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Julien Roussel, PhD,[*] François Labarthe, MD, PhD,[†] Jerome Thireau, PhD,[*] Fabio Ferro, PhD,[*†] Charlotte Farah, PhD,[*] Jerome Roy, MSc,[*] Masahisa Horiuchi, MD, PhD,[‡] Martine Tardieu, MD,[†] Bruno Lefort, MD,[†] Jean François Benoist, PhD,[§] Alain Lacampagne, PhD,[*] Sylvain Richard, PhD,[*] Jeremy Fauconnier, PhD,[*] Dominique Babuty, MD, PhD,[‖] Jean Yves Le Guennec, PhD[*]

From the [*]INSERM U1046, CNRS UMR 9214, Université de Montpellier, Montpellier, France, [†]Médecine Pédiatrique, INSERM U1069, CHRU de Tours, Université François Rabelais, Tours, France, [‡]Department of Biochemistry, Faculty of Medicine, Kagoshima University, Kagoshima, Japan, [§]Laboratoire de Biochimie Hormonologie, Hôpital R Debré, AP-HP, Paris, France, and [‖]Service de Cardiologie, CHRU de Tours, Université François Rabelais, Tours, France.

BACKGROUND Short QT syndrome is associated with an increased risk of cardiac arrhythmias and unexpected sudden death. Until now, only mutations in genes encoding the cardiac potassium and calcium channels have been implicated in early T-wave repolarization.

OBJECTIVE The purpose of this study was to confirm a relationship between a short QT syndrome and carnitine deficiency.

METHODS We report 3 patients affected by primary systemic carnitine deficiency and an associated short QT syndrome. Ventricular fibrillation during early adulthood was the initial symptom in 1 case. To confirm the relationship between carnitine, short QT syndrome, and arrhythmias, we used a mouse model of carnitine deficiency induced by long-term subcutaneous perfusion of MET88.

RESULTS MET88-treated mice developed cardiac hypertrophy associated with a remodeling of the mitochondrial network. The continuous monitoring of electrocardiograms confirmed a shortening of the QT interval, which was negatively correlated with the plasma carnitine concentration. As in humans, such alterations coincided with the genesis of ventricular premature beats and ventricular tachycardia and fibrillation.

CONCLUSION Altogether, these results suggest that long-chain fatty acid metabolism influence the morphology and the electrical function of the heart.

KEYWORDS Carnitine deficiency; Short QT syndrome; Electrophysiological remodeling; Sudden death; Ventricular arrhythmias

ABBREVIATIONS **ECG** = electrocardiogram/electrocardiographic; $\mathbf{I_{Kr}}$ = rapid potassium current; **LCFA** = long-chain fatty acid; **LV** = left ventricular; **OCTN2** = organic cation transport Na^+; **PCD** = primary carnitine deficiency

Introduction

Sudden cardiac arrest is one of the most common causes of early unexpected death, with an estimated incidence of 300,000 cases per year in the United States.[1] Genetic cardiac channelopathies have recently been described as an important cause of these sudden cardiac deaths. Accordingly, short QT syndrome is characterized by a reduced QT interval, which is associated with an increased risk of cardiac arrhythmias and sudden death at a young age.[2] To date, 3 mutations in genes encoding for cardiac potassium channels[2] and 3 affecting the calcium channels[3] have already been implicated in few patients with this syndrome. These mutations induce a gain of function in the outward potassium current or a loss of function in the inward calcium current, reducing the repolarization process and inducing a shortening of the QT interval.

Long-chain fatty acids (LCFAs) need to complex with carnitine and form acylcarnitines to diffuse through the mitochondrial membrane. Organic cation transport Na^+ (OCTN2), an ubiquitary membrane carnitine transporter, concentrates carnitine inside the cell.[4] Primary carnitine deficiency (PCD; MIM#212140) is a hereditary defect of fatty acid oxidation due to defective transport of carnitine. Most affected patients present during infancy with progressive heart failure, generalized muscle weakness, and recurrent episodes of fasting-induced hypoketotic hypoglycemia.[5,6] At the cellular level, this pathology is characterized by an LCFA accumulation and an increase in the number of mitochondria.[7,8]

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history, investigations, diagnosis, and outcome of the child and family. It was conducted between 2007 and 2012.

Results. Among the 19 newborns studied, all had severe hypotonia. Prenatal and perinatal features were similar. Their outcome was generally severe: the median survival as measured by the Kaplan-Meier method was 6.9 months. Thirteen children died at a median age of 61 days; ten of them were treated with a palliative procedure. Five children had achieved respiratory independence but suffered from a small delay in motor development. Among the three children who continuously required ventilatory support, only one survived (follow-up period: 23 months); he was the only one undergoing tracheostomy in the cohort. Diagnostic processes were different, leading to pathological and genetic diagnosis for only six infants. There was only histological orientation for seven and no specific diagnostic orientation for the last six. These difficulties have led us to propose an exploration process based on the literature.

Conclusion. This study highlights difficulties in obtaining a diagnosis and a precise prognosis for floppy ventilated infants. An exploration-standardized process for infants suspected of congenital neuromuscular diseases was made in order to standardize procedures. It could be used as a tool for all professionals involved.

Résultats. Dix-neuf nouveau-nés ont été inclus. L'anamnèse anténatale et néonatale était similaire. L'évolution avait été sévère : 13 enfants étaient décédés (âge médian : 61 jours), 10 dans une démarche palliative. Un enfant avait bénéficié d'une ventilation prolongée par trachéotomie. Cinq enfants avaient pu être sevrés de leur assistance ventilatoire ; leur développement moteur était subnormal. Les explorations diagnostiques, hétérogènes, avaient abouti à une identification anatomopathologique et génétique chez 6 patients seulement alors que pour un tiers, aucune orientation diagnostique n'avait été retenue. Ces difficultés diagnostiques ont motivé l'élaboration d'une stratégie d'exploration, à partir de l'analyse de la littérature.

Conclusions. Les données de notre cohorte, comparables aux séries publiées, illustrent la nécessité d'harmoniser les pratiques, notamment la démarche d'investigation des nouveau-nés suspects de maladie neuromusculaire congénitale. La stratégie proposée pourrait constituer un outil de travail auprès des professionnels concernés.

1. Introduction

Face à une hypotonie néonatale, les équipes soignantes doivent relever plusieurs défis : approcher au plus près le diagnostic étiologique et le pronostic tout en soutenant l'autonomie respiratoire et nutritionnelle de l'enfant, se heurtant parfois à des dilemmes éthiques. Si les causes centrales sont les plus fréquentes et bien connues (l'asphyxie périnatale au premier rang), les maladies neuromusculaires congénitales précoces restent confidentielles d'après les séries publiées [1–5], notamment lorsque l'on a écarté les diagnostics d'amyotrophie spinale infantile (SMA) et de dystrophie myotonique de type 1 (DM1). Pour ces deux maladies, en effet, la certitude diagnostique repose sur des techniques rapides de biologie moléculaire et des recommandations professionnelles internationales de prise en charge sont soutenues par des connaissances et expériences médicales solides [6,7]. Au contraire, les autres maladies en cause sont encore imparfaitement connues. Les diagnostics les plus fréquents, d'après l'analyse des séries publiées, sont rapportés dans le *tableau I*. Il est difficile d'en établir une certitude diagnostique car les arguments histologiques sont complexes et la documentation génétique est encore lacunaire. Les séries publiées sont rares et de faible effectif ; le pronostic est mal connu. Notre objectif est donc de proposer une stratégie d'exploration des nouveau-nés hypotoniques et ventilés, suspects d'être atteints d'une maladie neuromusculaire congénitale précoce, à partir des données de la littérature et de l'expérience du centre de référence des maladies neuromusculaires (CRMN) du Grand Sud-Ouest.

2. Matériel et méthodes

Nous avons réalisé une étude rétrospective, multicentrique, auprès des unités de réanimation et soins intensifs néonatals des centres hospitaliers universitaires (CHU) de Bordeaux, Montpellier et Toulouse, répertoriant, entre 2007 et 2012, les cas d'enfants ayant nécessité un support ventilatoire dans leur premier mois de vie du fait d'une probable maladie neuromusculaire congénitale précoce. Pour cela, nous avons soumis un questionnaire à chaque neuropédiatre référent du CRMN du Grand Sud-Ouest (régions Aquitaine, Languedoc-Roussillon et Midi-Pyrénées). L'inclusion des enfants était décidée par ce correspondant d'après les critères d'inclusion et d'exclusion. Les premiers étaient une détresse respiratoire ayant requis une aide ventilatoire dans le premier mois de vie (ventilation invasive ou ventilation spontanée en pression positive continue), et des signes cliniques évocateurs d'une origine neuromusculaire de la défaillance respiratoire. Les critères d'exclusion étaient la présence d'arguments cliniques ou para-cliniques en faveur d'une affection d'origine centrale, les diagnostics moléculaires de DM1, SMA ou du syndrome Prader-Willi (PWS). Le questionnaire évaluait l'anamnèse familiale, obstétricale et périnatale, la symptomatologie de l'enfant à la naissance, la démarche diagnostique réalisée et

hypoglycemia (1.2 mM; reference range 2.8–5.5 mM) without ketonuria. He rapidly recovered with glucose infusion, and plasma glucose levels remained in the normal range while he was fed a regular diet. This boy also had a permanent slight increase in plasma creatine kinase levels (300–400 U/L; reference range 0-170 U/L) and mild hyperammonemia (90–170 μM; reference range <40 μM). The plasma carnitine level was significantly decreased (free carnitine level 3 μM; reference range 25–55 μM, and total carnitine level 4 μM; reference range 35–65 μM), with undetectable levels of acylcarnitines in blood. The measurable urinary excretion of carnitine (44 μmol/mmol creatinine) despite the low plasma carnitine concentration was suggestive of PCD.[16] The organic acid profile in urine was normal. The defect in carnitine transport was further confirmed in cultured skin fibroblasts (patient: 0.06 pmol/(min · mg); control: 2.42 pmol/(min · mg)), and genotyping identified 2 mutations in the heterozygous state in the *SLC22A5* (OCTN2; NM_003060) gene. The first modification, in exon 1, corresponded to a G>A transition at position 186 in *SLC22A5* cDNA (c.186G>A) and was inherited from his father. At the protein level, it is predicted to result in the formation of a premature termination codon at position 62 (p.Trp62Ter). This mutation has never been reported to our knowledge but its deleterious impact is strong. The second mutation, inherited from his mother, was a C>T transition in exon 8 at a position corresponding to nt1411 in the *SLC22A5* gene (c.1411C>T). The resulting amino acid substitution, the basic arginine 471 replaced by a neutral cysteine, implicates a conserved amino acid and has already been reported (p.Arg471Cys).[17] Upon admission, echocardiography revealed mild dilated cardiomyopathy (LV end-diastolic diameter 36.5 mm for a body area of 0.54 m²; *z* score 3 SD)[18] with normal ventricular function.

Patient 2

The mother of patient 1, a 28-year-old woman with an unremarkable history, was investigated 1 year before for an aborted sudden death due to ventricular fibrillation. After resuscitation, her ECG showed a shortened QT interval (corrected QT interval 340 ms; Table 1) with increased T waves despite a concomitant low plasma potassium level (2.4 mM; reference range 3.2–5.8 mM), suggesting a short QT syndrome. Shortening of the QT interval was not modified after correction of the potassium level. QT dynamicity was estimated from the 24-hour ECG recording using the ELATEC Holter analysis QT software (ELA Medical, Montrouge, France). Daytime QT dynamicity (QTend/RR slope 0.238) was greater than nighttime QT dynamicity (QTend/RR slope 0.182). The morphology of the T wave was constant during the prolonged ECG recording in both derivations. Echocardiography, coronarography, and left and right ventricular functions were all normal. Because of the high risk of recurrence of cardiac arrhythmia, a cardiac defibrillator was implanted and the patient remained free of symptoms during the following year. Because of her son's story, the evaluation of her carnitine status was performed, which immediately confirmed the PCD with a significant decrease in plasma carnitine and acylcarnitine levels (Table 1) associated with sustained urinary excretion of carnitine (21 μmol/mmol of creatinine). Urinary organic acid profile, ammonemia, and plasma creatine kinase levels were in the normal range. The evaluation of carnitine status was performed, and kalemia was normal. Laboratory genetic testing later confirmed the PCD. The patient carries the missense mutation (p.Arg471Cys) she has transmitted to her son, with on the second allele a complete deletion of exon 2 of the *SLC22A5* gene. Mutations in *KCNQ1*, *KCNH2*, and

Table 1 Plasma carnitine concentrations and QT intervals before and after carnitine therapy in patients with systemic carnitine deficiency.

Age	Carnitine dosage (mg/(kg · d))	Plasma free carnitine level (μM)	Plasma total carnitine level (μM)	Corrected QT interval (ms)	QTend interval (ms)	QTapex interval (ms)
Reference range		25–55	35–65	392 ± 27		
Patient 1						
1 y 9 m*	0	3	4	309 ± 4	257 ± 21	195 ± 13
1 y 10 m	100	15	23	393 ± 14	305 ± 13	254 ± 13
2 y	100	16	28	406 ± 20	320 ± 12	250 ± 13
2 y 3 m	150	19	30	419 ± 15	321 ± 22	261 ± 14
2 y 7 m	150	15	28	383 ± 22	308 ± 16	236 ± 12
Patient 2						
30 y 3 m*	0	1	2	340± 6	320 ± 10	270 ± 12
30 y 4 m	50	12	16	385 ± 17	410 ± 24	333 ± 20
30 y 8 m	75	15	20	408 ± 19	410 ± 25	340 ± 17
Patient 3						
5 y 1 m*	0	nd	9	282 ± 2	280 ± 15	223 ± 9
5 y 3 m	100	nd	nd	360 ± 18	328 ± 20	276 ± 22
9 y 1 m	100	13	22	408 ± 13	367 ± 22	300 ± 22
15 y 3 m	100	21	26	400 ± 12	396 ± 8	325 ± 18

Plasma total and free carnitine concentrations were determined using the electrospray ionization MS-MS method. Total carnitine included free carnitine and the sum of all acylcarnitines. The corrected QT interval was calculated as the ratio between the QT interval (in ms) and the square root of the RR′ interval (in ms). Age is expressed in years (y) and months (m).

nd = not determined.

*Values before carnitine therapy.

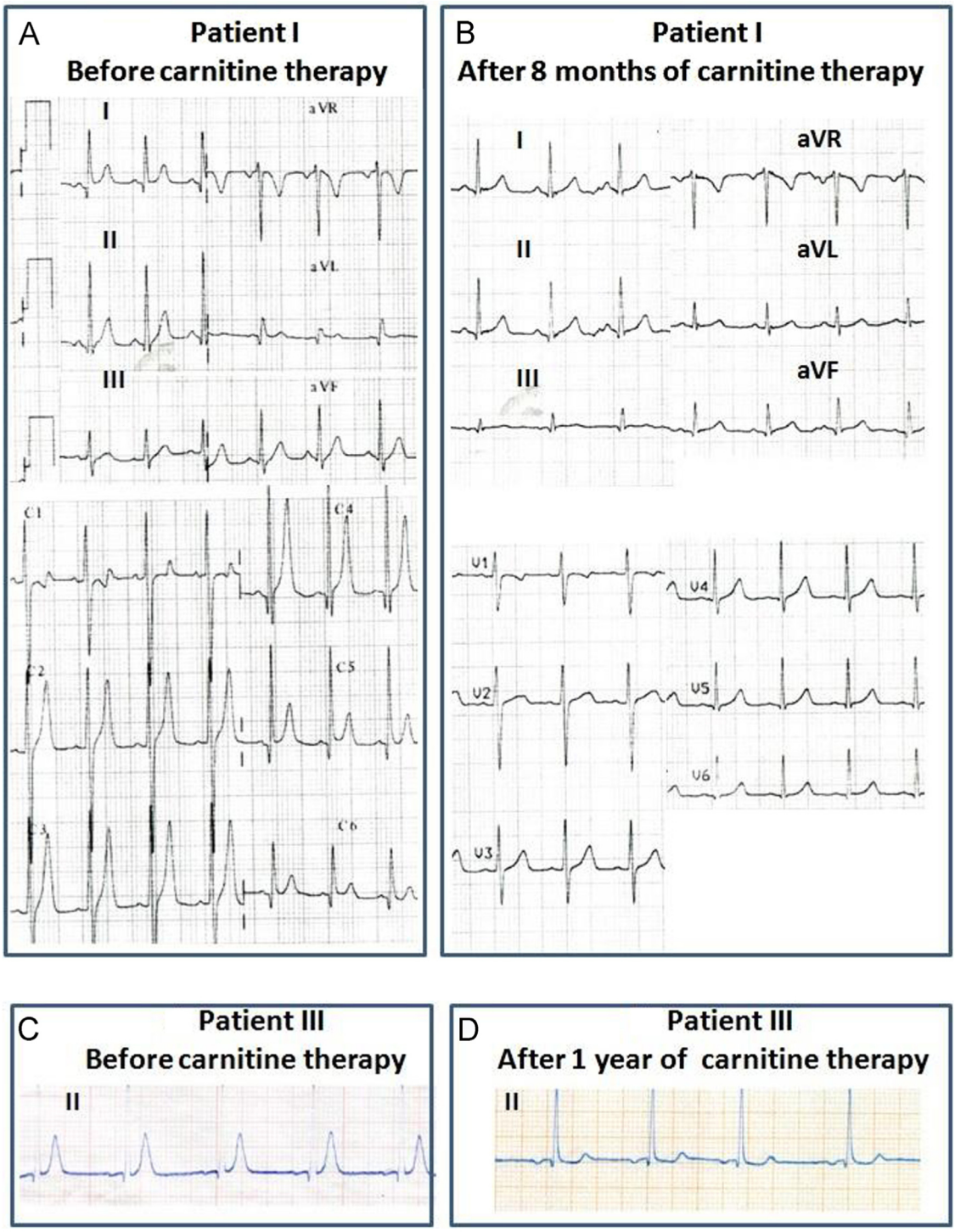

Figure 1 Electrocardiograms before and after carnitine therapy in patient 1 (A and B) and patient 3 (C and D) with primary systemic carnitine deficiency.

KCNJ2 genes encoding for potassium channel proteins involved in short QT syndrome were ruled out.

Patient 3

Patient 3 has already been reported a long time ago.[7] Briefly, this girl, second child of consanguineous Portuguese parents, had presented with recurrent episodes of fasting hypoglycemia since the age of 20 months. One year later, she progressively developed ventricular hypertrophy, associated with amyotrophy and hepatomegaly, that has evolved to severe cardiac heart failure at the age of 5 years. At that time, she had a severe carnitine deficiency in plasma (9 μM) and in muscle tissue (0.3 nmol/mg of tissue; reference range 2.8 ± 0.3 mmol/mg) associated with an inappropriate carnitine loss in urine (98 μmol/d; reference range 40 ± 8 μmol/d). The reduced carnitine uptake in cultured skin fibroblasts later confirmed the diagnosis. The patient fully recovered within a few months with oral DL-carnitine supplementation (5 g/d)

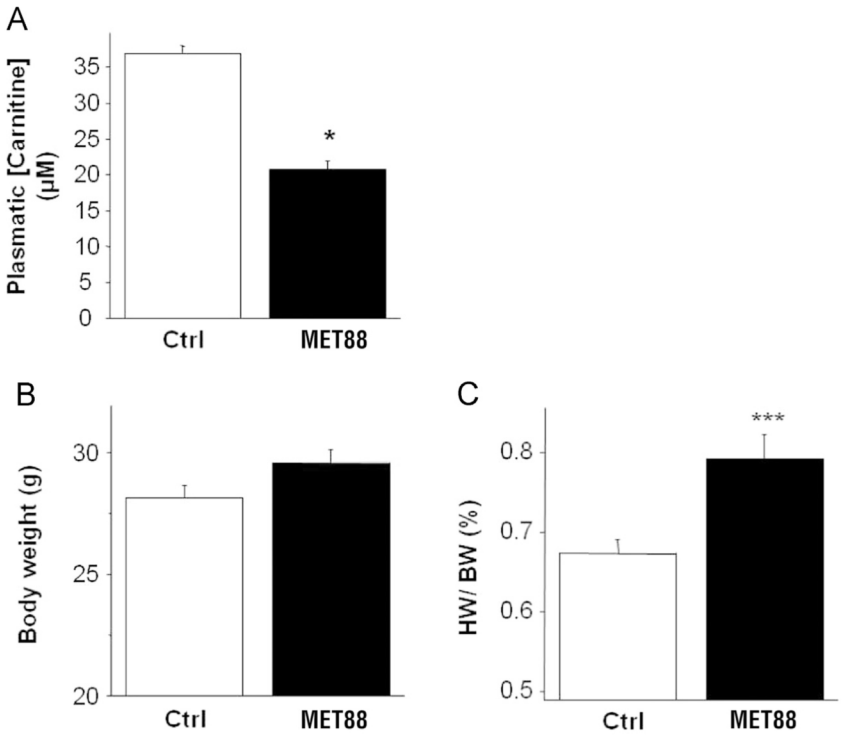

Figure 2 MET88 treatment decreases the plasma carnitine concentration and increases the heart size. **A:** Plasma carnitine concentration in control mice (Ctrl) and after 28 days of MET88 subcutaneous infusion. **B:** The body weight of mice was measured before and after the treatment. **C:** The heart weight (HW) to body weight (BW) ratio (in percentage) was calculated using the following formula: (HW/BW) × 100. $^{*}P < .05$; $^{***}P < .001$; N = 10.

and L-carnitine substitution a few years later. Still on therapy, this 40-year-old woman is actually free of any cardiac, muscular, or liver symptoms.

Statistical analysis

Data are presented as mean ± standard error of the mean. Statistical significance was defined as $^{*}P < .05$, $^{**}P < .01$, and $^{***}P < .001$ using the Student *t* test (paired or unpaired, as appropriate). N represents the animal number, and n represents the number of cells studied. The correlation between the QT interval and the plasma carnitine concentration was assessed using the Spearman rank correlation test.

Table 2 Effects of long-term treatment with MET88 on mice heart morphology and function.

Parameters	Baseline	After treatment
HR (beats/min)	452 ± 19	460 ± 12
M-mode parameters		
AWTd (mm)	0.72 ± 0.01	0.90 ± 0.03*
AWTs (mm)	1.15 ± 0.02	1.34 ± 0.05*
LVIDd (mm)	3.48 ± 0.14	3.39 ± 0.08
LVIDs (mm)	2.30 ± 0.13	2.29 ± 0.11
PWTd (mm)	0.72 ± 0.03	0.86 ± 0.34*
PWTs (mm)	1.16 ± 0.04	1.31 ± 0.05*
EF (%)	64 ± 3	62 ± 3
FS (%)	34 ± 2	33 ± 2
B-mode parameters		
FAC (%)	59 ± 3	59 ± 4
EF (%)	63 ± 4	61 ± 4

Data are expressed as mean ± standard error of the mean.

AWTd = anterior wall thickness in diastole; AWTs = anterior wall thickness in systole; EF = ejection fraction; FAC = fractional area change; FS = fractional shortening; HR = heart rate; LVIDd = left ventricular internal diameter in diastole; LVIDs = left ventricular internal diameter in systole; PWTd = posterior wall thickness in diastole; PWTs = posterior wall thickness in systole.

*$P < 0.05$ baseline vs after treatment. N= 10, and each animal was its own control.

Results

Systemic carnitine deficiency in patients

Herein, we report 3 patients with PCD from 2 unrelated families. The diagnosis was first based on the absence of plasma carnitine associated with sustained urinary excretion of carnitine in the absence of any organic acidurias and renal tubulopathy. This hypothesis was further confirmed by reduced carnitine uptake in cultured skin fibroblasts and/or genotyping. In patient 1, the ECG showed a shortened QT interval (corrected QT interval 300 ms; reference range 392 ± 27 ms; Table 1) associated with an abnormally tall, symmetrical peaking T wave (Figure 1A), which was consistent with a hereditary short QT syndrome. QT dynamicity was analyzed from a 24-hour ECG recording by using the same software used in his mother. Daytime QT dynamicity was low with a gentle slope (QTend/RR slope 0.0288), whereas nighttime QT dynamicity seemed preserved (QTend/RR slope 0.290). The morphology of the T wave was constant during the prolonged ECG recording in both derivations. Within 1 month, oral carnitine therapy (150 mg/(kg · d)) resulted in cardiac improvement while plasma carnitine and blood acylcarnitine levels increased (Table 1): the

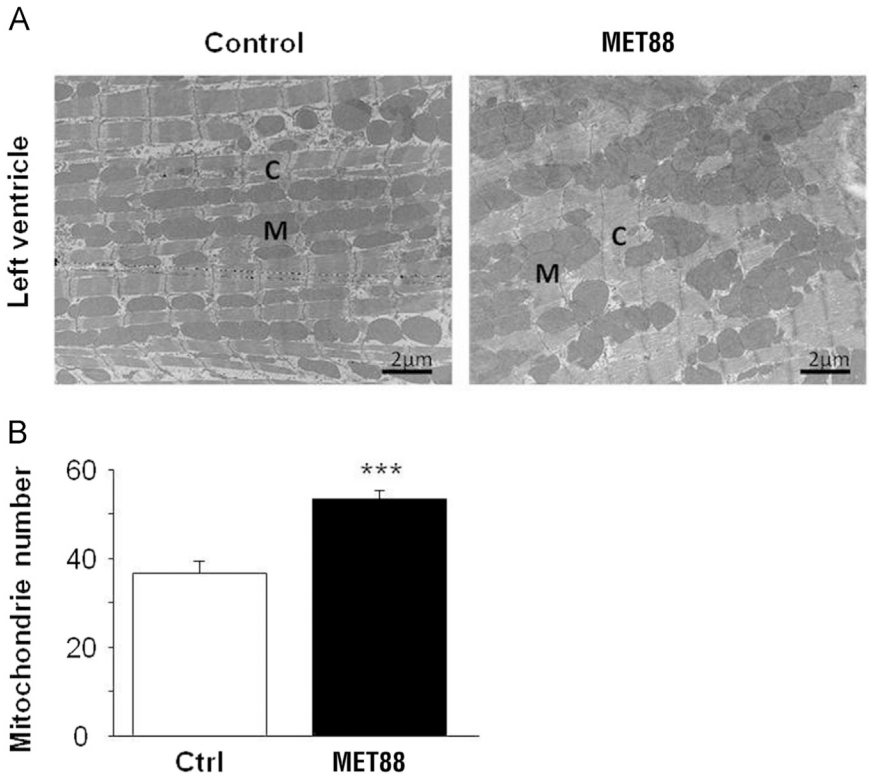

Figure 3 MET88 treatment induces a remodeling of the mitochondrial network. **A:** Typical examples of transmission electron microscopic images of ventricles. C = contractile protein; M = mitochondria. **B:** Mean number of mitochondria under control and MET88 conditions. $^{***}P < .001$; N = 3 for control and N = 4 for MET88.

LV end-diastolic diameter returned to the normal range on ultrasonography (34 mm for a body area of 0.6 m²; *z* score: 1.5 SD) and both the length of the QT interval and the T wave aspect normalized progressively on ECGs after 8 months (Table 1 and Figure 1B). Patient 2 also had a short QT interval. Under oral carnitine therapy (4 g/d), the ECG pattern normalized progressively (Table 1). The ECG data of patient 3 had already been published,[7] but not analyzed for the modification of the QT interval. The retrospective analysis of the ECG performed at the time of diagnosis and before carnitine therapy revealed a significantly shortened QT interval (corrected QT interval 280 ms; Figure 1C and Table 1) with an increased peaking T waves. These abnormalities had disappeared on the ECG after 1 year of carnitine therapy (Figure 1D and Table 1). A 24-hour ECG was not recorded.

Structural remodeling: Validation of the MET88 model

To induce carnitine deficiency in mice, we treated mice with MET88 for 28 days. Treatment with 100 mg/(kg · d) of MET88 significantly decreased the plasma carnitine concentration (Figure 2A), which was in agreement with Liepinsh data.[12] Body weight remained unchanged after carnitine deficiency (Figure 2B), but the heart weight to body weight ratio was increased, suggesting cardiac hypertrophy (Figure 2C). Increases in anterior wall thickness and posterior wall thickness observed by echocardiography confirmed major parietal hypertrophy (Table 2). This hypertrophy did not affect the LV internal diameter either in systole or in diastole. Clinical and experimental observations have indicated that a decrease in carnitine concentration is correlated with a modification of the cellular ultrastructure.[7,8] In order to validate our MET88 model, transmission electron microscopic images of cardiomyocyte ultrastructure were taken on ventricles. Carnitine deficiency induced severe modifications of the mitochondrial network (Figure 3A) with an increase in the number of mitochondria in LV cardiomyocytes (Figure 3B). Similar alterations have already been described.[7,8] We did not observe myocardial lipid inclusion after MET88 treatment. These results indicate that the decrease in plasma carnitine concentration induced by MET88 leads to structural remodeling at the ventricular level.

Electrophysiological remodeling

To investigate the consequences of carnitine deficiency on cardiac electrical activity, we recorded ECGs by telemetry on vigil unrestrained mice before and after long-term treatment with MET88. Analysis of ECGs revealed that the decrease in plasma carnitine concentration did not modify basal heart rate (Figure 4A), the time to electrical conduction from atria to ventricles (PR interval) (Figure 4B), or depolarization of ventricles (QRS complex duration) (Figure 4C). Interestingly, we found that after MET88 treatment, ventricular repolarization time (QT interval) was significantly shortened

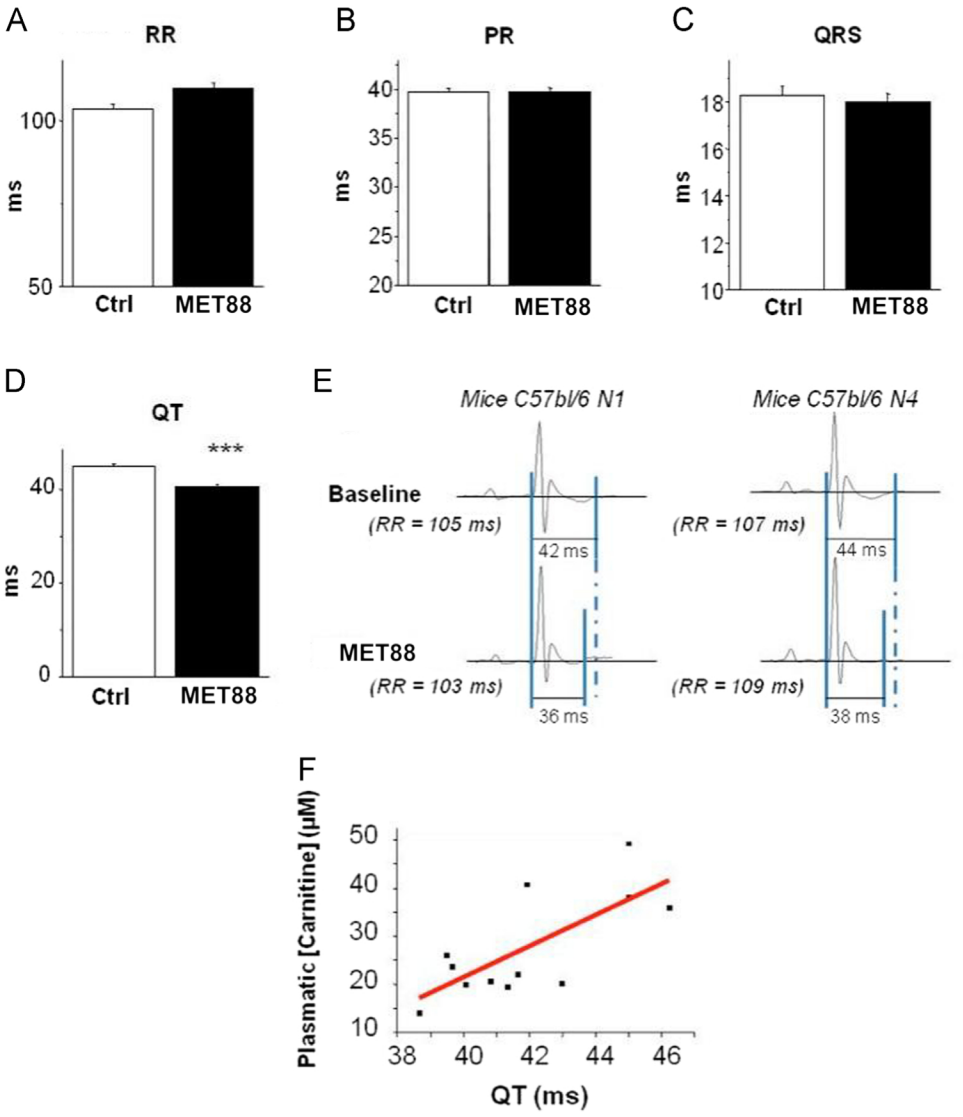

Figure 4 In vivo, plasma carnitine concentration is correlated with the QT interval duration. **A–D:** Effects of MET88 treatment on RR interval (panel A), PR interval (panel B), QRS complex duration (panel C), and QT interval duration (panel D). $^{***}P < .001$; N = 12. **E:** Typical electrocardiograms before and after MET88 treatment. **F:** Correlation between the plasma carnitine concentration and the QT interval ($r = 0.73$; $P < .01$).

(Figures 4D and 4E). Even a correlation between the QT interval and the plasma carnitine level was observed (Figure 4F). Moreover, although a short QT interval is unfavorably associated with ventricular events,[18] we highlighted that carnitine deficiency led to a high arrhythmogenic profile at the ventricular level (Figure 5). Mice developed frequent isolated and repetitive premature ventricular beats (Figures 5A–5D). Moreover, 7 of 10 mice (70%) that received MET88 developed spontaneous sustained ventricular tachycardia whereas no untreated mice developed spontaneous sustained ventricular tachycardia (Figures 5E and 5F). Half of the MET88-treated mice developed ventricular fibrillation (Figures 5G and 5H). We did not observe a significant difference for conduction-dependent events such as atrioventricular block and bundle branch block or for automaticity troubles such as sinus arrest between the 2 conditions, which was in accordance with the absence of cellular ultrastructural alteration in the atrium (not shown). In summary, our experimental model reproduces the clinical and electrocardiographic characteristics of the patients suffering from carnitine deficiency.

Discussion

Herein, we present a new pathophysiological condition promoting short QT syndrome. The 3 patients reported herein had systemic carnitine deficiency. The diagnosis was based on the almost nil plasma carnitine levels associated with sustained urinary excretion of carnitine in the absence of any organic acidurias and renal tubulopathy. This hypothesis was further confirmed by reduced carnitine uptake in cultured skin fibroblasts (patients 1 and 3) and genotyping (patients 1 and 2). This autosomal recessive disorder is due to a defective transport of carnitine into the

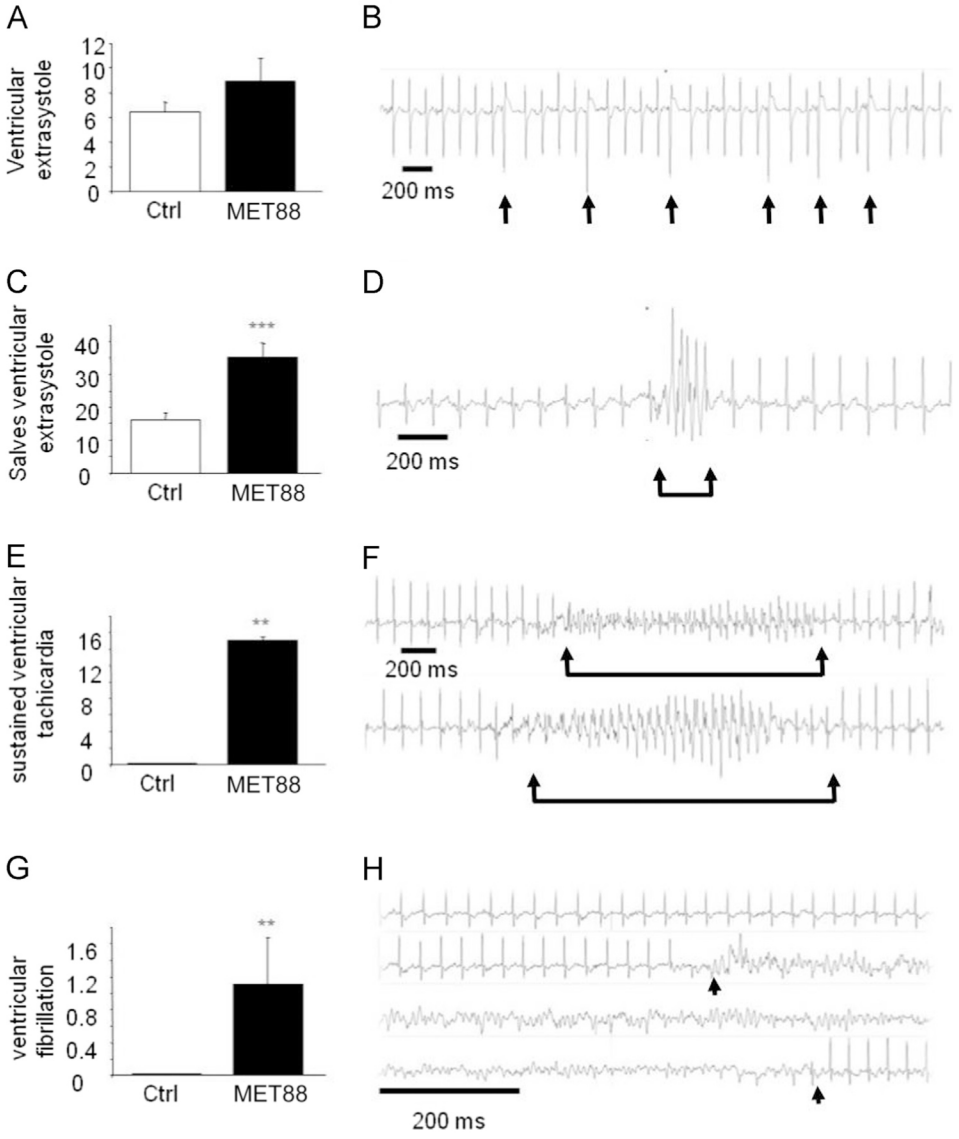

Figure 5 Arrhythmic events recorded after 28 days of MET88 treatment. Different kinds of arrhythmias were recorded (indicated by arrows) under basal conditions (blank column) and after MET88 treatment (filled column). **A and B:** Single premature ventricular beats. **C and D:** Salvos of premature ventricular beats. **E and F:** Episode of sustained ventricular tachycardia. **G and H:** Ventricular fibrillation events were counted. $^{**}P < .005$; $^{***}P < .001$; $N = 10$.

cell.[5,6] Carnitine is the essential cofactor for the mitochondrial carnitine shuttle that transfers LCFAs as acylcarnitine esters across the inner mitochondrial membrane. Intracellular carnitine deficiency induces an impairment of LCFA oxidation leading to broad clinical disorders. Most patients with carnitine transporter deficiency become sick during the initial years of life with recurrent hypoketotic hypoglycemia and/or cardiomyopathy, myopathy, hyperammonemia, and liver disease.[5,6] Some patients may present such clinical characteristics much later in childhood with isolated cardiomyopathy, and few asymptomatic adults have also been reported.[19,20] Biologically, the disorder is characterized by low levels of plasma carnitine contrasting with sustained urinary excretion of carnitine, abnormally low levels of acylcarnitines in blood, and the absence of dicarboxylic aciduria.[6] Cardiac arrhythmias due to defective mitochondrial fatty acid oxidation have been described in neonates affected by some carnitine shuttle defects, except for PCD.[9] The presentation of patient 2 with ventricular fibrillation as an inaugural symptom during adulthood and without overt cardiomyopathy was unusual. QT dynamicity was preserved in this condition, and daytime QT dynamicity reached the level (> 0.18) reported by Chevalier et al[21] as predictive of sudden death in patients with myocardial infarction. A similar presentation has previously been reported in a 15-year-old female patient who fully recovered with L-carnitine supplementation.[10] Unfortunately, this patient's ECG, and more particularly her QT interval, was not documented. QT dynamicity in patient 1 is difficult to analyze and interpret, because no data on QT dynamicity are available in young children.

PCD is classically due to consanguinity between the parents, with an autosomal recessive inheritance for PCD. However, it seems to be different in the first family where both mother and son were affected. This genetic inheritance

is unusual because the mutation inherited from the father was different from the 2 mutations inherited from the mother, suggesting a coincidental occurrence. The frequency of PCD has not been defined in the United States and Europe, but it remains rare and can be estimated at 1:50,000 from newborn screening data and reported cases.[22]

In mice, we observed that a decrease in carnitine concentration induced by MET88 dosage is responsible for the shortening of the QT interval and severe ventricular arrhythmias as well as of structural remodeling at the ventricular level.

The mouse model of carnitine deficiency reproduces many features—electrophysiological and structural—of the abnormalities observed in patients affected by PCD. These clinical results are obtained after a short period of MET88 treatment of the mice associated with a decrease of about 50% in the carnitine level. In PCD, the level of plasma carnitine is much lower. However, the level of plasma carnitine does not reflect the level of carnitine in different organs; for example, the level of plasma carnitine is normal in carnitine deficiency in muscle tissue, whereas the concentration of carnitine is low in muscle tissue. These differences between the level of plasma carnitine and the concentration of carnitine in different organs are explained by the differences in kinetic parameters of the transport system in various tissues (fibroblasts, muscle, liver, and heart).[23] The risk of developing ventricular fibrillation and sudden cardiac death is associated with the shortening of the QT interval.[24] In this study, we describe a close relationship between the carnitine concentration and the QT interval. But until now, the mechanisms connecting the carnitine concentration to the QT interval have remained elusive. Different studies have shown a direct influence of the high LCFA concentration on different ion-channel activities (I_{Na}, I_{Ca}, and I).[25–27] We previously showed that the absence of LCAC increases the rapid potassium current (I_{Kr}),[28] which plausibly participates in the shortening of the QT interval. This work[28] suggests that LCFAs can directly regulate I_{Kr}. Class III antiarrhythmic agents have not been tested in mice, but these drugs should be tested to assess the role of potassium currents, especially I_{Kr}, in our experimental model of short QT syndrome.

Conclusion

Our results show that carnitine deficiency must be suspected in situations of short QT syndrome or unexplained cardiac arrhythmias, which could be responsible for sudden death even during adulthood. In such situations, PCD can be easily corrected by oral carnitine supplementation, thereby normalizing the QT interval and probably preventing the recurrence of cardiac arrhythmias. Furthermore, these reports suggest that carnitine or its derivatives, especially long-chain acylcarnitines, play a pivotal role in cardiac ionic currents, possibly leading to electrophysiological dysfunction.

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References

1. Priori SG, Aliot E, Blomstrom-Lundqvist C, et al. Task Force on Sudden Cardiac Death of the European Society of Cardiology. Eur Heart J 2001;22:1374–1450.
2. Brugada R, Hong K, Cordeiro JM, Dumaine R, Short QT. syndrome. CMAJ 2005;173:1349–1354.
3. Hsiao PY, Tien HC, Lo CP, Juang JM, Wang YH, Sung RJ. Gene mutations in cardiac arrhythmias: a review of recent evidence in ion channelopathies. Appl Clin Genet 2013;6:1–13.
4. Ohashi R, Tamai I, Nezu JiJ, Nikaido H, Hashimoto N, Oku A, Sai Y, Shimane M, Tsuji A. Molecular and physiological evidence for multifunctionality of carnitine/organic cation transporter OCTN2. Mol Pharmacol 2001;59:358–366.
5. Stanley CA. Carnitine deficiency disorders in children. Ann N Y Acad Sci 2004;1033:42–51.
6. Tein I. Role of carnitine and fatty acid oxidation and its defects in infantile epilepsy. J Child Neurol 2002;17:3S57–3S82.
7. Morand P, Despert F, Carrier HN, Saudubray BM, Fardeau M, Romieux B, Fauchier C, Combe P. Lipidic myopathy with severe cardiomyopathy caused by a generalized carnitine deficiency: favorable course during carnitine hydrochloride treatment. Arch Mal Coeur Vaiss 1979;72:536–544.
8. Kuwajima M, Lu K, Sei M, et al. Characteristics of cardiac hypertrophy in the juvenile visceral steatosis mouse with systemic carnitine deficiency. J Mol Cell Cardiol 1998;30:773–781.
9. Bonnet D, Martin D, De Lonlay P, Villain E, Jouvet P, Rabier D, Brivet M, Saudubray JM. Arrhythmias and conduction defects as presenting symptoms of fatty acid oxidation disorders in children. Circulation 1999;100:2248–2253.
10. Rijlaarsdam RS, van Spronsen FJ, Bink-Boelkens MT, Reijngoud DJ, Wanders RJ, Niezen-Koning KE, Van der Sluijs FH, Dorland B, Beaufort-Krol GC. Ventricular fibrillation without overt cardiomyopathy as first presentation of organic cation transporter 2-deficiency in adolescence. Pacing Clin Electrophysiol 2004;27:675–676.
11. Spaniol M, Brooks H, Auer L, Zimmermann A, Solioz M, Stieger B, Krahenbuhl S. Development and characterization of an animal model of carnitine deficiency. Eur J Biochem 2001;268:1876–1887.
12. Liepinsh E, Vilskersts R, Skapare E, Svalbe B, Kuka J, Cirule H, Pugovics O, Kalvinsh I, Dambrova M. Mildronate decreases carnitine availability and upregulates glucose uptake and related gene expression in the mouse heart. Life Sci 2008;83:613–619.
13. Horiuchi M, Yoshida H, Kobayashi K, Kuriwaki K, Yoshimine K, Tomomura M, Koizumi T, Nikaido H, Hayakawa J, Kuwajima M, Saheki T. Cardiac hypertrophy in juvenile visceral steatosis (jvs) mice with systemic carnitine deficiency. FEBS Lett 1993;326:267–271.
14. Thireau J, Karam S, Roberge S, Roussel J, Aimond F, Cassan C, Gac A, Babuty D, Le Guennec JY, Lacampagne A, Fauconnier J, Richard S. B-adrenergic blockade combined with subcutaneous B-type natriuretic peptide: a promising approach to reduce ventricular arrhythmia in heart failure. Heart 2014;100:833–841.
15. Primassin S, Spiekerkoetter U. ESI-MS/MS measurement of free carnitine and its precursor γ-butyrobetaine in plasma and dried blood spots from patients with organic acidurias and fatty acid oxidation disorders. Mol Genet Metab 2010;101:141–145.
16. Lee NC, Tang NL, Chien YH, Chen CA, Lin SJ, Chiu PC, Huang AC, Hwu WL. Diagnoses of newborns and mothers with carnitine uptake defects through newborn screening. Mol Genet Metab 2010;100:46–50.
17. Kouchoukos NT, Blackstone EH, Doty DB, Hanley FL, Karp RB. Anatomy, dimensions, terminology. In: Kirklin JW, Barrat-Boyes BG, eds. Cardiac Surgery, vol 1. London: Churchill Livingston; 2003:3–65.

18. Patel C, Yan GX, Antzelevitch C, Short QT. syndrome: from bench to bedside. Circ Arrhythm Electrophysiol 2010;3:401–408.
19. Vijay S, Patterson A, Olpin S, Henderson MJ, Clark S, Day C, Savill G, Walter JH. Carnitine transporter defect: diagnosis in asymptomatic adult women following analysis of acylcarnitines in their newborn infants. J Inherit Metab Dis 2006;29:627–630.
20. Spiekerkoetter U, Huener G, Baykal T, Demirkol M, Duran M, Wanders R, Nezu J, Mayatepek E. Silent and symptomatic primary carnitine deficiency within the same family due to identical mutations in the organic cation/carnitine transporter OCTN2. J Inherit Metab Dis 2003;26:613–615.
21. Chevalier Ph, Burri H, Adeleine P, Kirkorian G, Lopez M, Leizorovicz A, André-Fouët X, Chapon Ph, Rubel P, Touboul P. QT dynamicity and sudden death after myocardial infarction: results of a long-term follow-up study. J Cardiovasc Electrophysiol 2003;14:227–233.
22. El-Hattab AW. Systemic primary carnitine deficiency. In: Pagon RA, Adam MP, Ardinger HH, Wallace SE, Amemiya A, Bean LJL, Bird TD, Dolan CR, Fong CT, Smith RJH, Stephens K, eds. GeneReviews®. Seattle, WA: University of Washington; 2012:2014. Updated.
23. Angelini C, Martinuzzi A, Vergani L. Treatment with L-carnitine of the infantile and adult form of primary carnitine deficiency. In: Ferrari R, Dimauro S, Sherwood G, eds. London: Academic Press; 1992:139–153.
24. Gaita F, Giustetto C, Bianchi F, Wolpert C, Schimpf R, Riccardi R, Grossi S, Richiardi E, Borggrefe M, Short QT. syndrome: a familial cause of sudden death. Circulation 2003;108:965–970.
25. Brugada J, Brugada R, Brugada P. Determinants of sudden cardiac death in individuals with the electrocardiographic pattern of Brugada syndrome and no previous cardiac arrest. Circulation 2003;108:3092–3096.
26. Bellocq C, van Ginneken AC, Bezzina CR, Alders M, Escande D, Mannens MM, Baro I, Wilde AA. Mutation in the KCNQ1 gene leading to the short QT syndrome. Circulation 2004;109:2394–2397.
27. Antzelevitch C, Pollevick GD, Cordeiro JM, et al. Loss-of-function mutations in the cardiac calcium channel underlies a new clinical entity characterized by ST-segment elevation, short QT intervals, and sudden cardiac death. Circulation 2007;115:442–449.
28. Ferro F, Ouille A, Tran TA, Fontanaud P, Bois P, Babuty D, Labarthe F, Le Guennec JY. Long-chain acylcarnitines regulate the hERG channel. PLoS One 2012;7:e41686.

CLINICAL PERSPECTIVES

Sudden cardiac death remains a common cause of death in young people. Genetic cardiac channelopathies are an important cause of sudden death in the absence of cardiomyopathy. Severe ventricular arrhythmias have also been observed in primary metabolic disorders associated or not with left ventricular hypertrophy or dilation (eg, primary carnitine deficiency and fatty acid oxidation defects). We reported 3 cases (2 children and 1 adult) of primary carnitine deficiency associated with short QT interval, one of whom also displayed ventricular fibrillation. The short QT interval was corrected by carnitine therapy, and no recurrence of ventricular fibrillation was observed. Clinical syndrome was reproduced by an experimental model of carnitine deficiency induced by MET88 infusion in mice. The carnitine level decreased, along with a shortening of the QT interval, and severe ventricular arrhythmias (ventricular tachycardia and ventricular fibrillation) were recorded in more than half of the mice treated with MET88. Severe modifications of the mitochondrial network were simultaneously observed in the left ventricle. These clinical and experimental results suggested that some alterations of ventricular repolarization may be induced by metabolic disorders, such as carnitine deficiency, without any mutation in genes encoding the channel proteins involved in ventricular repolarization. Genetic metabolic disorders appear to be a new field of investigation for physicians facing unexplained ventricular fibrillation. The measurement of the plasma carnitine level should be proposed in this particular clinical situation. The major interest of this investigation is the possibility of correcting the metabolic disorder by oral supplementation of the metabolite.