Research Article
Cirrhosis and Liver Failure

JOURNAL
OF HEPATOLOGY

Myosteatosis is closely associated with sarcopenia and significantly worse outcomes in patients with cirrhosis

Simone Di Cola[1], Gennaro D'Amico[2], Paolo Caraceni[3,4], Filippo Schepis[5], Simone Loredana[6], Pietro Lampertico[7,8], Pierluigi Toniutto[9], Silvia Martini[10], Sergio Maimone[11], Antonio Colecchia[12,13], Gianluca Svegliati Barone[14], Carlo Alessandria[15], Alessio Aghemo[16,17], Saveria Lory Crocè[18], Luigi Elio Adinolfi[19], Maria Rendina[20], Lucia Lapenna[1], Enrico Pompili[4], Giacomo Zaccherini[3,4], Dario Saltini[5], Massimo Iavarone[7], Giulia Tosetti[8], Carolina Martelletti[10], Veronica Nassisi[21], Alberto Ferrarese[12], Ilaria Giovo[15], Chiara Masetti[17], Nicola Pugliese[16,17], Michele Campigotto[18], Riccardo Nevola[19], Manuela Merli[1,*], for the EpatoSarco working group[‡]

Journal of Hepatology **2024**. vol. 81 | 641–650

Background & Aims: Sarcopenia and myosteatosis are common in patients with cirrhosis. This study aimed to determine the prevalence of these muscle changes, their interrelations and their prognostic impact over a 12-month period.

Methods: We conducted a prospective multicentre study involving 433 patients. Sarcopenia and myosteatosis were evaluated using computed tomography scans. The 1-year cumulative incidence of relevant events was assessed by competing risk analysis. We used a Fine-Gray model adjusted for known prognostic factors to evaluate the impact of sarcopenia and myosteatosis on mortality, hospitalization, and liver decompensation.

Results: At enrolment, 166 patients presented with isolated myosteatosis, 36 with isolated sarcopenia, 135 with combined sarcopenia and myosteatosis and 96 patients showed no muscle changes. The 1-year cumulative incidence of death in patients with either sarcopenia and myosteatosis (13.8%) or isolated myosteatosis (13.4%) was over twice that of patients without muscle changes (5.2%) or with isolated sarcopenia (5.6%). The adjusted sub-hazard ratio for death in patients with muscle changes was 1.36 (95% CI 0.99–1.86, $p = 0.058$). The cumulative incidence of hospitalization was significantly higher in patients with combined sarcopenia and myosteatosis than in patients without muscle changes (adjusted sub-hazard ratio 1.18, 95% CI 1.04–1.35). The cumulative incidence of liver decompensation was greater in patients with combined sarcopenia and myosteatosis ($p = 0.018$) and those with isolated sarcopenia ($p = 0.046$) than in patients without muscle changes. Lastly, we found a strong correlation of function tests and frailty scores with the presence of muscle changes.

Conclusions: Myosteatosis, whether alone or combined with sarcopenia, is highly prevalent in patients with cirrhosis and is associated with significantly worse outcomes. The prognostic role of sarcopenia should always be evaluated in relation to the presence of myosteatosis.

Introduction

Malnutrition is a frequent occurrence in patients with cirrhosis, with prevalence varying from 5% to 99% depending on the studied population and the diagnostic tools applied.[1–4] Several factors contribute to nutritional changes in these patients, including inadequate dietary intake, altered nutrient absorption and substrate utilisation modifications due to liver disease.[5] Moreover, a variety of disease-related acute and chronic complications can reduce a patient's ability to maintain their food intake[6] and/or increase their energy expenditure. Ultimately, malnutrition is associated with an increased risk of mortality, a higher incidence of complications related to portal hypertension and infections, and a longer hospital stay.[7–12]

Sarcopenia, a progressive and generalized loss of muscle mass and strength, is a significant feature of malnutrition in patients with cirrhosis.[13–16] Many studies have shown that sarcopenia is an independent predictor of morbidity and mortality,[11–13,17] and its inclusion in the evaluation of patients on liver transplantation (LT) waiting lists has been reported.[18] In obese patients with cirrhosis, loss of skeletal muscle can lead to a condition known as "sarcopenic obesity", which is associated with even worse prognosis and outcome.[17]

Muscle impairment in these patients is characterised not only by reduced muscle mass but also by changes in normal

* Corresponding author. Address: Department of Translational and Precision Medicine, Sapienza University of Rome, Rome, Italy.
E-mail address: manuela.merli@uniroma1.it (M. Merli).
‡ Collaborators list details are provided in Acknowledgements section.
https://doi.org/10.1016/j.jhep.2024.05.020

tissue structure and composition. Intramuscular fat accumulation, or myosteatosis, is known to reduce muscle quality.[17,19] Myosteatosis is more common in obese and elderly patients, but can also occur in normal weight individuals, reflecting a chronic inflammatory state.[20] Myosteatosis is associated with metabolic abnormalities and decreased muscle strength, and may lead to worsened median survival.[17,21]

Several studies have been conducted to investigate the role of sarcopenia and myosteatosis in the natural course of cirrhosis.[13,17,21] However, the available literature does not provide comprehensive information for various reasons. Firstly, most studies were retrospective. Second, the methods used to assess sarcopenia and myosteatosis, as well as the cut-offs used to define them, were heterogeneous.[13] Thirdly, most studies focus on patients with severe liver disease on LT waiting lists, while data on patients with less severe disease are still scarce.[22] Finally, the extent to which different muscle abnormalities coexist and impact patient prognosis and outcomes has not been previously defined.

To address this knowledge gap, we planned a multicentre prospective Italian study under the aegis of the Italian Association for the Study of the Liver.

Patients and methods

Study aims and design

The primary aim of the study was to investigate the impact of sarcopenia and myosteatosis, alone or in combination, on mortality, the need for hospitalization, and first or further liver decompensation. The secondary aim was to explore the correlations among muscle function, frailty and sarcopenia and myosteatosis in the subgroup of patients with available data.

Consecutive patients with cirrhosis from 26 Italian centres (see Fig. S1) were prospectively enrolled between January 2019 and December 2021 and followed-up for 1 year. The onset of the SARS-CoV-2 pandemic during the enrolment period significantly limited recruitment resources, especially from smaller centres. The cumulative incidence of the outcome of interest was calculated by competing risk analysis, and the prognostic role of muscle changes was assessed by multivariable analysis adjusted for major known prognostic indicators.

Eligibility and inclusion criteria

All patients with cirrhosis aged 40-75 years who underwent an abdominal computed tomography (CT) scan of the third lumbar (L3) vertebra for any clinical indication were eligible for the study. The cut-off age for inclusion was an arbitrary decision to reduce the potential confounding effect of age-related muscle abnormalities. We excluded patients on LT waiting lists or who had hepatocellular carcinoma (HCC), a history of LT, concomitant neuromuscular disease, current malignancy other than non-melanocytic skin cancer, a history of serious extrahepatic diseases, or HIV infection. Each patient was enrolled at the time of abdominal CT scan.

Patient characteristics and follow-up

Patient characterisation at inclusion was based on the following data: age, sex, liver disease aetiology, the presence and grade of ascites according to the International Club of Ascites, presence and grade of overt hepatic encephalopathy (OHE) according to the West Haven criteria, animal naming test (ANT) result of patients without OHE, oesophageal varices and size (according to the American Association for the Study of Liver Disease classification), history of gastrointestinal bleeding, and model for end-stage liver disease (MELD), MELD-Na and Child–Pugh scores.

Anthropometric data (including dry weight for patients with ascites and fluid retention) and hand grip test (HGT), liver frailty index (LFI) and 'timed up and go' (TUG) test results were also recorded.

Clinical visits to the outpatient clinic and biochemical exams were repeated at 6 and 12 months or more frequently when needed. During follow-up, data on hospitalizations and episodes of liver decompensation were collected during inpatient and outpatient visits, as well as through phone calls or emails.

The first episode of liver decompensation was defined as the presence of ascites, OHE, or variceal bleeding in a previously compensated patient. 'Further decompensation' was defined as the worsening of previous decompensation with recurrent, refractory, or complicated ascites; acute kidney injury; spontaneous bacterial peritonitis; variceal bleeding; or worsening of OHE. Death from any cause and LT were also recorded during the observation period.

Assessment of muscle changes

The assessment of sarcopenia and myosteatosis on CT images was centralised at the coordinating centre. All CT scans were assessed for sarcopenia and myosteatosis by two trained experts (S.DC. and L.L.).

According to the consensus statement of the EWGSOP (European Working Group on Sarcopenia in Older People),[23] abdominal muscle area was always evaluated by CT at the third or fourth lumbar vertebra in the present study. The Hounsfield unit (HU) limits used for assessing skeletal muscle mass ranged from -29 to +150 HU. The muscle area was normalised for height, resulting in a ratio (cm^2/m^2) known as the L3–L4 skeletal mass index (L3–L4 SMI). Patients were classified as having sarcopenia according to the validated SMI cut-off (<50 cm^2/m^2 for men and < 39 cm^2/m^2 for women).[24]

To assess myosteatosis across the entire muscle area, we computed the mean muscle attenuation in HU, which reflects the fat infiltration of muscles. We used the same CT image used for the SMI calculations. Patients were classified as having myosteatosis according to the following cut-off values: <41 HUs for patients with a BMI <24.9 kg/m^2 and <33 HU for those with a BMI ≥25 kg/m^2.[25]

The adipose tissue located within the peritoneal cavity was identified using HU thresholds for visceral adipose tissue ranging from -150 to -50 HU.[26] Subcutaneous adipose tissue was detected in the layer of adipose tissue beneath the skin and above the parietal peritoneal lining, using HU thresholds ranging from -190 to -30 HU.[27]

Assessment of muscle function and frailty

Assessment of patient muscle function at enrolment was based on the TUG test[28] and the HGT.[29] Patient frailty was assessed using the LFI.[30] Patients with LFI ≥4.5 were categorised as *frail*, those with LFI ≥3.2 and <4.5 were categorised as *prefrail*, and those with LFI <3.2 as *non-frail.*

Study approval and informed consent

The Local Ethical Committee of the Coordinator Center approved the study protocol and data collection (EC n° 94/19, 30/01/19), and each collaborating centre provided its own ethical committee approval. All patients provided informed consent to participate in the study.

Statistical analysis

Baseline patient characteristics are reported as proportions or means and standard deviations. The chi-square test assessed differences between proportions, and the Student's *t* test evaluated differences between means. We calculated the incidence rates of the events of interest as the number of observed events/100 patient-years of follow-up.

We assessed the cumulative incidence function (CIF) of death by competing risk analysis,[31] with LT as a competing event. Death and LT were considered competing events when assessing the CIF of first and further liver decompensation and of new hospitalizations. CIF differences were assessed by Gray's test.[32] Time zero for analyses of time to events (death, first or further decompensation and new hospitalization) was the date of the index CT for muscle assessment.

One-year incidence plots are shown. Throughout the text and figures, probabilities are expressed as percentages.

We assessed the adjusted impact of muscle damage on outcomes by a multivariable Fine-Gray model for competing risks.[33] We used Robust (sandwich) variance estimation for multivariable models.[34,35] The analysis included the following known prognostic indicators: sarcopenia, myosteatosis, serum bilirubin, INR, serum albumin, serum creatinine, age, sex, and the presence of OHE or ascites. The MELD[36] and Child-Pugh scores[37] were included in separate analyses excluding their individual components to avoid redundancy. A variable indicating participating centre was included in all multivariable models to account for between-centre heterogeneity. We performed variable reduction for the final multivariable models by a backwards procedure, based on the importance of risk predictors, clinical judgement and statistical significance. We included the number of variables in the multivariable models ≤1 per 10 observed outcome events.

In a subgroup of patients with available information on muscle function, we explored the relationships between HGT, TUG test and LFI results with muscle surface and muscle attenuation at the time of CT scan using regression analyses.

Statistical analyses were performed using STATA 16.1 (©2019 Stata Corporation, College Station, TX).

Results

Patient characteristics at inclusion and type of skeletal muscle damage

A total of 447 patients were eligible for the study, 14 of whom were excluded due to incomplete data, leaving 433 patients with available data for analysis. The mean follow-up (±SD) was 349±89 days.

The characteristics of the patients in the total population and in the subgroups according to the type of muscle changes are shown in Table 1.

The most common aetiology of cirrhosis was alcohol-related (39.9%), followed by hepatitis C (15.5%) and metabolic (15.0%). At enrolment, 158 patients were compensated, and 275 patients were decompensated, owing to ascites, OHE or portal hypertensive bleeding alone or in various combinations. The mean MELD score was 13±4.95 points, and the mean Child–Pugh score was 7.45±2.0 points; 162 patients (37.4%) were in class A, 196 (45.3%) in class B, and 75 (17.3%) in class C.

Isolated sarcopenia (I-sarcopenia) was diagnosed in 36 patients (8.3%), isolated myosteatosis (I-myosteatosis) in 166 patients (38.3%) and combined sarcopenia and myosteatosis in 135 patients (31.2%), while only 96 patients (22.2%) had no evidence of muscle damage (Fig. 1). Muscle changes were significantly more common in females than in males (88.8% *vs.* 73.4%, p <0.0001), mainly due to a significantly greater prevalence of I-myosteatosis in females (52.0% *vs.* 32.8%, p <0.0001) (Fig. S2). Overall, patients with any muscle changes had more advanced disease, with significantly greater Child–Pugh scores (7.6±2.0 *vs.* 7.0±1.8 points, p = 0.01) and a greater prevalence of ascites (53.4% *vs.* 36.5%, p = 0.003).

Myosteatosis was the most frequent muscle change, detected in 301 patients (69.5%). Compared to I-sarcopenia patients and patients without muscle changes, patients with myosteatosis were significantly older (60.8±8.8 *vs.* 57.2±9.1 years, p = 0.0001) and had significantly higher MELD (13±5 *vs.* 12±4 points, p = 0.006), Child–Pugh (7.6±2.1 *vs.* 7.0±1.8 points, p = 0.006) and ANT (18.5±5.5 *vs.* 16.5± points, p = 0.0037) scores. Comparisons between patients with and without myosteatosis, independent of the presence of sarcopenia, are shown in Table S1.

Patients with I-myosteatosis also had greater visceral and subcutaneous adiposity than patients with myosteatosis and sarcopenia (170±100 cm/m^2 *vs.* 131±79 cm/m^2, p = 0.0005) (Table 2).

Muscle function assessment was not always available, as some centres did not have a suitable handgrip dynamometer, which preventing LFI assessment in many patients. Specifically, we collected HGT results for 274 patients (63.3%), TUG test results for 228 patients (52.6%), and LFI results for 249 patients (57.5%). Compared to patients without myosteatosis, those with myosteatosis, either alone or associated with sarcopenia, had worse TUG test (13.3±5.7 *vs.* 9.4±3.8 points, p value <0.0001) and HGT (32.5±16.4 *vs.* 36.8±15.9 points, p = 0.049) results and were more frequently diagnosed as frail (33% *vs.* 13%, p = 0.002).

Impact of muscle changes on mortality

During the follow-up period, 51 deaths occurred, 45 of which were liver related. The major outcome events and corresponding incidence rates/100 patient-years are shown in Table 1. The incidence of death was higher in patients with muscle changes than in those without muscle changes. The difference was statistically significant for patients with I-myosteatosis (p = 0.015) and those with combined sarcopenia and myosteatosis (p = 0.012). The corresponding 1-year cumulative incidences of death with LT as a competing risk were 5.6% for patients without muscle changes, 5.2% for patients with I-sarcopenia, 13.8% for patients with I-myosteatosis and 13.4% for patients with combined sarcopenia and myosteatosis (Fig. 2). The differences between patients in each muscle change group and patients without muscle changes were,

Table 1. Patient characteristics at inclusion and major clinical outcomes.

	Whole cohort*	No muscle changes	I-sarco*	I-myo*	Combined sarco-myo*
Patients, n (%)	433	96 (22.2)	36 (8.3)	166 (38.3)	135 (31.1)
Age, years	57.1 (8.9)	56.7 (8.9)	58.6 (9.2)	60.8 (8.7) [0.0003]	60.7 (9.0) [0.0009]
Sex, M	308 (71.1)	82 (85.4)	31 (86.1)	101 (60.8) [<0.0001]	94 (69.6) [0.005]
Aetiology					
Alcohol	173 (39.9)	37 (38.5)	10 (27.7)	67 (40.4)	59 (43.7)
HCV	67 (15.5)	18 (18.8)	10 (27.7)	22 (13.2)	17 (12.6)
HBV	19 (4.4)	7 (7.3)	0	6 (3.6)	6 (4.4)
Alcohol+virus	38 (8.8)	12 (12.5)	1 (2.8)	12 (7.2)	13 (9.6)
NASH	65 (15.0)	12 (12.5)	8 (22.2)	31 (18.7)	14 (10.4)
Autoimmune/biliary disease	9 (2.1)	0	2 (5.6)	4 (2.4)	3 (2.2)
Others or undefined	62 (14.3)	10 (10.4)	5 (13.9)	24 (14.5)	23 (17.0)
Metabolic					
BMI, kg/m^2	27.8.7 (5.5)	28.7 (4.5)	24.4 (4.2) [<0.001]	28.6 (6.0)	24.6 (4.6) [<0.001]
Diabetes	138 (31.9)	33 (34.4)	8 (22.2)	61 (36.7)	36 (26.7)
Hypertension	160 (36.9)	30 (31.3)	10 (27.8)	72 (43.4) [0.05]	48 (35.6)
Dyslipidemia	75 (17.3)	17 (17.7)	5 (13.9)	36 (21.7)	17 (12.6)
Laboratory/clinical					
INR	1.4 (0.38)	1.34 (0.23)	1.34 (0.27)	1.43 (0.41) [0.036]	1.42 (0.41)
Albumin, g/L	35.8 (0.72)	37.4 (7.91)	37.7 (7.51	35.7 (7.0)	34.3 [0.0015]
Bilirubin, mg/dl	2.9 (4.5)	2.3 (2.4)	2.6 (4.0)	2.5 (3.0)	3.9 (6.7) [0.02]
Creatinine, mg/dl	0.90 (0.53)	0.88 (0.36)	0.83 (0.18)	0.87 (0.32)	0.97
Hb, g/dl	11.9 (2.4)	12.5 (2.3)	12.0 (2.4)	11.9 (2.3) [0.05]	11.3 [0.0001]
Oesophagogastric varices	277 (64.4)	68 (71.6)	23 (63.9)	105 (64.0)	81 (60)
Ascites	215 (49.7)	35 (36.5)	19 (52.8)	72 (43.4)	89 (65.9) [<0.0001]
OHE	86 (19.9)	17 (17.7)	10 (27.8)	36 (21.7)	23 (17.0)
ANT [n = 348]	17.1 (6.18)	18.4 (5.3)	18.8 (6.1)	16.2 (6.2) [0.01]	16.8 (6.5) [0.03]
Child–Pugh score	7.5 (2.0)	7.0 (1.78)	7.2 (1.9)	7.4 (2.1)	7.9 (2.0) [0.0009]
Child-Pugh A	162 (37.4)	44 (45.8)	12 (33.3)	68 (41.0)	38 (28) [0.018]
Child-Pugh B	196 (45.3)	40 (41.7)	18 (50)	68 (41.0)	70 (51.9) [0.018]
Child-Pugh C	75 (17.3)	12 (12.5)	6 (16.7)	30 (18.0)	27 (20) [0.018]
MELD score	12.9 (4.9)	12.2 (3.9)	11.6 (5.2)	12.9 (4.8)	13.9 (5.5) [0.007]
MELD-Na score	14.3 (5.4)	13.2 (4.3)	12.5 (6.3)	14.3 (5.2)	15.6 (5.9) [<0.001]
Ongoing therapies					
Primary prophylaxis†	221 (51)	52 (54.2)	22 (61.1)	79 (47.6)	68 (50.4)
NSBB	109 (25.2)	24 (25)	12 (33.3)	41 (24.7)	32 (23.7)
EVL	41 (9.5)	11 (11.5)	2 (5.6)	15 (9.0)	13 (9.6)
Secondary prophylaxis‡	71 (16.4)	17 (17.7)	8 (22.2)	23 (13.9)	23 (17.0)
Rifaximin	99 (24.5)	26 (27.7)	10 (31.2)	40 (26.0)	23 (18.6)
Lactulose	181 (44.8)	44 (46.8)	16 (50)	73 (47.4)	48 (38.7)
Albumin	59 (14.6)	15 (15.9)	4 (12.5)	24 (15.6)	16 (12.9)
Outcome events, N (incidence rate per 100 patient-years)					
Follow-up, patient-years	414	95	36	158	125
Death	51 (12.3)	5 (5.2)	2 (5.6)	24 (15.2) [0.015]	20 (16.0) [0.012]
Liver transplant	42 (9.7)	10 (10.5)	8 (22)	10 (6.3)	14 (11.2)
New hospitalization	207 (50.0)	33 (34.7)	17 (47.2)	88 (55.7) [0.0012]	69 (55.2) [0.0025]
All decompensation#	143 (31.4)	24 (25.3)	11 (30.6)	45 (28.5)	50 (40.0) [0.023]

Data are presented as number of patients or means and % or SD in brackets, as appropriate; between group differences were assessed by the Student's *t* test for means and Chi-square test for %, respectively.

ANT, animal naming test; EVL, endoscopic variceal ligation; INR, international normalised ratio; MELD, model for end-stage liver disease; NASH, non-alcoholic steatohepatitis; NSBB, non-selective beta blocker; OHE, overt hepatic encephalopathy.

*p value for significant differences from patients without muscle changes.

†Primary prophylaxis for variceal bleeding.

‡Secondary prophylaxis for variceal bleeding, mostly NSBB+EVL.

#All decompensation includes first or further decompensation.

however, not significant, although the increase in mortality in patients with combined sarcopenia and myosteatosis approached significance (p = 0.079).

Impact of muscle changes on hospitalization

During the 1-year follow-up, 207 liver-related hospitalizations were reported. The hospitalization incidence rate/100 patient-years in patients with combined sarcopenia and myosteatosis and I-myosteatosis was 55.7 and 55.2, respectively, both significantly higher (p = 0.0012 and 0.025, respectively) than that in patients without muscle changes (34.7). In patients with I-sarcopenia, the corresponding incidence rate was 47.2, which was not significantly different from that in patients without muscle changes (Table 1). The 1-year cumulative incidence of new hospitalization with death and LT as competing events

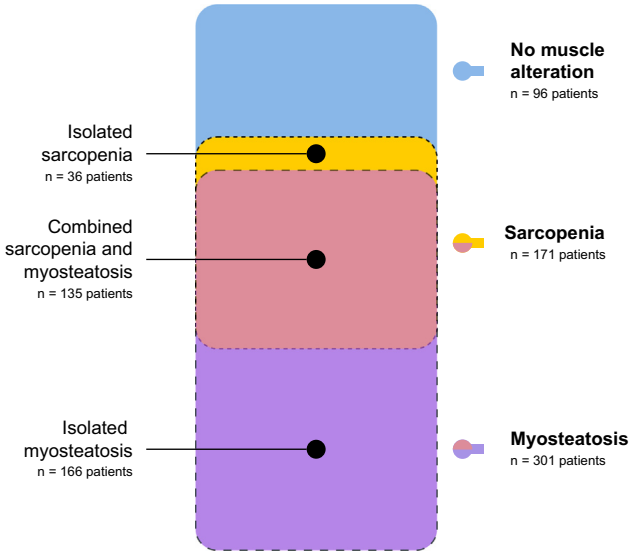


Fig. 1. Proportions and interactions of muscle abnormalities in our cohort. No muscle alterations (light blue), isolated sarcopenia (yellow), isolated myosteatosis (purple) and combined sarcopenia and myosteatosis (pink). (This figure appears in color on the web.)

was also significantly greater in patients with any type of muscle change than in those without muscle changes (Fig. 3).

Impact of muscle changes on first and further liver decompensation

At the start of the study, 158 patients with compensated cirrhosis were included, 43 without muscle changes, 12 with I-sarcopenia, 66 with I-myosteatosis and 37 with combined sarcopenia and myosteatosis.

Overall, decompensation occurred for the first time during follow-up in 5 of the 43 patients without muscle changes and 19 of the 115 patients with any type of muscle changes, with corresponding incidence rates per 100 patient-years of 11.3 and 16.7, respectively ($p = 0.39$). Among the 275 patients with decompensated cirrhosis at inclusion, 119 developed further decompensation: 19 of 53 without muscle changes, 11 of 24 with sarcopenia, 45 of 100 with myosteatosis, and 44 of 98 with combined sarcopenia and myosteatosis. The corresponding incidence rates per 100 patient-years were 37.6, 46.4, 50, and 48.9, respectively (p was not significant for differences between each type of change *vs.* no changes). In the overall population, the 1-year cumulative incidence of any decompensating event (either first or further, with death and liver transplant as competing events) was 39% for patients with combined sarcopenia and myosteatosis, 36% for those with I-sarcopenia, 34% for those with I-myosteatosis, and 30% for those with no muscle changes (Fig. 4). Compared with patients with no changes, the difference was significant for those with combined sarcopenia and myosteatosis ($p = 0.018$) and for those with I-sarcopenia ($p = 0.046$).

Adjusted prognostic role of muscle changes

Univariate analysis for death, hospitalization, and first or further decompensation is shown in Table S2. To investigate whether muscle changes had an independent impact on outcomes, we performed multivariable analyses. Multivariable models exploring the prognostic impact of muscle changes including MELD are shown in Table 3, while those including MELD-Na and Child-Pugh scores are shown in Table S3 and Table S4, respectively. The corresponding analyses excluding the MELD score and including its individual components are shown in Table S5.

Muscle changes had a significant effect on the incidence of hospitalization ($p = 0.012$) and tended to increase mortality ($p = 0.058$) but had no effect on first or further liver decompensation ($p = 0.60$). The c-statistic of the attenuation index for mortality was 0.69 (CI 0.62-0.76). In models including the individual components of the MELD score (ascites, OHE, and albumin) (Table S2), no independent prognostic effect of muscle changes was observed.

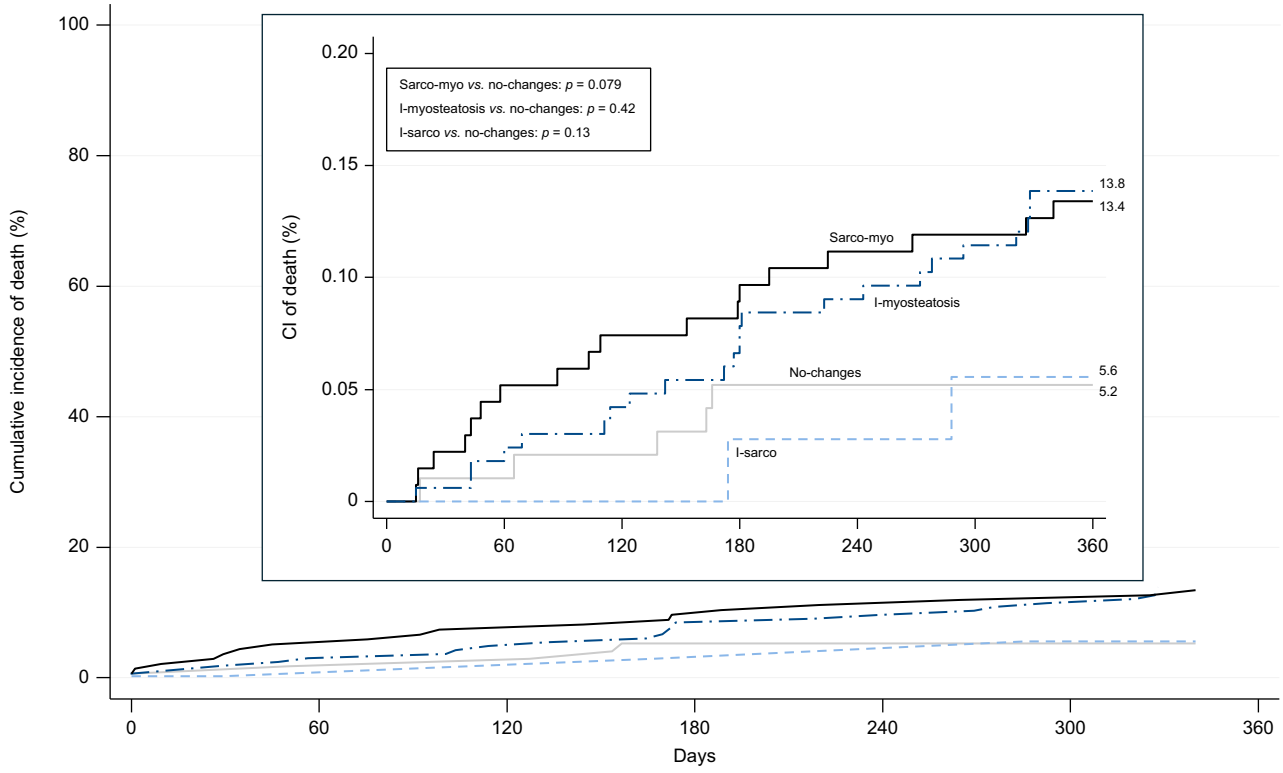
Correlations between muscle function and sarcopenia and myosteatosis

The frailty index was assessed in 238 patients, and the mean frailty index score was 3.7±0.85 points. A total of 65 patients were classified as frail, and 117 as prefrail. The mean frailty index score was significantly higher in patients with combined sarcopenia and myosteatosis (3.96±0.84) or I-myosteatosis (3.83±0.88) than in patients with I-sarcopenia (3.5±0.6) or no muscle changes (3.49±0.82). Differences in the same direction were found for HGT and TUG test results (Table 2). The HGT values correlated with the SMI ($r = 0.11$, $p < 0.0001$) but not with the HU (muscle attenuation utilised for the diagnosis of myosteatosis as a continuous value), while the TUG test and LFI did not correlate with the SMI but were inversely correlated with the HU (Fig. S3).

Table 2. Function tests and CT parameters of the whole population and of patient subgroups according to the type of muscle changes.

Parameter	Whole cohort	No muscle changes	I-sarco*	I-myo*	Combined sarco-myo*
Function tests, mean (SD)					
Frailty liver index (n = 238)	3.77 (0.85)	3.49 (0.82)	3.50 (0.60)	3.83 (0.88) [0.021]	3.96 (0.84) [0.0022]
Hand grip, kg (n = 263)	33.87 (16.3)	38.26 (17.33)	31.99 (8.88)	34.78 (17.03)	29.71 (15.17) [0.0021]
Up & go test, sec (n = 226)	11.8 (5.4)	9.9 (3.9)	8.0 (2.9)	13.3 (5.7) [0.0002]	12.6 (5.5) [0.003]
Computed tomography parameters, mean (SD)					
Patients, n	433	96	36	166	135
L3 SMI	50.3 (10.9)	59.1 (8.8)	43.4 (5.3) [<0.0001]	54.2 (9.4) [<0.0001]	41.2 (6.6) [<0.0001]
Muscle attenuation, %HU	31.6 (8.4)	39.8 (5.1)	40.1 (6.0)	27.8 (6.8) [<0.0001]	28.2 (6.7) [<0.0001]
VAT, cm/m^2	150.8 (91.8)	158.5 (87.7)	111.6 (80.7) [0.006]	170.2 (100.1)	131.9 (79.8) [0.017]
SAT, cm/m^2	196.8 (120.6)	213.5 (106.7)	137.5 (89.5) [0.0002]	236.1 (132.9)	152.8 (99.9) [<0.0001]

HU, Hounsfield unit; SAT, subcutaneous adipose tissue; SMI, skeletal muscle index; VAT, visceral adipose tissue.
*p value for significant differences compared with patients without muscle changes, computed by the Student's t test.



N° at risk							
No changes	96	94	92	87	86	85	84
I-sarco	36	34	33	30	29	28	26
Sarco-myo	135	123	117	112	107	105	103
I-myosteatosis	166	163	158	152	146	140	133

Fig. 2. Cumulative incidence of death with liver transplantation as a competing risk in four patient subgroups according to the type of muscle damage. The inset shows the same data on a larger scale. The numbers below the abscissa are the number of patients at risk. The numbers next to the curves are the cumulative incidence of death % at the end of the observation period. *p* values were computed by the Gray's test. I-myosteatosis, isolated myosteatosis; I-sarco, isolated sarcopenia; No changes, no muscle changes; Sarco-myo, sarcopenia combined with myosteatosis.

Discussion

In the present study, we aimed to assess the prognostic relevance of muscle alterations in a large prospective cohort of patients with cirrhosis and varying degrees of liver impairment.

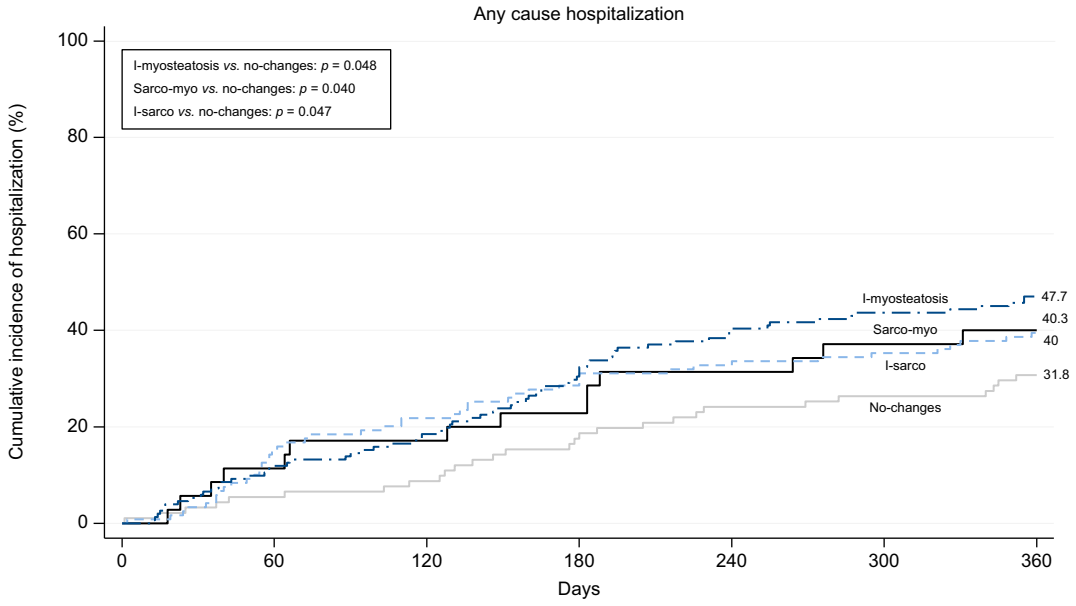
Seventy-eight percent of patients had some muscle changes at the time of enrolment. In our cohort, myosteatosis was the main muscle alteration affecting a significant proportion of the study population, while sarcopenia was rarely present in the absence of myosteatosis. In a retrospective cohort, Tachi *et al.*[38] reported a prevalence of myosteatosis of 82% and sarcopenia of 36%, with 93% of patients with sarcopenia having concomitant myosteatosis. Only a small proportion of patients had sarcopenia alone, as in our study. A recent study[39] examined the combination of reduced muscle function, quality, and quantity in 197 patients and suggested that myosteatosis may precede the onset of other muscle abnormalities. There may be a physiological explanation for this observation. Chronic hyperammonaemia, present in cirrhosis, induces mitochondrial dysfunction and a subsequent reduction in lipid oxidation, leading to intramuscular fat infiltration (myosteatosis).[40] Intramuscular fat, often associated with insulin resistance, has been linked to the development of a lipotoxic profile associated with the secretion of fatty acids and inflammatory adipokines, the latter having a detrimental effect on myocyte function.[41]

In our cohort, patients with myosteatosis, with or without sarcopenia, were generally older, more often female, had more visceral and subcutaneous fat, and had more advanced liver disease (see Table 1 and Table S1). In addition, these patients exhibited lower cognitive performance (as measured by the ANT) and were more likely to have ascites and bacterial infections. Functional performance was also impaired in these patients, as evidenced by lower TUG test scores and a greater tendency to frailty.

The role of myosteatosis, which has not always been evaluated in previous studies, was relevant in blunting that of sarcopenia, which has always been considered an important predictor of clinical outcomes in patients with cirrhosis.

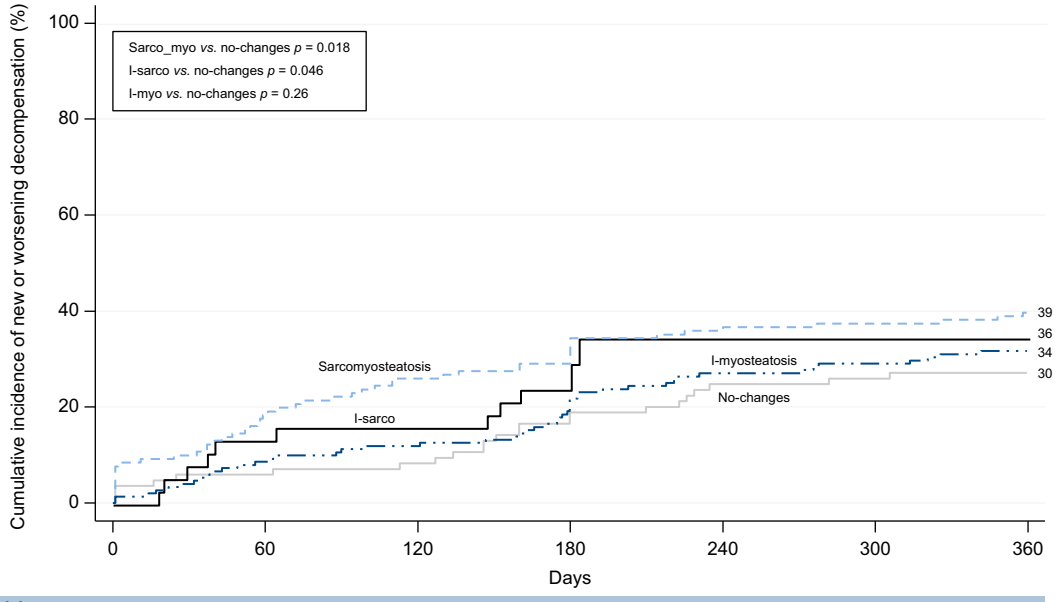
Indeed, a diagnosis of myosteatosis, even if not associated with sarcopenia, was a predictor of a greater risk of death (Fig. 2) and hospitalization (Fig. 3).

On the other hand, I-sarcopenia was not an independent predictor of mortality or hospitalization in our cohort. This finding may be influenced by the limited number of patients diagnosed with I-sarcopenia.



N° at risk							
No changes	96	87	83	73	67	65	61
I-sarco	36	29	26	22	18	16	15
Sarco-myo	135	95	85	74	67	64	59
I-myosteatosis	166	140	129	108	89	82	75

Fig. 3. Cumulative incidence of hospitalization, with death and liver transplantation as competing risks, in four patient subgroups according to type of muscle damage. The numbers below the abscissa are the number of patients at risk. The numbers next to the curves are the cumulative incidence of death % at the end of the observation period. *p* values were computed by the Gray test. I-myosteatosis, isolated myosteatosis; I-sarco, isolated sarcopenia; No changes, no muscle changes; Sarco-myo, sarcopenia combined with myosteatosis.



N° at risk							
No changes	96	90	87	76	69	68	67
I-sarco	36	29	27	23	18	17	17
Sarco-myo	135	103	91	84	74	72	68
I-myosteatosis	166	153	146	130	114	108	100

Fig. 4. Cumulative incidence of new liver decompensation (first or further) with death and liver transplantation as competing risks in four patient subgroups according to type of muscle damage. The numbers below the abscissa are the number of patients at risk. The numbers next to the curves are the cumulative incidence of death % at the end of the observation period. *p* values were computed by the Gray test. I-myosteatosis, isolated myosteatosis; I-sarco, isolated sarcopenia; No changes, no muscle changes; Sarco-myo, sarcopenia combined with myosteatosis.

Table 3. Adjusted prognostic role of muscle changes, by the Fine and Gray model, for death, hospitalization and liver decompensation, including the MELD score.

Variable	Score	Sub-hazard ratio	*p* value	95% CI	
Including the MELD score and excluding relevant single components					
Death (LT as a competing event)					
Muscle changes[†]	No = 0; I-sarco = 1, I-myo = 2; sarco-myo = 3	1.36	0.058	0.99	1.86
MELD	Continuous values	1.12	<0.0001	1.06	1.86
OHE	No = 0; yes = 1	2.09	0.011	1.18	3.69
Ascites	No = 0; yes = 1	2.73	0.005	1.35	5.52
Hospitalization (death and LT as competing events)					
Muscle changes[†]	No = 0; I-sarco = 1, I-myo = 2; sarco-myo	1.18	0.012	1.04	1.35
MELD[*]	Continuous values	1.36	0.191[*]	0.99	1.06
Albumin	g/L	1.28	0.052	0.998	1.65
Ascites	No = 0; yes = 1	1.86	0.062	0.98	1.91
Non-elective hospitalization (death and LT competing)					
Muscle changes[†]	No = 0; I-sarco = 1, I-myo = 2; sarco-myo = 3	1.12	0.25	0.92	1.36
MELD[*]	Continuous values	1.02	0.31[*]	0.98	1.07
HE	No = 0; yes = 1	2.26	0.001	1.38	3.72
Ascites	No = 0; yes = 1	2.86	<0.0001	1.67	4.87
First or further decompensation (death and LT as competing events)					
Muscle changes[†]	No = 0; I-sarco = 1, I-myo = 2; sarco-mio = 3	1.04	0.60	0.90	1.20
MELD	Continuous values	1.04	0.02	1.01	1.07
Ascites	No = 0; yes = 1	2.49	<0.0001	1.72	3.62
OHE	No = 0; yes = 1	1.48	0.038	1.02	2.15

LT, liver transplantation; MELD, model for end-stage liver disease; OHE, overt hepatic encephalopathy.
*MELD was forced in this analysis to show the effect of the other variables adjusted for MELD.
[†]In these analyses, each of the assessed muscle changes (*i.e.* I-sarcopenia, I-myosteatosis, and sarco-myosteatosis) was scored as absent/present; if none of them was significant, we included in the model a discrete variable scored as follows: no changes = 0, I-sarcopenia = 1, I-myosteatosis = 2, and combined sarcopenia and myosteatosis = 3.

Our study highlights that the occurrence of muscle changes, regardless of the type, represents a significant moment in the natural history of cirrhosis and adversely affects numerous outcomes. Multivariate analyses of competitive risk factors for mortality (with LT as a competing event), including the presence and type of muscle abnormalities, the MELD score and the presence of OHE or ascites (Table 3), showed that the presence of muscle changes negatively affected survival and hospitalization rates.

Altered muscle function and frailty have been shown to affect quality of life,[42] self-autonomy and prognosis of patients with cirrhosis.[43] In our study, many patients were categorised as frail or prefrail (76.5% in total). The correlation between frailty and TUG test results and muscle attenuation suggests that these tests, which are easy to perform, may be useful surrogates that overcome the need for imaging to assess changes in muscle quality.

The strengths of the present study include its prospective design, multicentre structure, use of CT scans (the gold standard for detecting muscle changes) in all patients, and detailed analysis of various types of muscle abnormalities.

Our study has several limitations. Nutritional assessment was only performed at baseline, without considering changes that may have occurred during follow-up. The different centres' policies regarding patient care in day services or in hospitals may have influenced the hospitalization rate, and laboratory data were not centralised. Muscle function tests were not performed in any of the centres participating in the study. Furthermore, although CT scans are widely used in patients with cirrhosis for various reasons (see Table S6), the availability of CT scans as an inclusion criterion may have introduced a baseline selection bias. Moreover, the prevalence of muscle alterations was sex dependent. Indeed, male patients represented 70% of the population, which could have impacted the overall cohort's distribution averages. Consequently, the study outcomes may not fully represent both sexes, highlighting the need for future larger studies designed with this in mind.

In conclusion, our study has shown that a comprehensive and integrated assessment of muscle changes is crucial for understanding their role in the natural history of cirrhosis. Our analyses revealed that myosteatosis is the most frequent alteration and has a significant impact on the course of liver disease. Many previous studies have focused on the assessment of sarcopenia, but concomitant myosteatosis is likely to play a major prognostic role. This may resize the predictive role of sarcopenia in favour of a more comprehensive consideration of muscle changes.

Our study suggests that muscle function tests could serve as a valuable and practical bedside tool for estimating prognosis and identifying patients at greater risk of mortality.

Affiliations
[1]*Department of Translational and Precision Medicine, Sapienza University of Rome, Rome, Italy;* [2]*Gastroenterology Unit, Ospedale V. Cervello, Palermo, Italy;* [3]*Unit of Semeiotics, Liver and Alcohol-related Diseases, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Italy;* [4]*Department of Medical and Surgical Sciences, Alma Mater Studiorum University of Bologna, Bologna, Italy;* [5]*Division of Gastroenterology, Azienda Ospedaliero-Universitaria of Modena and University of Modena and Reggio*

Emilia, Modena, Italy; [6]*Department of Gastroenterology and GI Endoscopy, Arcispedale S. Anna Ferrara, Italy;* [7]*Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Division of Gastroenterology and Hepatology, Milan, Italy;* [8]*CRC "A. M. and A. Migliavacca" Center for Liver Disease, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy;* [9]*Hepatology and Liver Transplantation Unit, Azienda Sanitaria Universitaria Friuli Centrale, University of Udine, Udine, Italy;* [10]*SSD Insufficienza Epatica e Trapianto, AOU Città della Salute e della Scienza di Torino, Italy;* [11]*Division of Medicine and Hepatology, University Hospital of Messina, Messina, Italy;* [12]*Gastroenterology, Verona University Hospital, Ospedale Borgo Trento, Verona, Italy;* [13]*Gastroenterology, Department of Medical Specialties, University of Modena and Reggio Emilia and Azienda Ospedaliero-Universitaria di Modena, Modena, Italy;* [14]*Liver Injury and Transplant Unit, Ospedali Riuniti Ancona, Italy;* [15]*Division of Gastroenterology and Hepatology, Città della Salute e della Scienza Hospital, University of Turin, Turin, Italy;* [16]*Department of Biomedical Sciences, Humanitas University, Pieve Emanuele (MI), Italy;* [17]*Division of Internal Medicine and Hepatology, Department of Gastroenterology, IRCCS Humanitas Research Hospital, Rozzano (MI), Italy;* [18]*Department of Medicine, Surgery and Health Sciences, University of Trieste, Trieste, Italy;* [19]*Department of Advanced Medical and Surgical Sciences, University of Campania "Luigi Vanvitelli", Naples, Italy;* [20]*Section of Gastroenterology, Department of Emergency and Organ Transplantation, University of Bari, Bari, Italy;* [21]*Internal Medicine Unit, Department of Clinical and Experimental Medicine, University of Messina, Messina, Italy*

Abbreviations

ANT, animal naming test; CIF, cumulative incidence function; CT, computed tomography; HGT, hand grip test; HU, Hounsfield unit; I-myosteatosis; isolated myosteatosis; I-sarcopenia; isolated sarcopenia; LFI, liver frailty index; LT, liver transplantation; MELD, model for end-stage liver disease; OHE, overt hepatic encephalopathy; SMI, skeletal muscle index; TUG, timed up and go.

Financial support

The authors did not receive any financial support to produce this manuscript.

Conflict of interest

The authors declare that they have no competing interests.

Please refer to the accompanying ICMJE disclosure forms for further details.

Authors' contributions

Manuela Merli: study concept and protocol, data collation, study supervision, analysis, results interpretation, and drafting and revision of the manuscript for important intellectual content.

Simone di Cola: study protocol, data coordinator, analysis plan, results interpretation, and drafting and revision of the manuscript. Gennaro D'Amico: analysis, results interpretation, revision of the manuscript for important intellectual content. Paolo Caraceni and Filippo Schepis: centre supervision, results interpretation, manuscript revision for important intellectual content. Simone Loredana, Pietro Lampertico, Massimo Iavarone, Pierluigi Toniutto, Silvia Martini, Sergio Maimone, Antonio Colecchia, Gianluca Svegliati-Barone, Alessio Aghemo, Saveria Lory Crocè;, Luigi Elio Adinolfi, Maria Rendina, Enrico Pompili, Federica Indulti, Dario Saltini, Giulia Tosetti, Paola Serri, Mariangela Bruccoleri, Carolina Martelletti, Veronica Nassisi, Alberto Ferrarese, Carlo Alessandria, Ilaria Giovio, Chiara Masetti, Nicola Pugliese, Michele Campigotto, Riccardo Nevol: centre supervision, results interpretation, manuscript revision. Gaetano Bertino, Clara Balsano, Nerio Lapadre, Marcello Maida, David Sacerdoti, Leonardo Antonio Natola, Carolina Ciacci, Antonella Santonicola, Raffaele Cozzolongo, Lorenzo Antonio Surace, Anna Ludovica Fracanzani, Annalisa Cespiati, Alessandro Federico, Mario Romeo, Antonio Grieco, Giuseppe Marrone, and Luca Vizioli: Patient selection and inclusion, follow-up, and data collection.

Data availability statement

The data that support the findings of this study are available from the corresponding author, M.M., upon reasonable request.

Acknowledgements

[‡]*EpatoSarco Group:* Giulia Iannone[1], Paola Serri[2], Gaetano Bertino[3], Clara Balsano[4], Nerio Iapadre[5], Marcello Maida[6], David Sacerdoti[7], Leonardo Antonio Natola[7], Carolina Ciacci[8], Antonella Santonicola[8], Raffaele Cozzolongo[9], Lorenzo Antonio Surace[10], Anna Ludovica Fracanzani[11], Annalisa Cespiati[11], Alessandro Federico[12], Mario Romeo[12], Antonio Grieco[13], Giuseppe Marrone[13], Luca Vizioli.[14]

[1]Department of Medical and Surgical Sciences, Alma Mater Studiorum University of Bologna, Bologna, Italy; [2]CRC "A. M. and A. Migliavacca" Center for Liver Disease, Department of Pathophysiology and Transplantation, University of Milan, Milan, Italy; [3]Hepatology Unit Department of Clinical and Experimental Medicine University of Catania, Catania - Italy Policlinico "G. Rodolico", Catania, Italy, [4]Department of Life, Health & Environmental Sciences, School of Emergency and Urgency Medicine, University of L'Aquila, 67100 L'Aquila, Italy, [5]Department of Infectious Disease, San Salvatore Hospital, L'Aquila, Italy, [6]Gastroenterology and Endoscopy Unit, S. Elia-Raimondi Hospital, Caltanissetta, Italy, [7]Liver Unit, Department of Medicine, University and Azienda Ospedaliera Universitaria Integrata of Verona, Verona, Italy, [8]Department of Medicine, Surgery and Dentistry, "Scuola Medica Salernitana", University of Salerno, Salerno, Italy, [9]Division of Gastroenterology, National Institute of Gastroenterology S De Bellis, Castellana Grotte, Bari, Italy, [10]Traveler and Migration Medicine Center, ASP Catanzaro, Catanzaro, Italy, [11]General Medicine and Metabolic Diseases, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy, [12]Department of Precision Medicine, University of Campania Luigi Vanvitelli, Naples, Italy, [13]Internal and Liver Transplant Medicine Unit, Fondazione Policlinico Universitario Agostino Gemelli IRCCS, Rome, Italy, [14]Internal Medicine Unit for the Treatment of Severe Organ Failure, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna, Italy.

Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jhep.2024.05.020.

References

[1] Bischoff SC, Bernal W, Dasarathy S, et al. ESPEN practical guideline: clinical nutrition in liver disease. Clin Nutr 2020;39(12):3533–3562. https://doi.org/10.1016/J.CLNU.2020.09.001.

[2] Merli M, Berzigotti A, Zelber-Sagi S, et al. EASL Clinical Practice Guidelines on nutrition in chronic liver disease. J Hepatol 2019;70(1):172–193. https://doi.org/10.1016/J.JHEP.2018.06.024.

[3] Merli M, Giusto M, Lucidi C, et al. Muscle depletion increases the risk of overt and minimal hepatic encephalopathy: results of a prospective study. Metab Brain Dis 2013;28(2):281–284. https://doi.org/10.1007/S11011-012-9365-Z.

[4] Kim G, Kang SH, Kim MY, Baik SK. Prognostic value of sarcopenia in patients with liver cirrhosis: a systematic review and meta-analysis. PLoS One 2017;12(10). https://doi.org/10.1371/JOURNAL.PONE.0186990.

[5] Tandon P, Raman M, Mourtzakis M, et al. A practical approach to nutritional screening and assessment in cirrhosis. Hepatology 2017;65(3):1044–1057. https://doi.org/10.1002/HEP.29003.

[6] Plauth M, Schütz ET. Cachexia in liver cirrhosis. Int J Cardiol 2002 Sep;85(1):83–87. https://doi.org/10.1016/s0167-5273(02)00236-x.

[7] Di Cola S, Nardelli S, Ridola L, et al. Ammonia and the muscle: an emerging point of view on hepatic encephalopathy. J Clin Med 2022;11(3). https://doi.org/10.3390/JCM11030611.

[8] Traub J, Reiss L, Aliwa B, et al. Malnutrition in patients with liver cirrhosis. Nutrients 2021;13(2):1–19. https://doi.org/10.3390/NU13020540.

[9] Bhanji RA, Moctezuma-Velazquez C, Duarte-Rojo A, et al. Myosteatosis and sarcopenia are associated with hepatic encephalopathy in patients with cirrhosis. Hepatol Int 2018;12(4):377–386. https://doi.org/10.1007/s12072-018-9875-9.

[10] Lucidi C, Lattanzi B, Di Gregorio V, et al. A low muscle mass increases mortality in compensated cirrhotic patients with sepsis. Liver Int 2018;38(5):851–857. https://doi.org/10.1111/liv.13691.

[11] Merli M, Giusto M, Lucidi C, et al. Muscle depletion increases the risk of overt and minimal hepatic encephalopathy: results of a prospective study. Metab Brain Dis 2013;28(2):281–284. https://doi.org/10.1007/S11011-012-9365-Z.

[12] Tandon P, Ney M, Irwin I, et al. Severe muscle depletion in patients on the liver transplant wait list: its prevalence and independent prognostic value. Liver Transpl 2012;18(10):1209–1216. https://doi.org/10.1002/LT.23495.

[13] Tantai X, Liu Y, Yeo YH, et al. Effect of sarcopenia on survival in patients with cirrhosis: a meta-analysis. J Hepatol 2022;76(3):588–599. https://doi.org/10.1016/J.JHEP.2021.11.006.

[14] Cruz-Jentoft AJ, Sayer AA. Sarcopenia. Lancet 2019;393(10191):2636–2646. https://doi.org/10.1016/S0140-6736(19)31138-9.

[15] Dasarathy S. Consilience in sarcopenia of cirrhosis. J Cachexia Sarcopenia Muscle 2012;3(4):225–237. https://doi.org/10.1007/S13539-012-0069-3.

[16] Cruz-Jentoft AJ, Baeyens JP, Bauer JM, et al. Sarcopenia: European consensus on definition and diagnosis: report of the European working group on sarcopenia in older People. Age Ageing 2010;39(4):412–423. https://doi.org/10.1093/AGEING/AFQ034.
[17] Montano-Loza AJ, Angulo P, Meza-Junco J, et al. Sarcopenic obesity and myosteatosis are associated with higher mortality in patients with cirrhosis. J Cachexia Sarcopenia Muscle 2016;7(2):126–135. https://doi.org/10.1002/JCSM.12039.
[18] Lattanzi B, Nardelli S, Pigliacelli A, et al. The additive value of sarcopenia, myosteatosis and hepatic encephalopathy in the predictivity of model for end-stage liver disease. Dig Liver Dis 2019;51(11):1508–1512. https://doi.org/10.1016/J.DLD.2019.09.004.
[19] Kalafateli M, Karatzas A, Tsiaoussis G, et al. Muscle fat infiltration assessed by total psoas density on computed tomography predicts mortality in cirrhosis. Ann Gastroenterol 2018;31(4):491–498. https://doi.org/10.20524/AOG.2018.0256.
[20] Li C wei, Yu K, Shyh-Chang N, et al. Pathogenesis of sarcopenia and the relationship with fat mass: descriptive review. J Cachexia Sarcopenia Muscle 2022;13(2):781–794. https://doi.org/10.1002/JCSM.12901.
[21] Correa-de-Araujo R, Harris-Love MO, Miljkovic I, et al. The need for standardized assessment of muscle quality in skeletal muscle function deficit and other aging-related muscle dysfunctions: a symposium report. Front Physiol 2017;8(FEB). https://doi.org/10.3389/FPHYS.2017.00087.
[22] de Franchis R, Bosch J, Garcia-Tsao G, et al. Baveno VII – renewing consensus in portal hypertension. J Hepatol 2022;76(4):959–974. https://doi.org/10.1016/j.jhep.2021.12.022.
[23] Cruz-Jentoft AJ, Bahat G, Bauer J, et al. Sarcopenia: revised European consensus on definition and diagnosis. Age Ageing 2019;48(1):16–31. https://doi.org/10.1093/AGEING/AFY169.
[24] Carey EJ, Lai JC, Wang CW, et al. A multicenter study to define sarcopenia in patients with end-stage liver disease. Liver Transpl 2017;23(5):625–633. https://doi.org/10.1002/LT.24750.
[25] Martin L, Birdsell L, MacDonald N, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013;31(12):1539–1547. https://doi.org/10.1200/JCO.2012.45.2722.
[26] Vehmas T, Kairemo KJ, Taavitsainen MJ. Measuring visceral adipose tissue content from contrast enhanced computed tomography. Int J Obes Relat Metab Disord 1996;20(6):570–573.
[27] Kvist H, Sjöström L, Tylén U. Adipose tissue volume determinations in women by computed tomography: technical considerations. Int J Obes 1986;10(1):53–67.
[28] Podsialdlo D, Richardson S. The timed "Up & Go": a test of basic functional mobility for frail elderly persons. J Am Geriatr Soc 1991;39(2):142–148. https://doi.org/10.1111/J.1532-5415.1991.TB01616.X.
[29] Lai JC, Tandon P, Bernal W, et al. Malnutrition, frailty, and sarcopenia in patients with cirrhosis: 2021 practice guidance by the American association for the study of liver diseases. Hepatology 2021;74(3):1611–1644. https://doi.org/10.1002/HEP.32049.
[30] Lai JC, Covinsky KE, Dodge JL, et al. Development of a novel frailty index to predict mortality in patients with end-stage liver disease. Hepatology 2017;66(2):564–574. https://doi.org/10.1002/HEP.29219.
[31] Pintile M. Competing risks. A practical perspective. Chichester: John Wiley and Sons Inc.; 2006.
[32] Lambert PC. The estimation and modeling of cause-specific cumulative incidence functions using time-dependent weights. The Stata Journal 2017;vol. 1:181–207.
[33] Fine PF, Gray RJ. A proportional hazards model for the subdistribution of a competing risk. J Am Stat Assoc 1999;94:496–509.
[34] Huber PJ. The behavior of maximum likelihood estimates under nonstandard conditions. In: Proceedings of the fifth berkeley symposium on mathematical statistics and probability. Berkley: University of California Press; 1967. p. 221–233.
[35] White HLJ. A heteroskedasticity-consistent covariance matrix estimator and a direct test for heteroskedasticity. Econometrica 1980;48:817–838.
[36] Kamath PSPS, Wiesner RHRH, Malinchoc M, et al. A model to predict survival in patients with end-stage liver disease. Hepatology 2001;33(2):464–470.
[37] Pugh RNH, Murray-Lyon IM, Dawson JL, et al. Transection of the oesophagus for bleeding oesophageal varices. Br J Surg 1973;60(8):646–649.
[38] Tachi Y, Kozuka A, Hirai T, et al. Impact of myosteatosis on skeletal muscle volume loss in patients with chronic liver disease. J Gastroenterol Hepatol 2018;33(9):1659–1666. https://doi.org/10.1111/JGH.14133.
[39] Geladari E, Alexopoulos T, Kontogianni MD, et al. The presence of myosteatosis is associated with age, severity of liver disease and poor outcome and may represent a prodromal phase of sarcopenia in patients with liver cirrhosis. J Clin Med 2023;12(9). https://doi.org/10.3390/JCM12093332.
[40] Davuluri G, Allawy A, Thapaliya S, et al. Hyperammonaemia-induced skeletal muscle mitochondrial dysfunction results in cataplerosis and oxidative stress. J Physiol 2016;594(24):7341–7360. https://doi.org/10.1113/JP272796.
[41] Kirk B, Al Saedi A, Duque G. Osteosarcopenia: a case of geroscience. Aging Med (Milton) 2019;2(3):147–156. https://doi.org/10.1002/AGM2.12080.
[42] Nathiya D, Raj P, Singh P, et al. Frailty predicting health-related quality of life trajectories in individuals with sarcopenia in liver cirrhosis: finding from BCAAS study. J Clin Med 2023;12(16). https://doi.org/10.3390/JCM12165348.
[43] Tandon P, Montano-Loza AJ, Lai JC, et al. Sarcopenia and frailty in decompensated cirrhosis. J Hepatol 2021;75(1):S147–S162. https://doi.org/10.1016/J.JHEP.2021.01.025. Suppl 1.

Keywords: cirrhosis; sarcopenia; myosteatosis; decompensation; survival; muscle function.
Received 11 February 2024; received in revised form 4 May 2024; accepted 13 May 2024; available online 21 May 2024

Journal of Hepatology, Volume *81*

Supplemental information

Myosteatosis is closely associated with sarcopenia and significantly worse outcomes in patients with cirrhosis

Simone Di Cola, Gennaro D'Amico, Paolo Caraceni, Filippo Schepis, Simone Loredana, Pietro Lampertico, Pierluigi Toniutto, Silvia Martini, Sergio Maimone, Antonio Colecchia, Gianluca Svegliati Barone, Carlo Alessandria, Alessio Aghemo, Saveria Lory Crocè, Luigi Elio Adinolfi, Maria Rendina, Lucia Lapenna, Enrico Pompili, Giacomo Zaccherini, Dario Saltini, Massimo Iavarone, Giulia Tosetti, Carolina Martelletti, Veronica Nassisi, Alberto Ferrarese, Ilaria Giovo, Chiara Masetti, Nicola Pugliese, Michele Campigotto, Riccardo Nevola, Manuela Merli, and for the EpatoSarco working group

Myosteatosis is closely associated with sarcopenia and significantly worse outcomes in patients with cirrhosis

Simone Di Cola, Gennaro D'Amico, Paolo Caraceni, Filippo Schepis, Simone Loredana, Pietro Lampertico, Pierluigi Toniutto, Silvia Martini, Sergio Maimone, Antonio Colecchia, Gianluca Svegliati Barone, Carlo Alessandria, Alessio Aghemo, Saveria Lory Crocè, Luigi Elio Adinolfi, Maria Rendina, Lucia Lapenna, Enrico Pompili, Giacomo Zaccherini, Dario Saltini, Massimo Iavarone, Giulia Tosetti, Carolina Martelletti[1], Veronica Nassisi, Alberto Ferrarese, Ilaria Giovo, Chiara Masetti, Nicola Pugliese, Michele Campigotto, Riccardo Nevola, Manuela Merli, for the *EpatoSarco* working group

Table of contents

List of investigators

Study site	Principal Investigator	Associate Investigator	No. patients enrolled
Rome	Manuela Merli (study coordinator)	Simone Di Cola Lucia Lapenna	30
Bologna	Paolo Caraceni	Enrico Pompili, Giacomo Zaccherini Giulia Iannone	44
Ferrara	Simone Loredana		44
Milan	Pietro Lampertico	Massimo Iavarone Giulia Tosetti Paola Serri Mariangela Bruccoleri	42
Udine	Pierluigi Toniutto		36
Turin	Silvia Martini	Carolina Martelletti	34
Modena	Filippo Schepis	Dario Saltini	30
Messina	Sergio Maimone	Veronica Nassisi	28
Verona	Antonio Colecchia	Alberto Ferrarese	23
Ancona	Gianluca Svegliati Barone		18
Turin	Carlo Alessandria	Ilaria Giovo	16
Milan	Alessio Aghemo	Chiara Masetti Nicola Pugliese Michele Campigotto	13
Trieste	Saveria Lory Crocè		12
Naples	Luigi Elio Adinolfi	Riccardo Nevola	11
Bari	Maria Rendina		11
Catania	Gaetano Bertino		9
Caltanissetta	Marcello Maida		7
L'Aquila	Clara Balsano	Nerio Iapadre	7

Verona	David Sacerdoti	Leonardo Antonio Natola	7
Salerno	Carolina Ciacci	Antonella Santonicola	6
Milan	Anna Ludovica Fracanzani	Annalisa Cespiati	5
Castellana Grotte	Raffaele Cozzolongo		5
Lamezia Terme	Lorenzo Antonio Surace		5
Naples	Alessandro Federico	Mario Romeo	4
Rome	Antonio Grieco	Giuseppe Marrone	3
Bologna	Luca Vizioli		3

Promotion and funding

This was a prospective observational study, performed on behalf of the *Italian Association for the Study of the Liver* (AISF). No funding was available for this study.

Supplementary tables

Table S1 . *Patients characteristics at inclusion according to the presence or absence of myosteatosis**

	Whole cohort	No Myosteatosis	Myosteatosis
Number	433	130 (30)	303 (70)
Age	57.1 (8.9)	56.7 (3.9)	61.0 (8.2) [0.0001]
Sex M	308 (71.1)	113 (85.6)	196 (64.7) [0.0009]
Etiology			
Alcohol	225 (51.9)	66 (50)	160 (52.8)
HCV	109 (25.2)	40(30,3)	69 (22.8)
HBV	27 (6.23)	11(7,6)	16 (5.3)
NASH	70 (16.5)	22 (16,7)	49 (16.2)
Autoimmune/Biliary disease	9 (2.1)	2 (1.5)	7 (2.3)
Others or undefined	60 (13.8)	15 (11.4)	45 (14.9)
Metabolic			
Diabetes	138 (31.8)	41 (31.1)	97 (32.0)
Arterial hypertension	160 (36.9)	40 (30.3)	121 (39.9)
Dyslipidemia	75 (17.3)	22 (16.7)	54 (17.8)
Clinical features			
Ascites	214 (49.4)	54 (40.9)	161 (53.1) [0.01]
E-G Varices╫	277 (63.9)	91 (68.9)	186 (61.4)
I-Prophylaxis ╞	221 (51)	74 (56.1)	147 (48.5)
NSBB	109 (25.2)	36 (27.7)	73 (24.1)
EBL	41 (9.5)	13 (10)	28 (9.24)
II-Prophylaxis §	71 (16.4)	25 (19.2)	46 (15.2)
Overt Hepatic encephalopathy	82 (18.9)	27 (20.5)	55 (18.5)
ANT [n=348]╪	17.1 (6.13)	18,5 (6.17)	16,4 (6.11) [<0.001]
AKI	19 (4.38)	4 (3.0)	16 (5.3)
Episodes of infection	55 (12.7)	7(5.3)	48 (15.8) [<0.001]
Severity of liver disease			
Child-Pugh			
A ┤	162 (37.4)	56 (43.1)	106 (35)
B ┤	196 (45.3)	58 (44.6)	139 (45.9)
C ┤	75 (17.3)	18 (13.8)	58 (19.1)
MELD score	12.99 (4.32)	12.0 (4.46)	13.47 (4.43) [<0.001]
MELD-Na score	14.3 (4.72)	13.0 (5.11)	14.9 (5.08) [<0.001]
MELD score < 15	303	108 (35.7)	195 (64.3)

MELD score ≥ 15	145	37 (25.5)	108 (74.5)
Bilirubin mg/dL	2.88 (4.52)	2.35 (4.53)	3.12 (4.52) [0.05]

*Data are presented as number of patients or means and % or SD in brackets, as appropriate, computed by the Student's t test for means and by the chisquare test for proportions.

^ The percentage is calculated based on the number of patients with alcoholic etiology.

¶ p value for significant differences from patients without muscle changes

╫ Esophago-Gastric varices

╪ANT= animal naming test, available in 348 patients

╞ Primary prophylaxis for variceal bleeding

§ Secondary prophylaxis for variceal bleeding, mostly NSBB+EVL

┤ Child-Pugh class A, B, C.

Abbreviations: MELD, model for end stage liver disease; NSBB, non-selective beta blockers; EVL, endoscopic variceal ligation;

Table S2. Univariable analysis for death, hospitalization and new decompensation by the Fine and Gray model.

Variable	Score	Sub-Hazard Ratio	p	95% Confidence Interval	
Death (LT competing)					
Sarcopenia	No=0; sarco=1	1.21	0.492	0.69	2.11
Myosteatosis	No=0; myo=1	2.86	0.010	1.28	6.41
Combined sarcopenia and myosteatosis	No=0; combined sarco-myo=1	1.39	0.011	1.07	1.79
Bilirubin	Continuous values	1.07	0.001	1.03	1.11
INR	Continuous values	2.98	<0.0001	1.89	4.71
Albumin	Continuous values	0.37	<0.0001	0.24	0.57
Creatinine	Continuous values	1.35	<0.0001	1.23	1.47
Age	Continuous values	1.01	0.327	0.98	1.04
Gender		1.06	0.841	0.58	1.95
OHE	No=0; yes=1	3.40	<0.001	1.95	5.94
Ascites	No=0; yes=1	4.34	<0.001	2.18	8.62
MELD	Continuous values	1.15	<0.001	1.10	1.21
Child-Pugh	5 tp 15 points	1.61	<0.001	1.40	1.84
Hospitalization (death and LT competing)					
Sarcopenia	No=0; sarco=1	1.01	0.907	0.78	1.31
Myosteatosis	No=0; myo=1	1.36	0.034	1.02	1.80
Combined sarcopenia and myosteatosis	No=0; combined sarco-myo=1	1.14	0.017	1.02	1.28
Bilirubin	Continuous values	0.99	0.659	0.96	1.02
INR	Continuous values	1.25	0.188	0.89	1.72
Albumin	Continuous values	1.06	0.590	0.85	1.31
Creatinine	Continuous values	1.33	<0.001	1.22	1.44
Age	Continuous values	1.00	0.546	0.99	1.01
Gender		1.11	0.445	0.84	1.45
OHE	No=0; yes=1	1.39	0.027	1.04	1.86

Ascites	No=0; yes=1	1.32	0.026	1.03	1.69
MELD	Continuous values	1.03	0.025	1.00	1.06
Child-Pugh	5 tp 15 points	1.10	0.006	1.03	1.18
First or further decompensation (death and LT competing)					
Sarcopenia	No=0; sarco=1	1.48	0.019	1.07	2.06
Myosteatosis	No=0; myo=1	1.29	0.166	0.89	1.87
Combined sarcopenia and myosteatosis	No=0; combined sarco-myo=1	1.07	0.268	0.94	1.23
Bilirubin	Continuous values	1.01	0.382	0.98	1.04
INR	Continuous values	1.36	0.055	0.99	1.86
Albumin	Continuous values	0.76	0.044	0.58	0.99
Creatinine	Continuous values	1.17	0.134	0.95	1.44
Age	Continuous values	1.00	0.997	0.98	1.01
Gender		0.98	0.937	0.68	1.41
OHE	No=0; yes=1	1.94	<0.001	1.67	2.77
Ascites	No=0; yes=1	2.99	<0.001	2.10	4.26
MELD	Continuous values	1.07	<0.001	1.04	1.10
Child-Pugh	5 tp 15 points	1.29	<0.001	1.19	1.39

Abbreviations: INR= international normalized ratio; OHE overt hepatic encephalopathy

Table S3. Significant risk predictors for death, hospitalization and new decompensation by the Fine and Gray model, and including MELD-Na.

Variable	Score	Sub-Hazard Ratio	p	95% Confidence Interval	
Including MELD-Na and excluding relevant single components					
Death (LT competing)					
Muscle changes ¶	No=0; I-sarco=1, I-myo =2; sarco-myo=3	1.33	0.086	0.95	1.84
MELD-Na	Continuous values	1.12	<0.0001	1.07	1.18
OHE	No=0; yes=1	2.08	0.011	1.18	3.66
Ascites	No=0; yes=1	2.51	0.010	1.24	5.09
Hospitalization (death and LT competing)					
Muscle changes ¶	No=0; I-sarco=1, I-myo =2; sarco-myo	1.18	**0.014**	1.03	1.35
MELD-Na	Continuous values	1.02	0.191	0.98	1.05
Albumin	g/L	1.28	0.047	1.00	1.63
Ascites	No=0; yes=1	1.39	0.054	0.99	1.95
First or further decompensation (death and LT competing)					
Muscle changes ¶	No=0; I-sarco=1, I-myo =2; sarco-mio=3	1.03	0.70	0.89	1.18
MELD-Na	Continuous values	1.03	0.039	1.00	1.06
Ascites	No=0; yes=1	2.52	<0.0001	1.73	3.67
OHE	No=0; yes=1	1.48	0.038	1.02	2.14

¶ In these analyses, each of the assessed muscle changes (i.e I-sarcopenia, I-myosteatosis, sarco-myosteatosis) were scored as absent/present: when none of them was significant, we included in the model a discrete variable scored as follows: no-changes=0, I-sarcopenia=1, I-myosteatosis=2, combined sarcopenia and myosteatosis=3.

Table S4. Adjusted prognostic role of muscle changes, by the Fine and Gray model, for death, hospitalization and new liver decompensation, including Child–Pugh score.

Variable	Score	Sub-Hazard Ratio	p	95% Confidence Interval	
Including Child-Pugh score and excluding relevant single components					
Death (LT as a competing event)					
Muscle changes¶	No=0; I-sarco=1, I-myo =2; sarco-myo=3	1.77	0.077	0.97	1.79
Child–Pugh score	5 tp15 points	1.59	<0.0001	1.38	1.83
Creatinine	mg/dL	1.29	<0.0001	1.19	1.41
Hospitalization (death and LT as competing events)					
I-Myosteatosis	No=0; yes=1	1.39	0.034	1.02	1.88
Child–Pugh score	5 tp15 points	1.1	0.022	1.01	1.18
Creatinine	mg/dL	1.31	<0.0001	1.20	1.43
Non elective hospitalization (death and LT as competing events)					
I-sarcopenia	No=0; yes=1	1.80	0.092	0.91	3.57
Child–Pugh score	5 tp15 points	1.35	<0.0001	1.22	1.50
New decompensation (death and LT as competing events)					
Muscle changes¶	No=0; I-sarco=1, I-myo =2; sarco-myo=3	0.94	0.44	0.80	1.10
Child–Pugh score	5 tp15 points	1.21	0.022	1.10	1.31

¶ In these analyses, each of the assessed muscle changes (i.e., I-sarcopenia, I-myosteatosis, and sarco-myosteatosis) was scored as absent/present; if none of them was significant, we included in the model a discrete variable scored as follows: no changes=0, I-sarcopenia=1, I-myosteatosis=2, and combined sarcopenia and myosteatosis=3.

Table S5. Significant risk predictors for death, hospitalization and new decompensation by the Fine and Gray model¶.

Variable	Score	Sub-Hazard Ratio	p	95% Confidence Interval	
Death (LT competing)					
Muscle changes	No=0; I-sarco=1, I-myo =2; sarco-myo	1.4	0.20	0.83	2.37
INR	Continuous values	3.06	0.002	1.53	6.11
Albumin	g/L	0.63	0.028	0.43	0.95
Creatinine	mg/dL	1.33	<0.0001	1.16	1.52
OHE	No=0; yes=1	2.39	0.003	1.33	4.29
Ascites	No=0; yes=1	3.25	0.005	1.44	7.3
Hospitalization (death and LT competing)					
I-Myosteatosis	No=0; yes=1	1.44	0.20	1.06	1.95
Creatinine	mg/dL	1.36	<0.0001	1.25	1.48
Ascites	No=0; yes=1	1.39	0.031	1.03	1.88
New decompensation (death and LT competing)					
Muscle changes	No=0; I-sarco=1, I-myo =2; sarco-mio	1.12	0.22	0.93	1.37
OHE	No=0; yes=1	2.38	<0.0001	1.47	3.87
Ascites	No=0; yes=1	3.04	<0.0001	1.85	5.01

¶ Models including model for end stage liver disease and Child-Pugh score are included in separate analyses shown in table 3 and supplementary table 4.

Abbreviations: I-sarco= isolated sarcopenia; I-myo= isolated myosteatosis; sarco-myo: sarcopenia and myosteatosis. INR= international normalized ratio; OHE hepatic encephalopathy

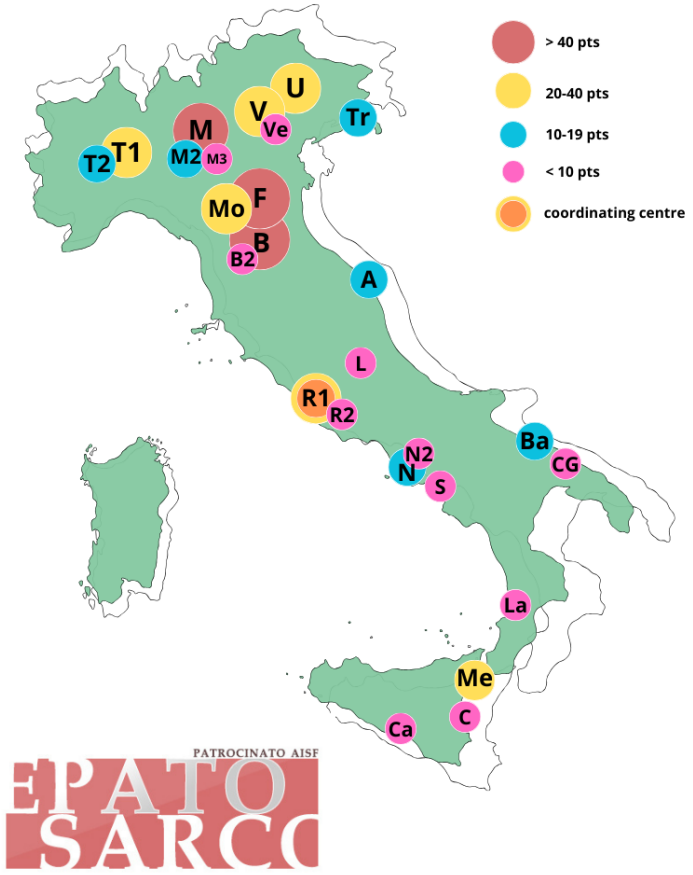
Table S6. Principal indications for abdominal CT-scan in our cohort.

CT-scan indication	**number (whole cohort 433 pts)**
Study of liver parenchyma (exclusion of HCC, US unreliable due to patients conformation …)	300
Evaluation of portal hypertension	44
Evaluation of splanchnic vein thrombosis	37
Miscellanea (feasibility of TIPS placement, abdominal pain, other …)	52

Abbreviations: HCC hepatocellular carcinoma; US, ultrasound; TIPS, Transjugular Intrahepatic Portosystemic Shunt

Supplementary figures

Fig. S1. Geographical distribution and details of centers involved in EpatoSarco multicenter study



Cod	City	Hospital	n
		NORTH OF ITALY	
B	BOLOGNA	Alma Mater Studiorum - University of Bologna	
F	FERRARA	Arcispedale S.Anna	
M	MILAN	Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico	
U	UDINE	Hepatology and Liver Transplantation Unit, Azienda Sanitaria Universitaria Friuli Centrale, University of Udine	
T1	TURIN	Gastrohepatology Unit AOU Città della Salute e della Scienza di Torino	
Mo	MODENA	Azienda Ospedaliero-Universitaria of Modena	
V	VERONA	Gastroenterology, Verona University Hospital, Ospedale Borgo Trento	
T2	TURIN	Division of Gastroenterology and Hepatology, Città della Salute e della Scienza Hospital, University of Turin	
M2	MILAN	IRCCS Humanitas Research Hospital, Rozzano	
Tr	TRIESTE	Clinica Patologie del Fegato, Azienda Sanitaria Universitaria Giuliano Isontina, Trieste, Italy	
Ve	VERONA	Liver Unit, Department of Medicine, University and Azienda Ospedaliera Universitaria Integrata of Verona	
M3	MILAN	General Medicine and Metabolic Diseases, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico	
B2	BOLOGNA	Internal Medicine Unit for the Treatment of Severe Organ Failure, IRCCS Azienda Ospedaliero-Universitaria di Bologna	
		CENTER OF ITALY	
R1	ROME	Department of Translational and Precision Medicine, Sapienza University of Rome	
A	ANCONA	Liver Injury and Transplant Unit, Ospedali Riuniti	
R2	ROME	Internal and Liver Transplant Medicine Unit, Fondazione Policlinico Universitario Agostino Gemelli IRCCS	
		SOUTH OF ITALY	
Me	MESSINA	Division of Medicine and Hepatology, University Hospital of Messina, Messina,	
N	NAPLES	AOU Vanvitelli, Napoli, Piazza Miraglia, UOC di Medicina Interna	
Ba	BARI	Section of Gastroenterology, Department of Emergency and Organ Transplantation, University of Bari	
C	CATANIA	Hepatology Unit Department of Clinical and Experimental Medicine University of Catania, Policlinico "Rodolico"	
Ca	CALTANISSETTA	Gastroenterology and Endoscopy Unit, S. Elia-Raimondi Hospital	
L	L'AQUILA	Department of Life, Health, Environmental Sciences, School of Emergency and Urgency Medicine	
S	SALERNO	Department of Medicine, Surgery and Dentistry, "Scuola Medica Salernitana", University of Salerno	
CG	CASTELLANA GROTTE	Division of Gastroenterology, National Institute of Gastroenterology S De Bellis	
La	LAMEZIA TERME	Traveler and Migration Medicine Center, ASP Catanzaro	
N2	NAPLES	Department of Clinical and Experimental Medicine, Second University of Naples	

Fig. S2. Different distribution of muscle alterations (red, isolated sarcopenia; blue, isolated myosteatosis; yellow, combinated sarco-myosteatosis; grey , no muscle alterations) between male and females

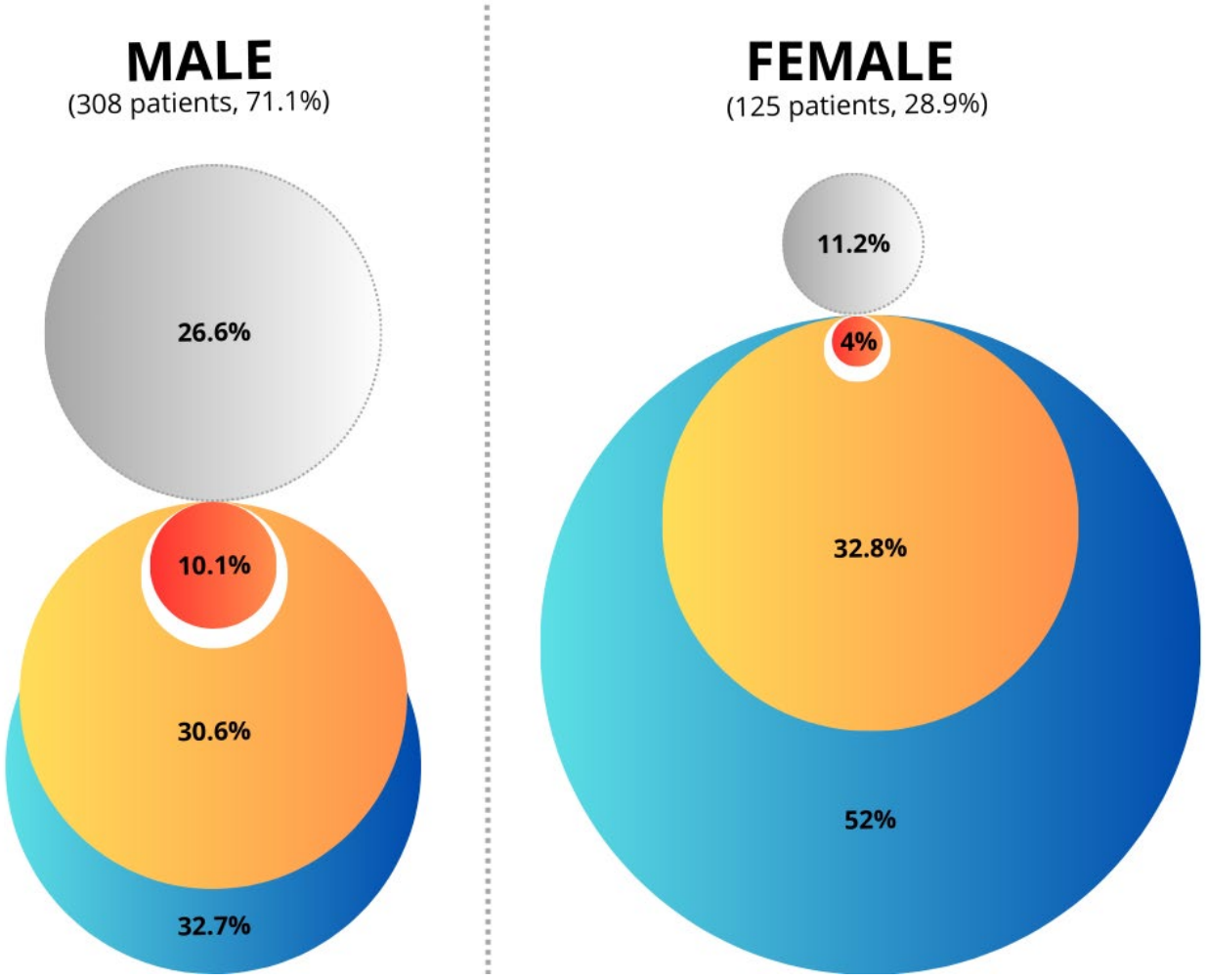


Fig. S3. Relationship between functional tests and frailty index with L3-SMI and muscle attenuation on CT scans. Each of the 4 panels represent the distribution of the assessed parameters in single patients (solid circles) and the corresponding fitted regression line. R 2 and p values refer to the corresponding regression analysis. Panel A: hand-grip test measured in Kilograms vs muscle surface in squared cm measured at the level of third or fourth lumbar vertebra. Panel B: hand- grip test vs muscle attenuation at CT scan, measured in Hounsfield units (HU). Panel C: up and go test measured in seconds vs muscle attenuation, HU. Panel D: frailty index vs muscle attenuation (HU)