoma

Introduction

Hepatocellular carcinoma (HCC) is the third commonest cause of cancer-related death worldwide [1]. Orthotopic liver transplantation (OLT) can be regarded as the best treatment modality where both tumor as well as diseased liver are removed and replaced with a new, disease-free liver. In OLT for HCC, United Network for Organ Sharing (UNOS) system for allocating organs in the United States and many other liver transplant centers in the world select patients based on Milan criteria i.e. single HCC lesion ≤5 cm or 2-3 lesions ≤3 cm each with no vascular invasion or distant metastasis [2]. Another commonly adopted selection criteria is that proposed by the University of California San Francisco (UCSF) that is a slight expansion of Milan criteria to benefit more patients with HCC for OLT (single HCC lesion ≤6.5 cm and 2-3 lesions ≤4.5 cm each and cumulative diameter not more than 8 cm with no local or systemic spread) [3-5]. Proper selection of HCC patients with the Milan and UCSF criteria has improved the recurrence-free survival and survival rates at 5-years post liver transplant to approximately 83% and 75% respectively.

Outcome of OLT for HCC depends on recurrence of HCC and site of recurrence [4,5]. The reported rate of recurrence of HCC following OLT varies between 10-20% despite strict adherence to the proposed criteria [6,7]. Recognized factors associated with recurrence of HCC in transplanted livers include tumor size beyond Milan and UCSF criteria, vascular invasion on explanted liver, poor differentiation of HCC on histopathology, previous hepatectomy and pretransplant alpha-fetoprotein (AFP) level ≥1000 ng/ml [8-15]. Post-transplant recurrence of HCC is associated with reduced survival [8,9].

Several recent studies have observed that unrecognized mixed HCC-cholangiocarcinoma are associated with poor post-transplant outcomes with increased recurrence rates and reduced survival [16-22]. The aim of our study was to examine the prevalence of incidental HCC-cholangiocarcinoma in liver explants from patients undergoing liver transplantation for HCC in our center and to examine its association with post-transplant disease recurrence and mortality.

Methods

A retrospective review was performed from the medical records of all patients who underwent liver transplantation at the Singapore General Hospital (SGH) since the inception of the liver transplant program in February 2006 to June 2012. SGH is the largest tertiary hospital and one of two government-funded liver transplant centers in Singapore. Majority of OLT in SGH are deceased donor liver transplantations (DDLT). Our program adopts the UCSF selection criteria to enroll HCC patients for OLT (single tumor <6.5 cm, maximum of 3 total tumors with none >4.5 cm, and a cumulative tumor size <8 cm). Patients who did not fulfill criteria for liver transplant were excluded from the study.

The socio-demographic data of patients transplanted for HCC were evaluated (age, race, gender, etiology of liver disease and AFP level). Pre-transplant imaging that included ultrasound, quadriphasic CT scan and contrast enhanced dynamic MRI scan were reviewed for number of lesions, size of individual and largest lesion, cumulative size of the lesions, vascular invasion, portal vein thrombosis, tumor thrombosis and evidence of distant metastasis. UCSF criteria was used to enroll the patients for transplant. These features were compared with operative findings, gross and histo-pathological evaluation of the explanted liver. Presence of mixed HCC-cholangiocarcinoma in the explanted livers was evaluated by experienced pathologists. Treatment of HCC prior to transplant with liver resection and loco-regional therapeutic procedures such as percutaneous ethanol injection (PEI), trans-arterial chemoembolization (TACE), trans-arterial embolization (TAE), radiofrequency ablation (RFA) were documented.

After OLT, patients were prospectively monitored for recurrence of disease. Post-transplant HCC surveillance was performed via 3-monthly AFP and CT scan or ultrasound abdomen. In cases of HCC recurrence, the site of recurrence and time interval of recurrence after OLT were recorded. Type of immunosuppressant medications used and in cases with underlying viral etiology, hepatitis B DNA and hepatitis C RNA quantitative levels were documented before and after transplantation. In patients who died, the cause of death and time interval from OLT was recorded.

IBM SPSS Statistics (version 19) was used for data analyses. Descriptive analyses of demographic variables were performed. Continuous variables were expressed as mean and standard deviations (SD) and categorical variables as proportions. Continuous variables were compared using Student t test and categorical variables were compared by chi-square or Fisher exact test and 95% confidence intervals (CI) was calculated for each association. All p-values were two sided and considered as statistically significant if $p < 0.05$. Survival comparisons were performed using Kaplan Meier analysis and compared using the log-rank association.

Results

A total of 54 patients underwent liver transplantation at our center during the period of study. Of these, 24 (44.4%) were performed for HCC as the primary indication. In patients who underwent OLT for HCC, 19 (79.2%) had DDLT and 5 (20.8%) had living donor liver transplantation (LDLT). Mean age of patients was 57 ± 9 years and 87.5% were male. Underlying liver diseases leading to HCC were hepatitis B cirrhosis in 16 (66.7%), hepatitis C cirrhosis in 4 (16.6%) and cryptogenic cirrhosis in 4 (16.7%). None of the patients had underlying primary sclerosing cholangitis. Mean MELD score was a HCC-assigned score of 15 as the indication of transplantation was HCC rather than severity of disease in majority of these cases (Table 1).

Demographics	N=24
Age in years (mean ± SD)	57 ± 9
Male gender	21 (87.5%)
Etiology of liver disease	
Hepatitis B	16 (66.7%)
Hepatitis C	4 (16.7%)
Cryptogenic Cirrhosis	4 (16.7%)
Pre-transplant imaging features	
Number of lesions	2.8 ± 2.8
Size of largest lesion (cm)	3.4 ± 3.0
Cumulative size of lesions (cm)	3.6 ± 3.1
Portal vein thrombosis	2 (8.3%)
Pre-transplant loco-regional therapies	
TACE	20 (83%)
RFA	13 (54%)
Resection	6 (25%)
Type of liver transplant	
Deceased donor	18 (75%)
Living donor	6 (25%)
Features on explanted liver	
Microvascular invasion	3 (12.5%)
Associated cholangiocarcinoma	4 (16.7%)
Tumor thrombosis	1 (4.2%)
Outcome	
Recurrence of HCC	3 (12.5%)
Recurrence within 6 months	1 (4.2%)
Recurrence within 18 to 24 months	2 (8.4%)
Number of deaths	8 (33%)
Number of deaths with HCC recurrence	2 (8.3%)

Table 1: Characteristics of patients transplanted for hepatocellular carcinoma. Data presented as mean ± SD or n(%). HCC: Hepatocellular Carcinoma, UCSF: University of California San Francisco, TACE: Trans Arterial Chemoembolization, RFA: Radiofrequency Ablation.

Incidental HCC-cholangiocarcinoma (HCC-CLC) was detected in 4 (16.7%) of the explanted livers on histological examination. All were males, with a mean age of 55 ± 7 years. There were no significant

8	Sepsis and MODS	DDLT	38 months

Table 3: Causes of death after liver transplantation for hepatocellular carcinoma. HCC: Hepatocellular Carcinoma, LDLT: Living Donor Liver Transplantation, DDLT: Deceased Donor Liver Transplantation, MODS: Multi-Organ Dysfunction Syndrome.

	Alive (n=16)	**Died (n=8)**	**95% CI**	**p**
>3 HCC lesions	2 (12.5)	3 (37.5%)	1.29-174	<0.001
Largest tumor >6.5 cm	3 (18.8%)	5 (62.5%)	0.92-53.23	<0.001
Fulfilled UCSF criteria	15 (93.7%)	5 (62.5%)	0.04-3.79	<0.001
DDLT	14 (87.5%)	4 (50%)	1.29-174	<0.02
AFP ≥1000 IU/ml	1 (6.25%)	2 (25%)	0.38-17.45	0.72
HCC-CLC	2 (12.5%)	2 (25%)	0.009-1.32	<0.001
Microvascular invasion	1 (6.25%)	2 (25%)	0.01-2.64	<0.01

observed in HCC-CLC patients is consistent with reports from other centers [18-21]. In fact, post-transplantation cholangiocarcinoma recurrence has been reported to have a dismal recurrence-free survival of 0% at 3 years [21].

Other factors associated with recurrence of HCC in this study on univariate analysis were size of largest lesion more than 6.5 cm, number of lesions more than three and micro vascular invasion on explanted liver. These findings were consistent with previously reported risk factors for recurrence of HCC after OLT [10-15]. In this series, our numbers were too low to calculate meaningful multivariate analysis 95% confidence interval. Many studies have also suggested that aggressiveness of HCC (as evident by poor differentiation of tumor on histopathology) are associated with increased recurrence. [10-15]. However, all 3 cases of recurrence in our study had well-differentiated HCC and this difference may be due to small number of HCC recurrence cases in this study. Contrary to reports that a pre-transplant alpha-fetoprotein (AFP) level of 1000 ng/ml or more is a predictor of recurrence, all of our cases of HCC recurrence had AFP level ≤1000 ng/ml [24,25]. In this series, all the recurrences of HCC occurred within two years of transplantation and none of the patients transplanted for HCC had recurrence beyond two years and similar observation was reported in other studies [10-15]. Therefore close surveillance for recurrence is recommended in the initial post transplantation period in order to initiate effective therapy earlier.

In our study, the median and disease-free survival at one and five years were 79%, 71%, and 96%, 88% respectively, which are similar to other reviews [10-15]. Pre-transplant evaluation of the HCC was by ultrasound, quadriphasic CT scan and thorax and dynamic contrast enhanced MRI scan. However despite advances in imaging techniques, there remains a discrepancy in pre-operative staging of tumor and actual number of tumor lesions in the explanted liver. In all the three patients with recurrence of HCC in this study, pre-operative imaging had underestimated the number, size and vascular invasion of the HCC. Disparity between pre-operative imaging and disease staging in the explanted liver has been reported in up to 20% of cases by Decaens et al. [5]; the authors suggest caution in expanding selection criteria based upon tumor size and number on pre-transplant imaging.

The immunosuppressant regime used in majority of our patients was a combination of prednisolone, tacrolimus and mycophenolate mofetyl (MMF). Sirolimus was used in only one patient because of significant renal insufficiency and low cell counts. Several studies in the literature has suggested that a mTOR inhibitor like sirolimus may have anti-neoplastic activity against HCC recurrence [26,27].

We acknowledge that the interpretation of the findings described in this retrospective single-center study is limited due to the small sample size and require further validation in a larger multi-center prospective

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