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A multicenter, randomized, double-blind, placebo-controlled trial of long-term ascorbic acid treatment in Charcot-Marie-Tooth disease type 1A (CMT-TRI*AA*L): The study protocol [EudraCT no.: 2006-000032-27]

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Abstract

There is no treatment for Charcot-Marie-Tooth disease 1A (CMT1A), but ascorbic acid (AA) is efficacious in the transgenic mouse model. Thus, a clinical trial of AA in CMT1A is warranted. The CMT-TRI*AA*L is a phase III randomized, double-blind, placebo-controlled study involving 222 CMT1A adults from eight Italian centers. Eligible for the study are symptomatic adults with genetically confirmed CMT1A. Treatment consists of 2-year oral AA (1500 mg/day) or placebo. The primary trial endpoint is an improvement in CMT Neuropathy Score. Secondary efficacy endpoints are changes in distal arm and leg maximum voluntary isometric contraction; 10 m timed walking; 9-hole-peg test; overall neuropathy limitations scale; pain and fatigue visual analog scales; health-related quality of life (SF-36); and electrophysiology. Clinical–electrophysiological assessments are performed at baseline and every 6 months thereafter. In consenting patients from three centers, skin biopsy is performed to evaluate PMP22 expression. The study will last 34 months, starting from March 2006.

Keywords: Charcot-Marie-Tooth disease; Ascorbic acid; Randomized controlled trial; Neuropathy; Genetic disorders; PMP22

1. Introduction

Charcot-Marie-Tooth disease (CMT) is the most common inherited neuromuscular disorder (estimated prevalence: 17–41:100,000) [1]. It is genetically heterogeneous, but almost one half of the patients carry a duplication on chromosome 17p11.2-p12 (CMT1A) [2–6]. Therefore, CMT1A is the most frequent monogenic CMT subtype (maximal estimated prevalence 1:5000). The 17p11.2-p12 genomic region encompasses the gene coding for the peripheral myelin protein 22 (PMP22), which is located in the compact myelin of peripheral nerves and plays a crucial role in the formation and maintenance of myelin, and possibly in cell growth regulation and shaping [5,7]. A gene-dosage effect is the hypothesized pathophysiologic mechanism underlying CMT1A: affected patients carry three copies of the PMP22 gene and overexpress the protein in peripheral nerves [5].

To date, only symptomatic or palliative treatments are available for CMT1A patients, such as physiotherapy and surgery

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for skeletal deformities and tendon tightening. Animal models of the human disease, namely transgenic rats [8] and mice [9] overexpressing PMP22, have been recently developed and are being employed for studying disease mechanisms and for testing potential treatments. As an example, progesterone and its derivatives increase PMP22 expression [10]. Accordingly, Sereda et al. [11] demonstrated that transgenic rats overexpressing PMP22 worsen with progesterone administration and improve when treated with the progesterone antagonist onapristone. Unfortunately, onapristone and other currently available progesterone antagonists bear unacceptable side effects and are not suitable for CMT therapy. A recently published pilot trial of Neurotrophin-3 (NT-3) in eight CMT1A patients reported significant improvement in sensory and reflex components of the neuropathy impairment score [12].

Passage et al. [13] reported that chronic treatment with ascorbic acid (AA, Vitamin C) effectively treats PMP22-overexpressing mice. In particular, Passage et al. showed that neuropathy was less severe in AA-treated mice, which also performed much better in a series of clinical tests, i.e. rotarod, beam-walking, grip, and slip tests. In some clinical motor tests, namely the grip test, performance was better at the end than before treatment, indicating that AA not only prevents disease progression, but also partially reverts the phenotype. Histological analyses revealed that the number and percentage of myelinated fibers was higher and myelin was thicker in treated mice than in untreated animals. Finally, treated mice had a normal life span, as opposed to the shorter one of untreated animals. AA promotes myelination *in vitro*, and decreases PMP22 expression both *in vitro* and *in vivo*, possibly through a cAMP-mediated mechanism [13,14]. It is therefore plausible that AA improves the neuropathy by decreasing PMP22 expression. The potential application of AA therapy to the human disease is easy and feasible, as AA is devoid of severe side effects [15]. For these reasons, the effects of AA in the mouse deserve to be tested in humans. The 136th international ENMC workshop discussed issues on clinical trials in CMT1A and agreed on inclusion criteria and primary and secondary outcome measures [16].

We designed a randomized, double-blind, placebo-controlled trial of AA in CMT1A patients, termed the CMT-TRial Italian with Ascorbic Acid Long-term (CMT-TRI*AA*L).

2. Methods/design

The study protocol was approved by the Ethic's committee of the "C. Besta" National Neurological Institute and of all the participating centers. This study complies with the Helsinki Declaration in its most recent version, and with the principles of Good Clinical Practice (GCP) guidelines. The trial is also carried out in keeping with the Italian Health Ministry regulatory requirements.

2.1. Study design

This is a multicenter phase III prospective, randomized, double-blind, placebo-controlled trial, meant to evaluate the safety and efficacy of long-term (2 years) oral therapy with AA (1500 mg/day) or placebo in two doses.

Table 1
List of the CMT-TRI*AA*L eligibility criteria

Inclusion criteria
1. Clinical diagnosis of CMT1A
2. Genetic confirmation (17p11.2-p12 duplication)
3. CMT Neuropathy Score between 1 (excluding the electrophysiological component) and 35 (including the electrophysiological component)
4. Age 18–70 years
5. Ability to accomplish the primary outcome measures
6. Women of child-bearing age only if they declare not to be pregnant or breast feeding at the inclusion into the study and to avoid becoming pregnant during the study
7. Signed informed patient consent
Exclusion criteria
1. Clinical or echographic diagnosis of nephrolithiasis
2. Positive history of recurrent renal colics
3. One or more episodes of renal colic in the 6 months prior to screening
4. Deficit of Glucose-6P-Dehydrogenase
5. Acquired or hereditary hemochromatosis; thalassemia major; sideroblastic anemia
6. Treatment with potential therapeutic agents for CMT1A in the 3 months prior to screening
7. Extra-dietary AA consumption in the 3 months prior to screening
8. Other causes of neuropathy (e.g. diabetes, monoclonal gammopathy, cryoglobulinemia, neoplasms, B12 deficiency, HCV-related liver disease)
9. Other neurological disorders (e.g. multiple sclerosis, cerebrovascular diseases, movement disorders), or major comorbidities (e.g. definite cognitive impairment, psychiatric disease, heart or lung failure, orthopedic or rheumatologic disorders)
10. Limb surgery in the 6 months prior to screening, or planned before final assessment

All of the inclusion criteria and none of the exclusion criteria must be satisfied.

2.2. Patient selection

CMT-TRI*AA*L focuses on symptomatic CMT1A adults of both sexes, followed by eight Italian referral centers. A detailed overview of the eligibility criteria is given in Table 1. Briefly, we decided to include patients of both sexes, aged 18–70, with clinical and genetic diagnosis of CMT1A, with Charcot-Marie-Tooth Neuropathy Score (CMTNS) between 1 and 35 [17]. Patients with contraindication to AA assumption (i.e., nephrolithiasis, glucose 6P-dehydrogenase deficiency, and iron overload), major copathologies and other causes of neuropathy are excluded. Patients must not take supplements of AA in the 3 months prior to screening. Symptomatic treatments for the neuropathy (e.g. drugs for pain) and physiotherapy are allowed; such treatments should not be changed during the study unless necessary, and should be thoroughly recorded.

2.3. Randomization and treatment schedule

All consenting patients fulfilling the inclusion criteria are randomized (central phone randomization). Randomization is stratified by center and disease severity (CMTNS for symptoms and signs, ≤ 8 and >8) [17].