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Renal outcome and plasma methylmalonic acid levels after isolated or combined liver or kidney transplantation in patients with methylmalonic acidemia: A multicenter analysis

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ABSTRACT

Background: Methylmalonic acidemia (MMAemia) is characterized by accumulation of methylmalonic acid (MMA) in all body tissues. To minimize disease-related complications, isolated kidney (KTx), liver (LTx) or combined liver-kidney transplantation (LKTx) have been suggested. However, the impact of these different transplant strategies on outcome are unclear.

Methods: In this multicenter retrospective observational study, we compared plasma MMA levels and estimated glomerular filtration rate (eGFR) data of 83 patients. Sixty-eight patients (82%) had a mut^0-type MMAemia, one patient had a mut$^-$-type MMAemia, and seven (7.3%) had an inherited defect in cobalamin metabolism (cblA- or cblB-type MMAemia). Median observation period was 3.7 years (0–15.1 years).

Results: Twenty-six (31%) patients underwent KTx, 24 (29%) LTx and 33 (40%) LKTx. Posttransplant, mean plasma MMA concentration significantly decreased in all three cohorts; but at month 12, plasma MMA in KTx (1372 ± 1101 μmol/L) was 7.8-fold higher than in LTx (176 ± 103 μmol/L; $P < 0.001$) and 6.4-fold higher than in LKTx (215 ± 110 μmol/L; $P < 0.001$). Comparable data were observed at month 24. At time of transplantation, mean eGFR in KTx was 18.1 ± 24.3 mL/min/1.73 m^2, in LTx 99.8 ± 29.9 mL/min/1.73 m^2, and in LKTx 31.5

Abbreviations: eGFR, estimated glomerular filtration rate; KTx, kidney transplantation; LTx, liver transplantation; LKTx, combined liver-kidney transplantation; 2-MCA, 2-methylcitrate; MMA, methylmalonic acid; MMAemia, methylmalonic acidemia.

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± 21.2 mL/min/1.73 m^2. At month 12 posttransplant, mean eGFR in KTx (62.3 ± 30.3 mL/min/1.73 m^2) was 33.4% lower than in LTx (93.5 ± 18.3 mL/min/1.73 m^2; $P = 0.0053$) and 25.4% lower than in LKTx (83.5 ± 26.9 mL/min/1.73 m^2; $P = 0.0403$).

Conclusions: In patients with isolated MMAemia, LTx and LKTx lead to markedly lower plasma MMA levels during the first 2 years posttransplant than KTx and are associated with a better preservation of kidney function. LTx should therefore be part of the transplant strategy in MMAemia.

1. Introduction

Methylmalonic acidemia (MMAemia) defines a heterogeneous group of autosomal recessive disorders characterized by impaired metabolism of methylmalonyl-CoA, an intermediary product of the common catabolic pathways of the amino acids isoleucine, methionine, threonine and valine, the side chain of cholesterol, and odd-chain fatty acids, as well as propionate arising from the intestinal microbiome [1]. It is a rare disorder with an estimated cumulative incidence of approximately 1–2 cases per 100,000 newborns [2]. The most frequent form of isolated MMAemia is caused by inherited deficiency of 5′-deoxyadenosylcobalamin-dependent methylmalonyl-CoA mutase (MMUT). MMUT deficiency can be complete (mut^0) or partial (mut^-); the former does not respond to pharmacological doses of hydroxocobalamin (vitamin B_{12}). Furthermore, defects in the genes encoding key enzymes of cobalamin metabolism, particularly *MMAA* (cblA-type MMA), *MMAB* (cblB-type MMA), and *MMADHC* (variant 2 cblD-type MMA), are alternative causes of isolated MMA, commonly responding to hydroxocobalamin [2].

MMA is biochemically characterized by the accumulation of methylmalonic acid in all body tissues and fluids [3]. Most patients present within the first days or weeks of life with acute metabolic crises. Target organ damage include recurrent pancreatitis, neurological symptoms with seizures, developmental delay, movement disorders, stroke-like events, psychiatric symptoms and chronic kidney disease [2]. Even though medical management and diet can be effective in reducing the frequency and severity of symptoms, recurrent metabolic crises are not reliably prevented in all affected individuals, and target organ damage and neurological impairment progress. Long-term complications of patients include short stature, global developmental delay and cognitive deficits, movement disorders due to basal ganglia lesions, sensorineural hearing loss, optic atrophy, epilepsy, neurological deficits after encephalopathy and/or cerebrovascular accidents, pancreatitis, prolonged QT_c intervals and cardiomyopathy, and progressive kidney disease [2,4]. Progressive kidney diseases in MMUT-type MMAemia has recently been explained by metabolite-induced mitochondrial dysfunction, exacerbated by anomalies in PINK1/Parkin-mediated mitophagy with concomitantly increased organelle ageing and epithelial stress [4–7]. Noteworthily, disease severity varies between known forms of isolated MMA, depending on their responsiveness to hydroxocobalamin treatment [8,9]. Individuals with non-responsive MMUT-type MMAemia present with a severe form, while those with the responsive cblA-type MMAemia show a more attenuated clinical phenotype. Hence, increasing knowledge about the progressive multiorgan phenotype and disease-specific differences of MMAemia patients who survived into adulthood highlight the urgent need for more effective therapeutic measures [10], which should be adapted to the individual clinical severity.

Liver transplantation [11–15], frequently performed during early infancy [16,17], or combined liver-kidney transplantation [14,15,18,19] have been considered as alternative options to medical treatment, but also isolated kidney transplantation has been proposed [15,20–23]. Several reports suggest this procedure as an alternative and safer strategy, because the enzyme activity provided by the transplanted kidney could be sufficient to partially restore the metabolic defect [20].

Following transplantation, plasma and urine methylmalonic acid levels improve and the frequency of metabolic crises decreases despite persistent metabolic abnormalities [12,15,24]. Neurological complications may still arise, because MMA levels in cerebrospinal fluid samples remain elevated after transplantation [15,24,25,27], possibly as a result of the persisting underlying enzymatic defect in the central nervous system combined with the physiological lack of an effective efflux transporter for di- and tricarboxylic acid across the blood-brain barrier, resulting in entrapment of toxic MMA and 2-MCA in the brain compartment [28].

From the conflicting data reported above, it is clear that the best transplant strategy is still a matter of debate. Kidney transplantation is obviously required when severe chronic kidney disease, but it only partially restores enzyme activity, which may not be sufficient for the prevention of MMAemia-related symptoms in more severely affected patients [20]. Possible alternatives include liver transplantation or combined liver-kidney transplantation, which provide a larger enzymatic activity, but are associated with a higher perioperative mortality [3,4,11–14]. In this retrospective observational multicenter study from Europe and the United States (US), we aimed to compare plasma MMA levels and renal outcome following different transplant strategies in patients with MMAemia.

2. Methods

2.1. Study design and conduct

We conducted a retrospective, international, multicenter, longitudinal cohort analysis by use of the CERTAIN Registry (www.certain-registry.eu). A comprehensive description of the CERTAIN Registry including the data validation process has been published previously [29]. Written informed consent was obtained from all parents/guardians to participate in the registry, with assent from patients when appropriate for their age. The CERTAIN Registry has been approved by the ethics committee of each contributing center and is kept in full accordance with the principles of the Declaration of Helsinki and Good Clinical Practice guidelines. Transplant centers not being part of the CERTAIN network contributed to this study through the European Society for Pediatric Nephrology in Europe and through the Midwest Pediatric Nephrology Consortium in the United States. All contributing centers obtained authorization from their respective institutional review board. The study was designed, analyzed and reported according to the STROBE guidelines (https://www.strobe-statement.org).

Inclusion criteria were (i) patients with a proven genetic or biochemical diagnosis of isolated MMAemia (*i.e.* MMUT-type, cblA-type, and cblB-type MMAemia, and variant 2 cblD), and (ii) kidney transplantation, liver transplantation or combined liver-kidney transplantation in the time period between January 2003 and December 2018. No exclusion criteria were considered. Because of the multicenter design, data capture was limited and included the type of transplantation, causes and point in time of deaths, estimated glomerular filtration rate (eGFR) [30] and plasma MMA concentration. Data were recorded before transplantation and at month 1, 6 and 12, and 24 posttransplant. MMA plasma levels were measured locally by means of gas-chromatography coupled with mass spectrometry [30]. Median observation period was 3.7 years (0–15.1 years).

2.2. Statistical analysis

All analyses were performed with Stata 13.1 (StataCorp LLC, College Station, TX) and R Version 4.1.1 software. Patient demographic and clinical data were summarized for categorical variables as counts and percentages and for continuous variables as mean and standard deviation. Percentages were calculated excluding missing values. Chi-squared and Mann–Whitney *U* test were used for the comparison of demographic characteristics. To analyze MMA and eGFR profiles over time, generalized linear mixed models (GLMM) were used (R package 'glmmTMB'). For MMA, a GLMM with Gamma family function and log-link, for eGFR a GLMM with Tweedie family function and log-link was chosen. Selection of family function for GLMM based on Akaike information criteria (AIC) and deviance. Gamma and Tweedie function appropriately deals with right-skewed distribution of MMA and eGFR values as well as with their positive range. In addition, GLMM with the covariates sex (female or male) and mutation (mut^0-type or other) were computed to test for further effects on MMA and eGFR values. At least two MMA or eGFR values per patient were required for including a patient in this analysis. Post-hoc comparisons in GLMM were computed with estimated marginal means (R package 'emmeans') and Tukey method for *P* value adjustment. There were several missing values for MMA and eGFR at each posttransplant point in time. Therefore, we computed a sensitivity analysis for MMA and eGFR values at the time points pretransplant, month 1, month 6, month 12, and month 24 posttransplant with a sub-sample of patients in whom at least three MMA or eGFR values were available. To correlate pooled MMA and eGFR values, Pearson Product moment correlation coefficient was used.

3. Results

Eighty-three transplanted patients with MMAemia were analyzed, 48 patients from 15 European centers and 35 patients from four centers in the United States (US). Patient and transplant characteristics are shown in Table 1. The median observation period was 3.7 years (range, 0–15.1 years). One-year data were obtained from 67 patients and 2-year data from 49 patients (Fig. 1). Sixty-eight patients (82%) had a mut^0-type MMAemia, one patient had a mut^--type-MMAemia and seven (7.3%) had a defect in the synthesis of 5′-deoxyadenosylcobalamin (cblB-type, $n = 6$; cblA-type, $n = 1$). A mutational analysis was not performed in the seven remaining cases, in whom the diagnosis was based on biochemical criteria (median MMA plasma level, 835 μmol/L; range, 535–6940).

Twenty-six (31%) patients underwent kidney transplantation, 24 (29%) liver transplantation and 33 (40%) combined liver-kidney transplantation. No patient received a sequential liver-kidney transplant. Twenty-five of 26 kidney transplantations (96%) were performed in Europe, one (4%) in the US. The respective distribution for liver transplantation ($n = 24$) were 10 (42%) in Europe and 14 (58%) in the US, for combined liver-kidney transplantation ($n = 33$) 13 (39%) in Europe and 20 (61%) in the US. Patients of the liver transplant cohort were significantly younger (median age 1.8 years) than patients of the kidney (median 11.1 years) and of the liver-kidney transplant cohort (median 9.5 years) (Table 1). Of 68 patients with a mut^0-type MMAemia, 18 (26.5%) received a kidney transplant, 23 (33.8%) a liver transplant, and 27 (39.7%) underwent combined liver-kidney transplantation.

The number and causes of deaths in the three transplant cohorts is given in Table 3. Five of 26 (19%) patients in the kidney transplant cohort, 3 of 24 (13%) in the liver transplant cohort and 2 of 33 (6%) in the combined liver-kidney transplant cohort died during the time of observation. Numbers were too low for a meaningful statistical analysis.

Pretransplant mean plasma MMA concentration in the kidney transplantation cohort (3113 ± 2636 μmol/L) was 7-fold higher than in the liver transplant (433 ± 269 μmol/L; $P < 0.001$) and 2.3-fold higher than in the combined liver-kidney transplant cohort (1368 ± 1801 μmol/L; $P = 0.006$; Table 1). The respective plasma MMA concentrations at month 1, 6 and 12 posttransplant are given in Table 1 and

Table 1
Patient demographics and clinical characteristics.

	Entire cohort	Kidney Tx	Liver Tx	Combined liver-kidney Tx	*P* value LTx *vs.* KTx	*P* value LKTx *vs.* KTx	*P* value LTx *vs.* LKTx
Patient number, n (%)	83	26 (31)	24 (29)	33 (40)	n.s.	n.s.	n.s.
Age at transplantation (years)	8.7 (0.6–38.9)	11.1 (4.4–38.9)	1.8 (0.6–16.8)	9.5 (2.7–21.6)	<0.001	n.s	<0.001
Male gender, n (%)	46 (55.4)	16 (34.8)	12 (26.1)	18 (39.1)	n.s.	n.s.	n.s.
mut^0-type MMAemia, n (%)	68 (81.9)	18 (26.5)	23 (33.8)	27 (39.7)	n.s.	n.s	n.s
Duration of follow-up (years)	3.7 (0.0–15.1)	3.6 (0.7–12.6)	2.6 (0.4–15.1)	3.7 (0.0–13.2)	n.s	n.s	n.s.
Plasma MMA concentration (μmol/L) mean ± SD (n)							
Pretransplant		3113 ± 2636 ($n = 19$)	433 ± 269 ($n = 20$)	1368 ± 1801 ($n = 27$)	<0.0001	0.0009	0.0001
Month 1 posttransplant		813 ± 575 ($n = 15$)	121 ± 95.4 (n = 20)	152 ± 116 ($n = 28$)	<0.0001	<0.0001	0.3809
Month 6 posttransplant		1291 ± 1274 (n = 14)	171 ± 132 ($n = 18$)	213 ± 101 ($n = 21$)	<0.0001	<0.0001	0.4134
Month 12 posttransplant		1372 ± 1101 (n = 18)	176 ± 103 ($n = 13$)	215 ± 110 (n = 19)	<0.0001	<0.0001	0.4916
eGFR (mL/min/1.73 m^2) mean ± SD (n)							
Pretransplant (n)		18.1 ± 24.3 ($n = 23$)	99.8 ± 29.9 (n = 19)	31.5 ± 21.2 ($n = 29$)	<0.0001	0.0024	<0.0001
Month 1 posttransplant		60.9 ± 22.0 (n = 21)	100.5 ± 37.0 ($n = 17$)	87.6 ± 33.6 (n = 29)	0.0007	0.0095	0.4691
Month 6 posttransplant		69.1 ± 22.2 (n = 21)	95.0 ± 41.1 (n = 17)	82.0 ± 26.5 ($n = 24$)	0.0432	0.3192	0.5010
Month 12 posttransplant		62.3 ± 30.3 (n = 21)	93.5 ± 18.3 (n = 15)	83.5 ± 26.9 ($n = 25$)	0.0053	0.0403	0.5725

Data are given as median (range) or mean ± SD, as indicated. Abbreviations: KTx, kidney transplantation, LTx, liver transplantation; LKTx, combined liver-kidney transplantation; SD, standard deviation; Tx, transplantation.

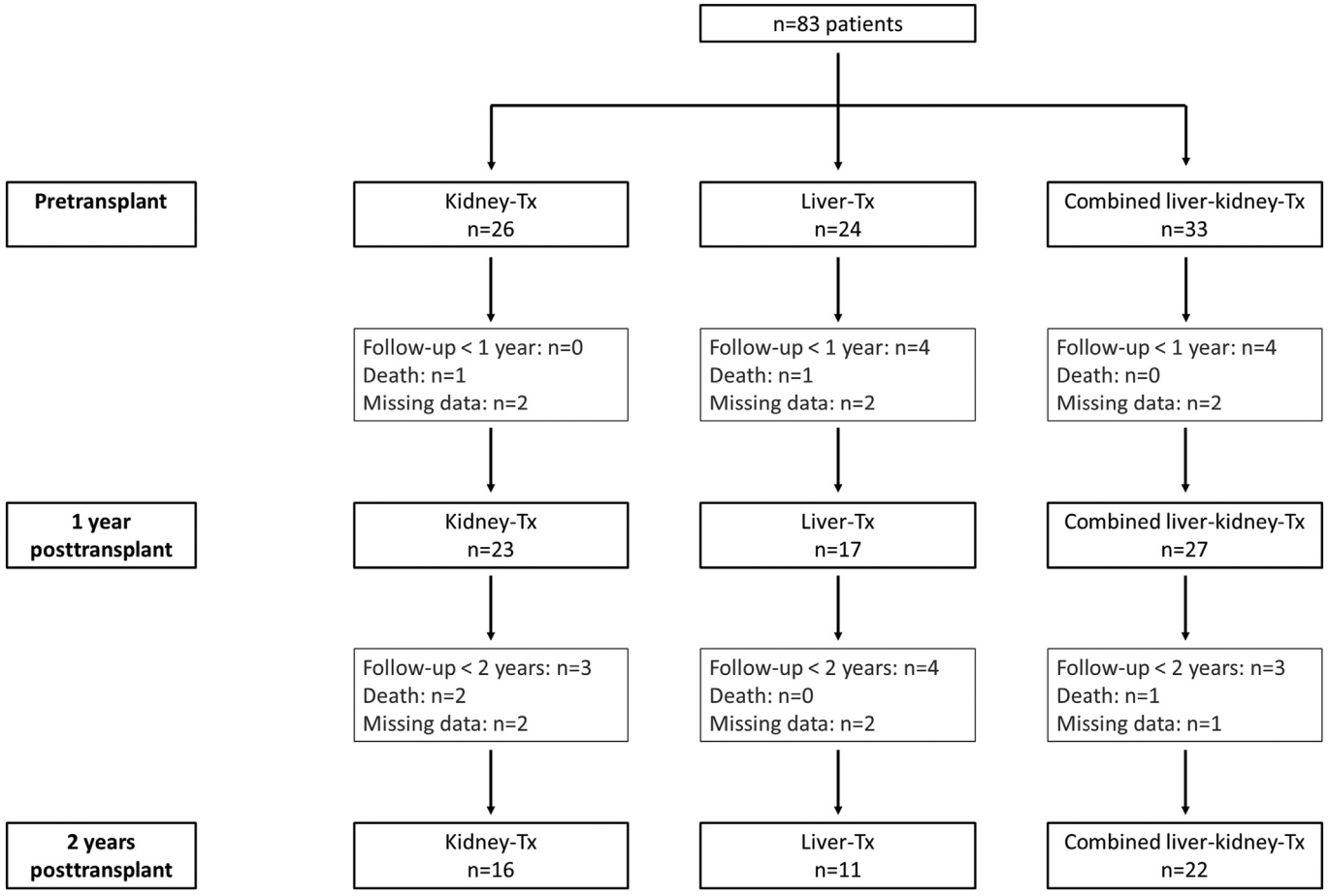

Fig. 1. Flowchart showing the number of patients available for analysis.

depicted in Fig. 2. Compared to baseline (pretransplant), mean plasma MMA concentrations significantly decreased in all three cohorts at month 1 and 6 posttransplant; but, at both time-points, the mean plasma MMA concentration of the kidney transplant cohort was significantly higher ($P < 0.001$) than of the liver transplant or combined liver-kidney transplant cohort. At month 12 posttransplant, plasma MMA

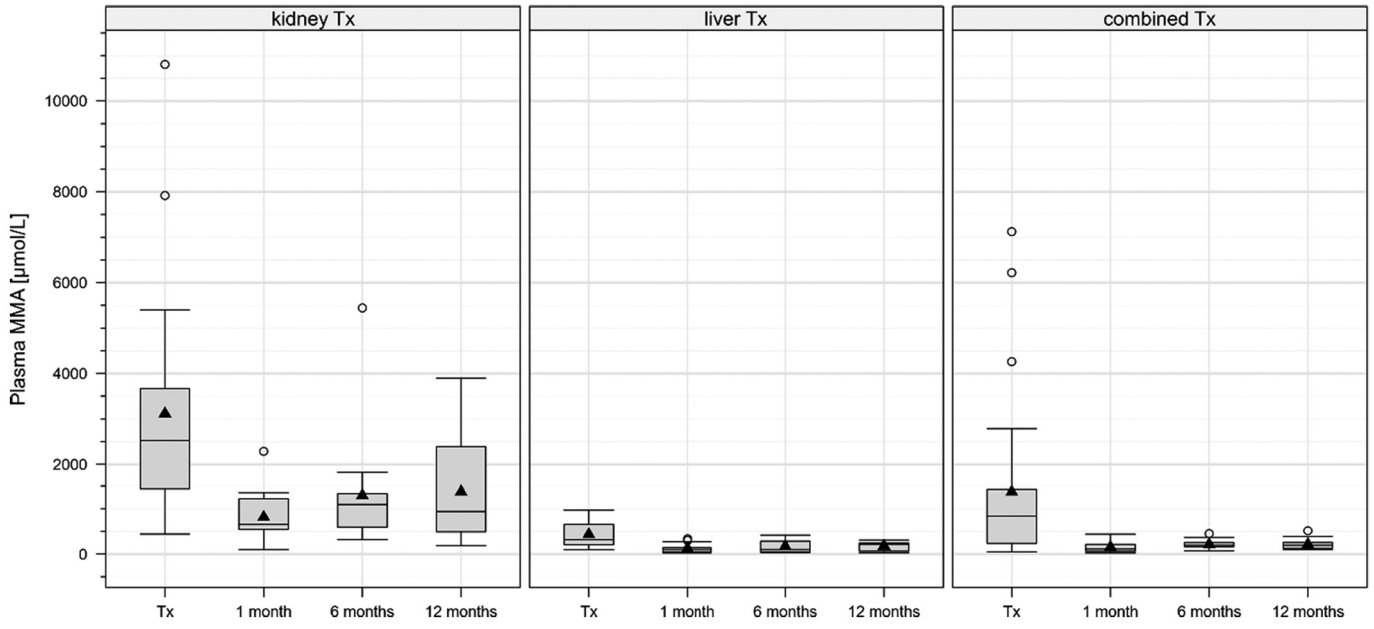

Fig. 2. Plasma MMA levels prior to transplantation (Tx) and at month 1, 6, and 12 posttransplant in patients after kidney, liver, or combined liver-kidney transplatation. Mean is shown as a black triangle; box and whisker plots with the box represent the median, the 25th and 75th percentiles, while whiskers show the highest and lowest non-outlier values. Outliers were identified using upper/lower quartile ±1.5 times IQR.

remained low in the liver transplant and the combined liver-kidney transplant cohort, while it tended to increase again in the kidney transplant cohort ($P = 0.06$ compared to month 1). At month 12 posttransplant, mean plasma MMA concentration in the kidney transplant cohort (1372 ± 1101 μmol/L) was 7.8-fold higher than in the liver transplant (176 ± 103 μmol/L; $P < 0.001$) and 6.4-fold higher than in the combined liver-kidney transplant cohort (215 ± 110 μmol/L; $P < 0.001$; Table 1). Comparable data were observed at month 24: mean plasma MMA remained low in the liver transplant (217 ± 94 μmol/L) and combined liver-kidney transplant cohort (265 ± 105 μmol/L), while elevated plasma MMA persisted in the kidney transplant cohort (1075 ± 937 μmol/L).

At time of transplantation, 12 of 26 patients (46%) of the kidney transplant cohort and 7 of 33 patients (21%) of the combined liver-kidney transplant cohort had kidney failure requiring chronic dialysis therapy. Most of the patients in the kidney transplant cohort who did not yet receive chronic dialysis therapy ($n = 14$) had Stage IV or V chronic kidney disease (mean eGFR 18.1 ± 24.3 mL/min/1.73 m^2; range, 14–80). In the liver transplant cohort, mean eGFR at time of transplantation was 99.8 ± 29.9 mL/min/1.73 m^2, in the combined liver-kidney transplant cohort 31.5 ± 21.2 mL/min/1.73 m^2.

At month 12 posttransplant, mean eGFR in the kidney transplant cohort (62.3 ± 30.3 mL/min/1.73 m^2) was 33.4% lower than in the liver transplant cohort (93.5 ± 18.3 mL/min/1.73 m^2, $P = 0.0053$) and 25.4% lower than in the combined liver-kidney transplant cohort (83.5 ± 26.9 mL/min/1.73 m^2, $P = 0.0403$) (Table 1, Fig. 3). Mean eGFR in the kidney transplant cohort at month 12 posttransplant was also significantly lower ($P = 0.0078$) than the mean eGFR (71.4 ± 34.8 mL/min/1.73 m^2) of 1112 patients aged <18 years documented in the CERTAIN Registry, who had received a kidney transplant for primary kidney diseases other than MMA. Because there were several missing values for MMA and eGFR at each posttransplant point in time we computed a sensitivity analysis for MMA and eGFR values with a sub-sample of patients in whom at least three MMA or eGFR values were available over a period of 24 months posttransplant (Table 2). Comparable results were obtained as for the entire cohort (Table 1). At month 24, the observed difference among the three transplant cohorts regarding plasma MMA concentrations and eGFR values were comparable to the differences observed at month 12. Three of five deaths in the kidney transplant cohort occurred during the second posttransplant year; their respective eGFR data were not included in this analysis.

Fig. 4 shows the relationship between eGFR and plasma MMA concentrations. This analysis is based on pooled data from months 1, 6 and 12 posttransplant for patients, in whom both MMA plasma concentrations and eGFR data were available. There was a significant negative correlation ($P < 0.0001$) between eGFR and plasma MMA concentration in all three transplant cohorts; patients in the kidney transplant cohort had by far the highest MMA plasma concentrations and lowest eGFR values.

4. Discussion

This multicenter cohort study of patients affected by isolated MMAemia and having received an organ transplantation represents the largest series up to now. These patients underwent different transplant procedures, isolated kidney transplantation, isolated liver transplantation or combined liver/kidney transplantation. The main result is that patients after liver transplantation alone or in combination with a kidney transplant had a better outcome than those who had received an isolated kidney transplant.

Pretransplant, patients in the liver transplant cohort had lower plasma MMA levels than the other two cohorts. This is most likely due to their young age and preserved kidney function. Plasma MMA is cleared from the circulation by glomerular filtration, and loss of kidney function contributes to MMA accumulation [31,32]. Kidney transplantation or combined liver/kidney transplantation was usually performed in children at a later stage of the disease when the detrimental effect of toxic metabolites, impaired mitophagy, enhanced mitochondrial ageing and dysfunction with concomitant epithelial stress had already induced chronic kidney disease [6]. Increased plasma MMA is a specific diagnostic marker of the disease [2]; its circulating concentration together with 2-methylcitrate (2-MCA) are higher in older patients and negatively correlate with eGFR, consistent with the concept that these molecules are cleared from the circulation by the kidney. Loss of kidney function contributes to their accumulation in plasma in parallel with the

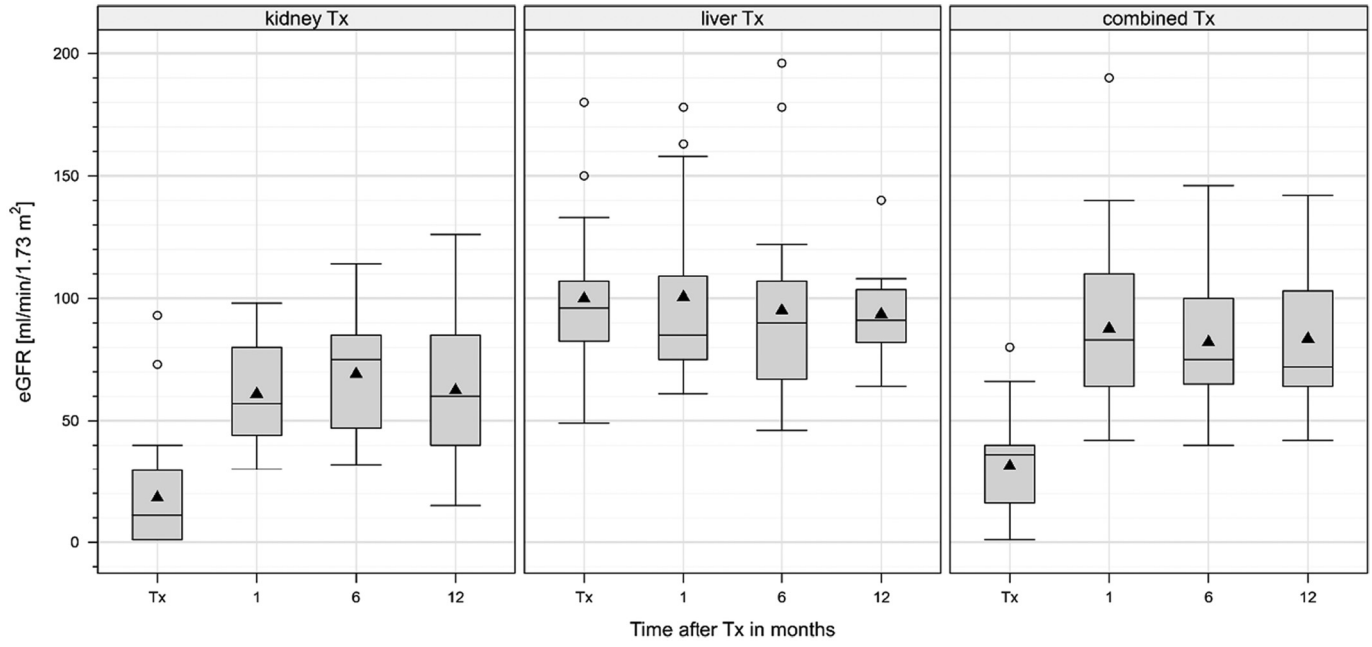

Fig. 3. Estimated glomerular filtration rate (eGFR) data prior to transplantation (Tx) and at month 1, 6, and 12 posttransplant in patients after kidney, liver, or combined liver-kidney transplatation. Mean is shown as a black triangle; box and whisker plots with the box represent the median, the 25th and 75th percentiles, while whiskers show the highest and lowest non-outlier values. Outliers were identified using upper/lower quartile ±1.5 times IQR.

Table 2
Sensitivity analysis for MMA and eGFR values with a sub-sample of patients in whom at least three MMA ($n = 59$ patients) or eGFR values ($n = 68$ patients) were available.

	Entire subsample	Kidney Tx	Liver Tx	Combined liver-kidney Tx	*P* value LTx *vs.* KTx	*P* value LKTx *vs.* KTx	*P* value LTx *vs.* LKTx
Plasma MMA concentration (μmol/L)							
Patient number, n (%)	59	16 (27)	19 (32)	24 (41)	n.s.	n.s.	n.s.
Age at transplantation (years)	8.55 (0.6–25)	11.8 (4.4–25.0)	2.9 (0.6–16.8)	10.6 (2.7–21.6)	<0.001	n.s	<0.001
Male gender, n (%)	32 (54.2)	9 (28.1)	10 (31.3)	13 (40.6)	n.s.	n.s.	n.s.
mut^0-type MMAemia, n (%)	68 (81.9)	18 (26.5)	23 (33.8)	27 (39.7)	n.s.	n.s	n.s
Duration of follow-up (years)	4.2 (0.0–15.2)	4.1 (1.2–10.7)	4.7 (0.4–15.1)	3.9 (0.0–13.2)	n.s	n.s	n.s.
Pretransplant		3113 ± 2636 (n = 19)	433 ± 269 (n = 20)	1368 ± 1801 (n = 27)	<0.0001	0.0009	0.0001
Month 1 posttransplant		817 ± 573 (n = 15)	114 ± 85 (n = 19)	139 ± 98 (n = 24)	<0.0001	<0.0001	0.3590
Month 6 posttransplant		1279 ± 1229 (n = 15)	179 ± 128 (n = 19)	218 ± 118 (*n* = 24)	<0.0001	<0.0001	0.2256
Month 12 posttransplant		1434 ± 1079 (n = 15)	182 ± 101 (n = 14)	249 ± 148 (*n* = 22)	<0.0001	<0.0001	0.1801
Month 24 posttransplant		1149 ± 694 (*n* = 8)	217 ± 149 (*n* = 9)	244 ± 188 (n = 13)	<0.0001	<0.0001	0.9828
eGFR (mL/min/1.73 m^2)							
Patient number, n (%)	68	22 (32)	17 (25)	29 (43)	n.s.	n.s.	n.s.
Age at transplantation (years)	9.86 (0.6–38.9)	13.9 (4.2–38.9)	2.6 (0.6–16.8)	11.0 (2.7–21.6)	<0.001	n.s	<0.001
Male gender, n (%)	37 (54.4)	13 (35.2)	8 (21.6)	16 (43.2)	n.s.	n.s.	n.s.
mut^0-type MMAemia, n (%)	56 (89.9)	17 (30.3)	16 (28.6)	23 (41.1)	n.s.	n.s	n.s
Duration of follow-up (years)	4.8 (0.0–15.1)	4.9 (1.2–12.6)	4.5 (0.4–15.1)	4.6 (0.0–13.2)	n.s	n.s	n.s.
Pretransplant mean ± SD (n)		18.1 ± 24.6 (n = 22)	97.2 ± 32.7 (n = 17)	28.3 ± 19.8 (n = 29)	<0.0001	0.0388	<0.0001
Month 1 posttransplant		61.1 ± 22.3 (n = 21)	98.5 ± 39.4 (*n* = 17)	82.8 ± 36.2 (n = 28)	0.0022	0.0730	0.2687
Month 6 posttransplant		69.3 ± 22.3 (*n* = 21)	93.2 ± 42.7 (n = 17)	78.2 ± 27.9 (n = 27)	0.0600	0.5757	0.3079
Month 12 posttransplant		62.5 ± 29.6 (n = 22)	90.5 ± 23.0 (n = 15)	79.3 ± 28.4 (n = 28)	0.0090	0.1356	0.3508
Month 24 posttransplant		58.6 ± 21.8 (n = 14)	80.0 ± 29.1 (*n* = 10)	69.9 ± 20.7 (n = 20)	0.0367	0.1922	0.5462

Data are given as median (range) or mean ± SD, as indicated. Abbreviations: KTx, kidney transplantation, LTx, liver transplantation; LKTx, combined liver-kidney transplantation; SD, standard deviation; Tx, transplantation.

progressive reduction of GFR [32]. Before onset of CKD, plasma and urine concentrations of MMA allow discrimination of presumed severity levels and predict a more severe clinical course. Once the disease has progressed to CKD, the plasma concentration of MMA is only of limited predictive value for the severity of the disease [26].

Following organ transplantation, plasma MMA levels decreased at month 1 posttransplant in all three cohorts, but the mean MMA level in the kidney transplant cohort was 6.7-fold higher than in the liver transplant cohort and 5.3-fold higher than in the combined liver/kidney transplant cohort (Table 1). During the first year posttransplant, plasma MMA increased again to 1372 ± 1101 μmol/L at month 12 posttransplant, while it remained markedly lower in the liver transplant cohort (176 ± 103 μmol/L) and the combined liver/kidney transplant cohort (215 ± 110 μmol/L). This difference is most likely due to a lower enzymatic activity provided by the kidney transplant; lower kidney function after isolated kidney transplantation might also contribute to higher plasma MMA levels. Metabolic impairment associated with high MMA plasma levels has been repeatedly reported [32–34]; these data should

Table 3
Number and causes of deaths in the three transplant cohorts.

Transplant type and number of deaths, n (%)	Cause of death	Age at transplantation (years)	Time from transplantation to death (years)
Kidney transplantation, 5/26 (19%)	Metabolic decompensation	14.6	0.9
	Infection	6.6	9.4
	Cardiomyopathy	15.9	1.9
	Pancreatitis	10.8	1
	Hepatoblastoma	9.5	1.9
Liver transplantation 3/24 (13%)	Metabolic decompensation	1	12.2
	Infection	16.8	4.4
	Intra-operative cardiac arrest	1.3	0
Combined liver-kidney transplantation 2/33 (6%)	Metabolic decompensation	9.1	11.4
	Liver failure/rejection	16.1	7.2

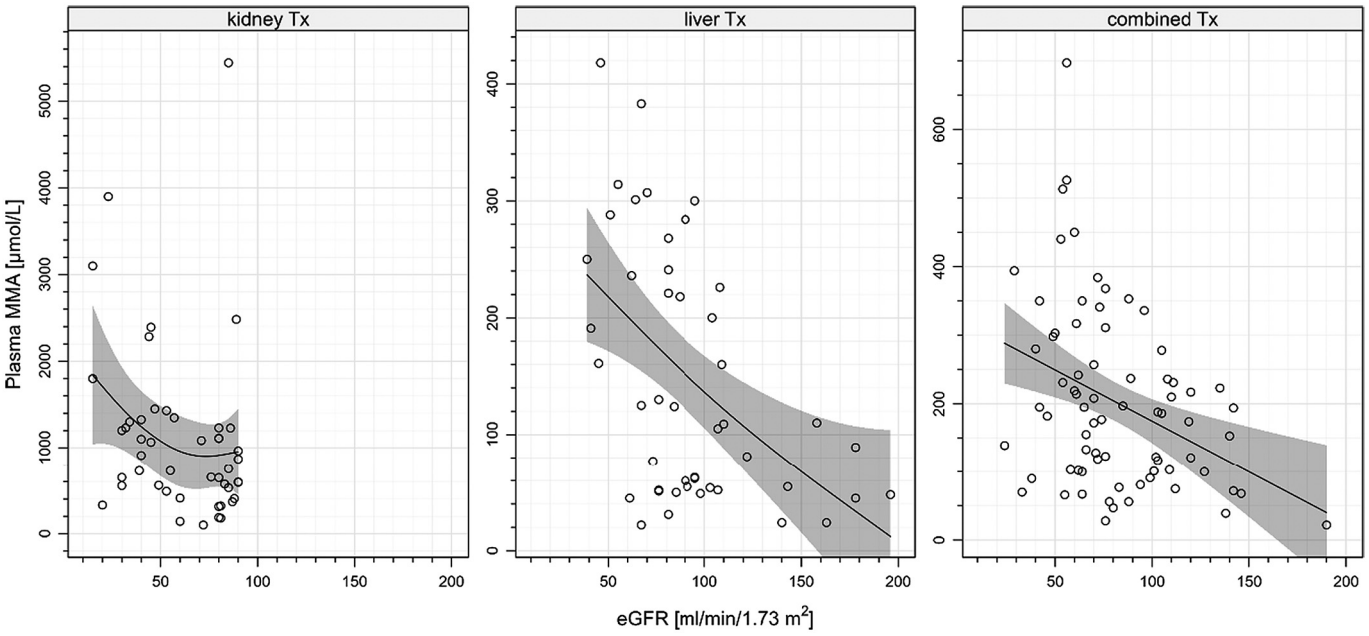

Fig. 4. Plasma MMA concentration as a function of estimated glomerular filtration rate (eGFR) in in patients after kidney (n = 59), liver ($n = 63$), or combined liver-kidney transplatation ($n = 100$). Data are pooled from observations at month 1, 6 and 12 posttransplant in patients in whom both plasma MMA concentrations and eGFR data were available. Please not the different scales of the y-axes. There was a significant negative correlation ($P < 0.0001$) between eGFR and plasma MMA concentration in all three patient cohorts. Solid lines show estimates from scatter-plot smoother, and gray areas are corresponding 95% confidence bands.

be considered for the definition of a transplant strategy in MMA patients.

During the first year posttransplant, kidney function may be impaired by several factors such as rejection, urinary tract infections, drug toxicity, and viral infection, beside others [35]. It is likely that impaired allograft function leads to reduced renal filtration of MMA and lower intrarenal enzymatic activity, both of which increase plasma MMA levels driving further renal tissue injury and decline of kidney function. In fact, mean eGFR of MMAemia patients after isolated kidney transplantation was by 9.1 mL/min/1.73 m^2 lower than the eGFR of 1112 patients aged <18 years documented in the CERTAIN Registry, who had received a kidney transplant for primary kidney diseases other than MMAemia. Moreover, eGFR in the MMAemia population may overestimate kidney function, because patients have less muscle mass than children with other primary kidney diseases. Hence, persistently high plasma MMA concentrations after isolated kidney transplantation appear to contribute to tissue injury of the transplanted kidney.

Our observations are consistent with a recent retrospective study on a smaller French cohort of 23 patients with MMAemia receiving either isolated kidney transplantation, liver transplantation, or combined liver-kidney transplantation [15]. Also, in this study, combined liver-kidney transplantation was associated with a better metabolic outcome than isolated kidney transplantation.

We observed that the transplant strategy for MMAemia patients differs between the US and Europe. While in the US isolated kidney transplantation was only exceptionally performed, 25 of 48 (52.1%) of transplantations performed in Europe were isolated kidney transplantations. The reasons for this difference are not evident. In the absence of clear indications from existing guidelines on treatment of MMAemia [2,36], the transplantation strategy in MMA patients is a personal decision of the caring physicians and transplant team at each center. The current guidelines suggest that liver and/or kidney transplantation should be considered as an alternative therapy to conventional medical treatment [2]. They do not exclude any options, including isolated kidney transplantation. This is relevant, because also liver transplantation does not cure the disease but attenuates the clinical phenotype [12–15], and severe neurological complications may occur even after liver transplantation because of persistent increased production of di- and tricarboxylic toxic metabolites in the brain [15,24,25,27,36].

The main strengths of this study are the large sample size and sufficient numbers in the three transplant cohorts to allow a meaningful comparison. The main limitation is the variable observation period, missing data at the different observation time points, no data on long-term outcome, lacking data on extrarenal disease manifestations of MMAemia and the type of conservative therapy, and differences among age at transplantation and laboratory methods for measurements of plasma MMA. There is also lack of other measures of kidney function such as cystatin C. Also, no data on immunosuppressive therapy in the three transplant groups are available. Patients after liver transplantation may need less immunosuppressive therapy with calcineurin inhibitors, which are nephrotoxic. We cannot exclude that some of the differences in eGFR among transplant groups are due to differences in nephrotoxic medication. No data on dietary management of MMAemia in the three transplant groups are available. Older patients with MMAemia need less dietary protein. We cannot exclude that higher plasma MMA levels in patients after kidney transplantation, who were older than patients after liver transplantation, are partially due to a more liberal diet which will lead to higher plasma MMA levels posttransplant. Finally, no details surrounding the causes of death are available. The source of metabolic instability in MMAemia is the liver; hence, seeing fatal decompensation after liver transplantation or combined liver-kidney transplantation is unusual. No data are available whether these decompensations have been preceded by liver graft dysfunction.

In conclusion, our data show that liver transplantation and combined liver-kidney transplantation lead to markedly lower plasma MMA levels during the first 2 years posttransplant than kidney transplantation alone and are associated with a better preservation of kidney function. We suggest that liver transplantation should therefore be part of the transplant strategy in MMA patients. In the future, new molecular therapeutic approaches in MMAemia could make organ transplantation unnecessary. There are promising trials in animal models for systemic mRNA therapy [37] and hepatotropic gene replacement therapy [38], both of which should be considered analogous to liver transplantation in terms of effect strength, but may have a more favorable side effect

profile. It will also be interesting to investigate whether a liver targeted mRNA or gene therapy or hepatic cell therapy is useful in patients with MMAemia who have already received a kidney transplant.

Data availability statements

The data underlying this article will be shared on reasonable request to the corresponding author.

Data availability

Data will be made available on request.

Declaration of Competing Interest

The authors declare that they have nothing to disclose and that there is no conflict of interest.

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