Fondazione Policlinico Universitario Agostino Gemelli IRCCS
Università Cattolica del Sacro Cuore

Study Protocol

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Università Cattolica del Sacro Cuore

STUDY PROTOCOL

Surgical or Medical Treatment of Breast Cancer Liver Metastasis: a Multicenter Observational Study

ACRONYM: SurMed_BLCM

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Background and rationale

The liver represents the third most common site of breast cancer (BC) metastases behind the lymphatics and bone [1]. Breast cancer that spreads to the liver can include cancers with different cell characteristics, including hormone receptor-positive, HER2 -positive, or triple-negative breast cancers [2]. People of any background or age may develop liver metastasis. The process of liver metastasis consists of multiple steps and involves various factors from breast cancer cells and the liver microenvironment [3].

About 50% of people diagnosed with metastatic breast cancer develop liver metastases (BCLM), and 5% to 12% of these people develop liver metastases as the main site of breast cancer recurrence [4,5]. Survival in breast cancer with liver metastases (BCLM) typically does not exceed 8 months if left untreated.

The primary treatment for BCLM remains chemo-therapy and, more recently, targeted immunotherapy. Liver metastases from other primary tumors, such as colorectal cancers, are routinely resected as part of standard management. Breast cancer rarely develops isolated liver metastases because neo-plastic cells at this stage have often already reached systemic circulation with the possibility of further localization, unlike colon cancer, where the liver, through the portal system, is the first organ to be colonized. Therefore, the role of liver resection in BCLM remains controversial [6-8].

Hepatic resection and/or ablation was not associated with a survival advantage in some studies [9], whilst in other disclosed a remarkable improvement as compared to systemic treatment alone [10,11], thus indicating surgical intervention as suggested in highly selected patients with the goal of providing time off of systemic chemotherapy.

Our study thus aims to provide further insights in this complex and still unclear field of research.

OBJECTIVES

Primary Objective

Primary aim of the study is to compare the efficacy of liver resection vs. medical therapy alone in Breast Cancer Liver Metastasis (BCLM) patients.

Secondary Objectives

- To assess the recurrence rate after liver resection or medical treatment alone;
- To assess Recurrence free survival (RFS);
- To assess potential factors involved in both overall survival and RFS.

METHODS

Study design

Multicenter observational ambispective study.

Population

We will enroll all patients with BCLM underwent liver resection or medical treatment alone afferent to the Fondazione Policlinico Gemelli IRCSS of Rome (Italy), and other 8 International centers (as detailed in the attached document), following the inclusion and exclusion criteria hereafter detailed.

Study duration

The study will overall last 68 months since the approval of the present protocol by the sponsor center institutional review board (IRB), i.e. Fondazione Policlinico Gemelli IRCSS of Rome (Italy), and of all the satellite centers. The prospective enrolment of patients will last till the December 31, 2023 and follow-up phase will last for other 5 years, till December 31, 2028.

Study phases

The study foresees two distinct phases:

- **I phase (only retrospective phase)** → it will cover all patients underwent to either medical treatment or surgery for which all data are available, with a follow up of 5 years at March 31, 2023. We will retrieve all patients data from January 1994 till March 31, 2018. At the end of "phase I", we foresee an "ad interim" analysis.
- **II phase (ambispective)** → we will further include all patients retrieved from the archives between April 1, 2018 and the April 30, 2023, as well as all prospectively enrolled patients till December 31 2023, so that the last patient enrolled will end the 5-years follow-up phase within December 31, 2028.

Inclusion criteria

- Age >18 years old;
- Presence of BCLM;
- BCLM and extra-hepatic disease;
- Any treatment;
- Ability to understand and sign the informed consent.

Exclusion criteria

- Inability to provide the informed consent;
- Patients who underwent Best Supportive Care or palliation.

Variables and procedures

The following data will be retrieved from medical archives of each participating center for all retrospectively enrolled patient.

Prospectively enrolled patients' data will instead be collected in the related medical record and reported, alongside all other patients' data, in an electronic dataset protected by password and accessible only to the PI and the co-investigators, and to any other person authorized by PI.

In depth, the following data will be included:

- Anonymized ID;
- Demographic and anthropometric data (e.g., age, sex, BMI, etc..);
- Name of the participating center;
- ER, PR and HER2 status,
- Presence of BRCA1-2 mutation,
- Neoadjuvant-radiotherapy,
- Neoadjuvant endocrine therapy,
- Neoadjuvant-chemotherapy,
- Neoadjuvant-anti-HER2,
- Date of breast surgery,
- Breast tumor T and N,
- Adjuvant radiotherapy,

- Ad-endocrine therapy, Ad-chemotherapy, and Ad-anti-HER2,
- Date of liver metastasis diagnosis,
- Date of liver surgery,
- Liver surgery,
- Liver resection major/minor (Segmental or wedge resection or Left Lateral),
- R status,
- Number of liver metastasis,
- Size of the bigger lesion (mm),
- Distribution (uni or bilobar),
- Postoperative morbidity,
- Hormone receptor expression in the liver metasasis (ER+, PR+),
- Extra-hepatic disease,
- Site of Extra-hepatic disease,
- Mortality and cause;
- Date of death or last FU observation,
- Recurrences,
- Site of recurences,
- Date of recurrences.

ENDPOINTS

Primary endpoint

The primary endpoint is to analyze the efficacy of liver surgery as compared to medical therapy alone in terms of 5-years overall survival in patients with breast cancer who underwent to liver resection or medical therapy alone.

Secondary endpoints

- Recurrence rate;
- Post-operative mortality (in patients who underwent to surgical treatment).
- Post-operative complication (in patients who underwent to surgical treatment).
- To assess potential predictive factors of 5-years overall survival in patients with breast cancer who underwent to either liver resection or medical therapy alone.
- To assess recurrence free survival (RFS) of liver surgery as compared to medical therapy alone in terms of 5-years overall survival in patients with breast cancer who underwent to liver resection or medical therapy alone.
- To assess potential predictive factors of 5-years RFS in patients with breast cancer who underwent to either liver resection or medical therapy alone.

STATISTICAL ANALYSIS PLAN

Sample size calculation

The sample size was computed based on the assessment of an improvement in the OS of patients surgically treated as compared to systemic therapy alone.

Based on the evidence from Ruiz and colleagues [10], which reported a 5-years OS of 24% among women treated only with medical therapy and of 69% among those surgically treated, we hypothesize to obtain in our study sample an OS of 60% in the treated group and of 25% in the systemic therapy group.

As such, considering a treatment/control ratio of 0.15/0.85, a two-sided log-rank test with an overall sample size of 117 subjects (99 in the systemic therapy group and 18 in the surgical treatment group) achieves 90% power at a 0,05 significance level to detect a hazard ratio of 0,37 when the proportion of 5-year OS in the control group is 25%. The study will consider a time-window of 30 years of total enrolment (from 1994 to 2023) and a follow-up period of 5 years, for a total time of 35 years. The accrual pattern across time periods is uniform (all periods equal). We expect no subjects drop out both in the control group and treatment groups. The expect no. of events is of 96,5 in the control group and 14,3 in the treatment group. The sample size calculation was performed with PASS2022 v22.0.3 [12,13]. Such sample size estimation is consistent with the disease specific metastasis prevalence and on the no. of involved centers. In fact, we overall expect to include in both retrospective and

prospective phase 75-80 patients/center, for overall sample of 675-720 patients. Particularly, the retrospective phase, which will include patients till 2017 will consist of about 500 patients, already consistent with the sample size estimate provided, whilst the other will pertain to the period 2018-2023.

Statistical analysis

The sample will be described in its clinical and demographic characteristics by descriptive statistics techniques. In depth, qualitative data will be expressed as absolute and relative percentage frequency, whilst quantitative variables either by mean and standard deviation (SD) or median and interquartile range (IQR), as appropriate. To verify the Gaussian distribution of quantitative variables, the Shapiro-Wilk test will be applied.

Missing values, as <5% in all cases, were treated by *imputeR* package, using multiple imputation with Lasso Regression methods centered on the mean as for quantitative data, whilst classification trees for imputation by "rpartC" function, centered on the mode, i.e. most represented class object, were applied on qualitative data [14].

Between groups differences were analyzed by either the Chi squared test or the Fisher-Freeman-Halton test for qualitative data, whilst either the Student's t test or non-parametric Mann-Withney U test will be applied on quantitative data, as appropriate.

Differences between groups will be further exemplified by clustered violin plots or plots describing pre-post variation in individual groups. Such plots will be produced with the R packages "*ggplot2*", "*ggstatsplot*", "*ggpubr*", "*ggprism*", and "*ggsignif*" [15-19].

OS will be calculated from the date of treatment initiation through the date of death from any cause; subjects alive at the last follow-up will be censored. Overall survival and RFS probabilities at 5 years will be estimated using the Kaplan Meier method and compared by the log-rank test. The KM curve will also be reported. The analysis will be performed with the R package "*ggplot2*" and "*survminer*" R packages [15,20].

To assess potential prognostic factors of 5-year RFS and OS, both univariable and multivariable Cox proportional hazards regression models will be fitted, and hazard ratios (HR) and 95% confidence intervals (CI) consequently reported. The proportionality of the hazards will be assessed by visual inspection of the hazards and Schoenfeld residuals. In case of doubtful proportionality, Cox weighted regression models will be fitted. "*survival*" and "*coxphw*" R packages will be used for the whole analysis [21-24]. The predictors to be included in the multivariable model will be selected on the basis of univariable analysis ($p<0.05$ or suggestive, i.e. $0.05 \leq p < 0.10$) and expert opinion, also considering the possibility of overcoming the rule of 10 events per variable (EPV) [25,26]. Statistical significance will be set at a p-value <0.05. Suggestive p-values will be further reported ($0.05 \leq p < 0.10$). All analyses will be performed with R software version 4.2.0 (CRAN ®, R Core 2022) [27].

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