Observational Study Protocol

Study title	MODELING CANCER-SPECIFIC PROGNOSIS IN LIVER TRANSPLANTATION FOR HEPATOCELLULAR CARCINOMA (HCC) WITH PRE-TRANSPLANT RADIOLOGY ASSESSMENT AND ALPHA-FETOPROTEIN (α-FP) Version 1 (final), 2nd March 2016
Rationale	Liver transplantation (LT) is one of the curative treatment options for patients with HCC associated to chronic liver disease (cirrhosis). All current international guidelines (EALS J Hepatol. 2012; AASLD Hepatology 2011; APASL Hepatol Int 2010 JHS) recommend LT for HCC only within pre-defined criteria. The use of restrictive criteria to select patients affected by HCC for LT was originally proposed by our centre (N Engl J Med. 1996) with the Milan criteria. These criteria were based on pathologic assessment of number and size of the HCC nodules on the explanted liver. Subsequently, many authors proposed the expansion of such restrictive criteria (e.g. UCSF, Tumour Volume, Up-To-Seven etc.). All these attempts, based on different combinations of morphologic parameters, have been defined on the pathologic staging of the tumor made on the removed liver, namely after LT, once decision on treatment and treatment itself could not be changed. Over time, various attempts have also been made to correlate postoperative pathologic findings of HCC to preoperative clinical staging in order to anticipate decision-making. Should a reliable conversion algorithm correlating pre-operative staging with post-transplant prognosis be available a selection of LT candidates based on routine radiologic HCC findings would optimise survival patient survival and organ resource allocation. Although post-LT pathology / pre-LT radiology correlation have improved over time, significant biases still affect clinical assessment of HCC stage and no reliable protocols has entered clinical practice yet. In addition, robust evidence indicates that other biological markers of aggressiveness (such as α-FP levels and clinical response to bridge therapies) have to be added when evaluating pre-operative variables. Another important issue to be considered when dealing with LT for HCC is the striking progress in treating chronic viral infections (HCV and HBV) that has been achieved over the last few years. Due to the possibility of treating recurrence of viral infections in the transplanted liver, these conditions do not significantly affect mortality following LT as they did in the past.

	In the current scenario, the decreasing role of non-oncologic factors in survival of patients treated by LT for HCC enhances the need to define new **prognostic models oriented towards cancer-specific survival.** These models should include, besides conventional morphological parameters, also the response to bridge therapies delivered in the pre-transplant setting as well as other biologic markers commonly used in clinical practise such as α-FP levels. Although many studies have been conducted, prognostic calculators of cancer-specific survival for HCC patients undergoing an evaluation for LT are not yet available. Such models should be able to provide survival estimates based on pre-treatment oncologic variables. A tailored assessment of cancer-specific prognosis is strongly advocated also by regulatory authorities, to optimize the allocation/distribution criteria of the limited source of available organs. Therefore, these criteria could be useful in the daily practise of transplant Centres to define different priority levels within the waiting list for LT, both for patients with and without HCC.
Endpoints and clinical relevance	The main goal of the study is the **definition of a cancer-specific prognostic model based on pre-operative features** (radiologic staging and α-FP levels) of a wide population of patients who underwent LT for HCC. Considering the competitive risk of cancer-specific mortality and death due to other causes, we aim to **redefine the Up-To-Seven criteria**, as they were developed by our centre on the base of pathologic staging (Lancet Oncology 2009). Primary endpoints: besides conventional outcome endpoints, the study aims to **develop and validate a prognostic calculator of recurrence rate and cancer-specific survival for HCC patients** undergoing evaluation for LT. The prognostic algorithm will be based on the **competitive risk analysis** of cancer-related recurrence and survival vs. non-cancer related outcome. Clinical relevance: the development of an on-line available prognostic calculator based on pre-operative oncologic factors would provide precise estimates of survival. This could help in the comparison of patients with different disease stage and comorbidities, by defining progressive priority levels to be applied to the waiting list for LT. The final goal is to **provide reliable survival estimates for patients at different disease stage and therefore with different priority**. This perspective may have great clinical impact as would allow the definition of different priority levels for LT in HCC, both to provide a prognosis-oriented treatment for each patient and to optimise the global outcome of the population of patients eligible for LT.
Study population	**Main inclusion criteria** are the following: - > 18-year-old patients with a definite pre-operative diagnosis of HCC (either radiologic or pathologic) who underwent their first LT in Italian tertiary centres for liver cancer (Milan-INT, Milan-Niguarda, Bologna Sant-Orsola

	Malpighi), over a period of at least 10 years after 2000; - Available clinical parameters of HCC (morphology and α-FP levels) both in the evaluation process and during their stay within the waiting list; - Evaluable clinical response to either medical or interventional anti-cancer treatments delivered prior to LT, by means of RECIST criteria (1.1 or mRECIST/EASL); - Fully described co-morbidities; - Completed oncologic follow-up (disease recurrence and/or cancer-specific death) and hepatologic/transplant follow-up (liver disease recurrence or onset of non-oncologic disease and/or death from any cause). These **inclusion criteria are applicable both to the training set** for the development of the cancer-specific prognostic model **and to any external and independent validation set.** The validation set should belong to non-Italian series, preferably among Institutions applying less restrictive criteria than those currently used in Italy. Also, the population of the validation set should be different in terms of aetiology of chronic liver disease underlying HCC, which in Italy mainly concerns HCV-related liver disease. Ideally, if the development of a cancer-specific prognostic model is achieved, the validation set may consist of an Eastern series of patients, in which HBV-related liver disease is prevalent.
Observational period	Retrospective analysis of a population of **patients who underwent LT for HCC from January 2000 to December 2013.**
Study population size	The expected number of patients in the 3 Italian centres is **more than 1000 cases**, 350 of which belonging to our series. If the training set will allow the development of a cancer-specific prognostic model, **at least 300 other cases will have to be added as a further set of external validation**, preferably belonging to an Eastern series (see above). Three Italian centres among the most active in LT and integrated treatment of HCC will take part in the study: Milan-INT (P.I. and study coordination), Milan-Niguarda, Bologna Sant-Orsola Malpighi. Contacts have been initiated with the Fudan University Centre in Shanghai, China, and the Montreal University Centre, Canada, and both are willing to provide the validation set should a reliable model emerge from the training phase of the study.
Informed consent	Considering the retrospective nature of the study, it is impossible to obtain an informed consent from all patients of the study populations. However, it should be mentioned that all patients, followed-up at dedicated clinics in each Centre, already agreed for their personal data to be used for scientific research and prognostic evaluation at the time of the pre-transplant evaluation and transplant enlistment, (see form MOD-PRO-P-03-CDTF-10). A copy of signed informed consent to transplant and to personal data management is filed and is available in each patient medical record belonging to the study and, in addition to the reference Organ Procurement Organization (in Italy Nord-Italia Transplant and Emilia Romagna Transplant Organization, Bologna).