Effect of desipramine on patients with breathing disorders in RETT syndrome

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Effect of desipramine on patients with breathing disorders in Rett syndrome

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Abstract

Objective: Rett Syndrome (RTT) is a severe neurodevelopmental condition with breathing disorders, affecting around one in 10,000 female births. Desipramine, a noradrenaline reuptake inhibitor, reduced the number of apneas in MeCP2-deficient mice, a model of RTT. We planned a phase 2 trial to test its efficacy and its safety on breathing patterns in 36 girls with RTT. Methods: The trial was a 6-month, multicenter, randomized, double-blind, placebo-controlled study registered with ClinicalTrials.gov, number NCT00990691. Girls diagnosed according to clinical examination and confirmed by genotyping were randomly assigned in a 1:1:1 ratio to receive 2–3 mg/kg Desipramine per day (high Desipramine), 1–2 mg/kg Desipramine per day (low Desipramine), or a placebo. The primary outcome was the change of apnea hypopnea index (AHI), defined by the number of apnea and hypopnea events per hour, assessed at 6 months from baseline. Intention-to-treat analysis was applied. Results: The median change in AHI from baseline to 6 months was −31 (IQR: −37 to −11) for the high Desipramine, −17.5 (IQR: −31 to 13) for the low Desipramine, and −13 (IQR: −31 to 0) for the placebo group. We did not find any significant difference in these changes between the groups (P = 0.781). A significant inverse correlation between Desipramine plasma concentration and AHI (r = −0.44; P = 0.0002) was underlined. Interpretation: This first clinical trial of desipramine did not show clinical efficacy. Although required further studies, the significant correlation between Desipramine concentrations and improvement of AHI provided additional and relevant reasons to test the noradrenergic pathway in RTT.
Introduction

Rett Syndrome (RTT) is a complex and severe neurodevelopmental disorder, affecting around one in 10,000 female births.\(^1\) RTT is caused by mutations in the methyl-CpG-binding protein 2 (MECP2) gene.\(^2\) After an apparently normal development until 6–18 months of age, a progressive disorder occurs, characterized by a loss of motor and cognitive skills, gait impairment, stereotypic hand movements, ataxia, seizure, decrease in BMI (Body Mass Index), osteopenia, and scoliosis.\(^1\) Among the symptoms of RTT, breathing dysrhythmia may represent a life-threatening event.\(^3\) Breathing disorders are present in 65 to 93% of the patients and characterized by hyperventilation, hypoventilation, breath holds and apneic events.\(^4\) Studies in validated mouse models of RTT have shown that MeCP2 null male and heterozygous female mice exhibited breathing disturbances with different level of severity (\(^5\)–\(^11\)). Even if the real genetic model of RTT is the heterozygous female mouse, most of the in vivo studies have been done using null male mice as preclinical model mainly due to the potential confounding effects of variable X chromosome inactivation in female mice. The male mouse models exhibit severe breathing dysfunctions between 1 and 2 months of age. During their last week of life, MeCP2 null male mice had frequent apneas. Their medulla had a drastically reduced number of tyrosine hydroxylase (TH)-expressing neurons, Noradrenaline (NA) content, and serotonin (5-HT) content. In vitro experiments using transverse brainstem slices of mice, between 2 and 3 weeks of age, revealed that the rhythm produced by the isolated respiratory network was irregular in MeCP2 null male mice but could be stabilized with exogenous NA.\(^10\) When these mice were treated in vivo with desipramine (DMI), which specifically inhibits NA reuptake, the number of apneas was strongly reduced in comparison to the placebo group (\(\sim 75\%)\) after 3 days of treatment. In addition, the treatment significantly extended their lifespan.\(^12\) This effect, observed with administration by intraperitoneal route was confirmed in another independent study giving DMI by oral route.\(^13\) At cellular level, DMI maintained the number of TH-expressing neurons in the brainstem of MeCP2-deficient mice.\(^12\) On the basis of these preclinical data and the scientific literature showing that RTT patients also present a reduced amount of available NE in the CNS,\(^14\) orphan designation was granted by the European Commission for the treatment of RTT with desipramine in 2011.\(^15\) In view of the aforementioned results in RTT mouse models and the well-known pharmacology of DMI in humans (including children), we aimed to assess its efficacy and safety on breathing patterns in girls with RTT.

Methods

Study design and participants

A multicenter, randomized, double-blind, placebo-controlled phase 2 trial was performed from February 2009 to September 2014. Girls with a clinical diagnosis of RTT and confirmation by genotyping (mutation in the MECP2 gene) were enrolled from six French university hospital sites specialized in neurodevelopmental disorders (Marseille, Lille, Lyon, Paris, Toulouse and Tours). Inclusion criteria were being aged 6–18 years old, weighing up to 60 kilograms, and having breathing disorders outlined during respiratory monitoring in awake patients (apnea-hypopnea index \(\geq 5/h\)). Exclusion criteria were male subjects, patients under 6 years old, patients with a history of status epilepticus, uncontrolled cardiopathy, renal insufficiency or liver disease.

The protocol was approved by the local ethics committee and regulatory authority (Agence Nationale de Sécurité du Médicament et des Produits de Santé). The study was carried out in accordance with the Declaration of Helsinki and with the principles of Good Clinical Practice. All parents gave written informed consent before their daughters participated.

Randomization and masking

After a screening visit, eligible patients were randomly assigned in a 1:1:1 ratio to receive a daily DMI dose between 25 mg to 150 mg according to their weight and the treatment allocation groups corresponding either to the DMI high dosage (2–3 mg/kg per day) or DMI low dosage (1–2 mg/kg per day) or matching placebo for 6 months (Table 1).

DMI and placebo capsules had identical appearance, were centrally blistered, packed, and labeled by AMAST-SIGROUP (Crid Pharma, Montpellier, FRANCE) then sent to the pharmacy of each study site. The randomization sequence was a computer-generated list by AMAST-SIGROUP, which was independent of the study. Randomization was done in blocks of 6 and stratified according to the study site. The sequence was available in opaque sealed envelopes at every site for emergency unmasking. Treatment allocation was masked for patients and physicians.

<table>
<thead>
<tr>
<th>Table 1. Treatment allocation.</th>
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<tr>
<td>Patient weight</td>
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<tr>
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<td>Low dose Group</td>
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<tr>
<td>Placebo Group</td>
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</tbody>
</table>

and their families, treating physicians and physicians assessing outcomes at baseline and during follow up. The randomization list was sent to the Department of Pharmacometry CIC-UPCET, Marseille, France after the database had been cleaned up and locked for statistical analysis.

**Procedures**

After informed consent, RTT girls underwent clinical history and structured clinical examination by experienced physicians including physical examination, breathing recording, and laboratory testing at screening. Disease severity was determined by investigators using a clinical rating scale, the Clinical Severity Score (CSS), which was specifically developed for RTT. The CSS was developed as a measure of clinical severity based on key diagnostic and developmental features for RTT. The CSS is based on 13 individual, ordinal categories. Scores for all items were summed to create a total score (range: 1–58). Higher scores indicate more severe clinical status. Eligible girls returned for visit 1 (baseline) and were randomly assigned to treatment groups. Treatment began with a gradual increase on the day after randomization and a gradual reduction on the day of the 6-month visit, according to the dose schedule. Study visits were at 2 weeks, 4 weeks, 9 weeks, 3 months, and 6 months after randomization. As the treatment was progressively decreased, over 6 weeks for the highest doses, a follow-up visit consisting of a clinical examination and adverse event records was done at 8 months, 15 days after the last capsule. Breathing disorders were assessed using a specific and light device ApneaLink® (ResMed Germany Inc., ref 22301) in all study sites, allowing a non-invasive measurement of respiratory parameters. This device has been previously described and used in several studies for identification of sleep apnea in patients. A Symptom Severity Index (SSI) is a single-channel screening device that measures airflow through a simple nasal cannula connected to a pressure transducer, providing an apnea-hypopnea index (AHI) based on recording time. The ApneaLink® is battery operated, has a sampling rate of 100 Hz, and a 16-bit signal processor. The internal memory storage is 15 MB, which allows for approximately 10 h of data collection. It automatically analyses and derives flow limitation and to exclude them from analysis.

An apnea was defined as a decrease in airflow by 50–80% of baseline for at least 10 sec. The apnea-hypopnea index was defined by the number of apnea and hypopnea events per hour. The ApneaLink® AHI used for analysis was automatically generated by the ApneaLink® software. After the automated analysis, a blinded respiratory physician (JCD) reviewed each recording to eliminate aberrant or invalid AHI data and to exclude them from analysis.

The ApneaLink® firmware version 2.97 and the scoring software version 5.13 were used. During each study visit there was 1 h of monitoring, coupled with a pulse oximetry measurement. Particular attention was paid to the fact that the patient’s mouth had to remain closed during recording. An ancillary study was performed in 6 awake girls from one center (Lyon Centre) to correlate AHI collected with the ApneaLink® and during a polysomnography.

The primary outcome was the change of Apnea-Hypopnea Index at 6 months from baseline. The secondary outcomes were respiratory parameters (respiration rate, cumulated length of breathing disturbances, oxygen saturation). Careful cardiac monitoring including an electrocardiogram was planned at each visit to record heart rate and QT length. All seizures were recorded by parents or caregivers on paper diaries and reviewed by the investigator at each visit. Adverse events were assessed by the investigators throughout the study. Serious adverse events were rated by investigators; these included death and events that were life threatening, persistently or permanently disabling, led to admission to hospital, or a prolonged stay in hospital. All adverse events were collected, monitored and categorized according to the common terminology criteria for adverse events and rated for severity and association with the study drug.

A Symptom Severity Index (SSI) is a 16-item assessment tool filled in by the parents, with each item individually rated from 0 to 8, with 8 being the best possible score and 0 the worst. Each item included a question regarding parental perception of change.

Venous blood samples were collected for DMI plasma concentration and laboratory tests (liver and kidney function tests, complete blood cell count, serum electrolytes, glucose, calcium, cholesterol, uric acid). DMI samples had to be immediately frozen at −20°C or under. Plasma concentrations of DMI at every visit were measured using liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS), in a central laboratory (Pharmacology, Timone Hospital, Marseille, France).

Clinical examination (including recording of respiratory parameters by ApneaLink®), weight, height, and vital signs were recorded at screening, baseline and 1 week, 2 weeks, 4 weeks, 3 months, 6 months and 8 months. CSS was assessed at baseline and 6 months.
Concomitant medications and adverse events were recorded at each visit. Capsule counts were also undertaken at all visits to monitor treatment compliance. Electrocardiogram and DMI concentration were also obtained at each visit. Blood tests were performed at screening, baseline, 2 months and 6 months.

All patient data were recorded on case report forms, which were reviewed by the independent trial monitor. After validation, all results were double-entered into an EpiData base version 3.1. Data entry was controlled for consistency using an SAS version 9.3 according to the protocol and validated in the data management plan.

Statistical analysis
We had little evidence in humans to guide the power of calculation since no data were available on the effects of DMI on RTT girls. The study sample size was not predetermined but similar to that generally used in pilot phase 2 trials and based on patient enrollment study at the study sites.

Descriptive statistics were expressed as median and interquartile range (IQR, 1st quartile–3rd quartile) for continuous variables and as proportions for categorical variables. Continuous variables were expressed as mean and standard deviation (SD). We compared baseline clinical and demographic variables, using Fisher’s exact tests or Kruskal–Wallis tests. A modified intention-to-treat analysis was done at 3 and 6 months follow-up, consisting of all randomized patients who had a valid respiratory assessment at baseline and at least one valid post-baseline assessment. P values of less than 0.05 were considered significant. Comparisons between the DMI and placebo groups were assessed on changes from baseline, using a Kruskal–Wallis test. In an exploratory analysis, Spearman’s correlation coefficients were calculated between AHI and DMI concentration and AHI and change in AHI from baseline to 6 months. A negative change corresponds to a decrease of the number of apnea and hypopnea events per hour.

Safety analysis was based on the reported treatment-emergent adverse events (TEAE) and other safety data (vital signs, ECG). Each adverse event was coded as a so-called preferred term and associated system organ according to an established and validated adverse reaction dictionary (Medical Dictionary for Regulatory Activities version 17.1 MedDRA). Endpoints for TEAE were the number of patients with at least one event related to the study treatment. Statistical analysis was done, using SAS® software version 9.3. Figures was generated from SAS 9.3 and Microsoft® Excel® 2013 (Fig. 3B). This study is registered with ClinicalTrials.gov, number NCT00990691.

Role of the funding source
The sponsor had no role in the study design, data collection, data analysis, data interpretation, or the writing of the report. All authors had full access to the study data and had final responsibility for the decision to submit for publication.

Results
Here, 11 of the 47 girls initially screened did not meet the inclusion criteria (lack of breathing disorders) and two patients withdrew (Fig. 1). Thus, 34 girls were treated, 11 with high dose DMI, 12 with low dose DMI, and 11 with placebo. Also 26 patients completed 6 months of treatment leading to a mean DMI exposure of 50 mg (SD: 0) per day (high DMI Group) and 34.4 mg (SD: 12.9) per day (low DMI group) (Fig. 1). Baseline characteristics

47 patients screened
13 not included
11 did not meet inclusion criteria
2 declined to participate
34 randomly assigned
11 allocated to High
Dose desipramine
12 allocated to Low
Dose desipramine
11 allocated to Placebo

4 discontinued
1 lost to follow up
3 adverse events*
4 discontinued
4 adverse events*

7 completed study
6 completed study
11 completed study

11 included in the safety population
7 included in mITT population at 3 months
5 included in mITT population at 6 months

12 included in the safety population
7 included in mITT population at 3 months
6 included in mITT population at 6 months

11 included in the safety population
9 included in mITT population at 3 months
7 included in mITT population at 6 months

Figure 1. Trial profile, modified intention-to-treat, including randomized patient with valid respiratory assessment. *Adverse events were hypersensitivity (2 weeks), prolonged QTc interval (2 weeks) and constipation (5 weeks) in high dose DMI and status epilepticus (2 weeks), insomnia (2 weeks), and motor dysfunction (9 weeks and 3 months) in low dose DMI.
were similar in all three groups (Table 2) (Appendix S3). Mean age was 10.4 years old (SD: 2.7) and the mean age at the RTT diagnostic was 3.1 years old (SD: 1.3). The mean of BMI was 15.1 kg/m² (SD: 2.2). The mean of CSS was 24.3 (SD: 6.3).

A significant correlation was found for AHI recorded with ApneaLink® or during a polysomnography in the 6 awake girls ($r = 0.738$, $P = 0.037$, Fig. 2). Concerning breathing patterns in awake patients, the median of AHI was 38 (IQR: 18 to 58) and the median of the number of apneas was 19 per hour (IQR: 8 to 31) (Table 1). Although they were not significant, the lowest values were observed in the placebo group (Table 2).

![Figure 2](image)

**Figure 2.** Relation between AHI measured with Apnealink® and AHI measured with polysomnography. AHI, Apnea-Hypopnea Index.

There was no significant difference between groups in the AHI after 6 months (high DMI: $-31$ (IQR: $-37$ to $-11$), low DMI: $-17.5$ (IQR: $-31$ to $13$), placebo: $-13$ (IQR: $-31$ to 0); $P = 0.781$) (Fig. 3). No significant differences were observed between treatment groups in the changes from baseline to month 6 in breathing patterns (number of apneas, number of hypopneas, number of respiratory cycles, respiratory rate, O2 desaturation; % SpO2 below 90%, O2 saturation) (Appendix S1). No significant effect was observed in changes of CSS at 6 months (high DMI: 0 (IQR: $-3$ to 0), low DMI: $-1$ (IQR: $-2$ to $-1$), placebo: $-0$ (IQR: $-1$ to 0); $P = 0.554$). No significant effect was observed for the Symptom Severity Index (Appendix S2).

At 6 months, plasma concentration of DMI was significantly different between groups, consistent with allocation treatment. The plasma concentration of DMI was $119.3$ ng/mL (SD: 46.3) in the group treated with 2–3 mg/kg DMI per day, and $44.8$ ng/mL (SD: 41.9) in the group treated with 1–2 mg/kg DMI per day (Fig. 4). Treatment compliance was 96.9% and there was no difference between groups. In an exploratory analysis on all pharmacological samples ($n = 70$), there was a significant inverse correlation between DMI plasma concentration and AHI ($r = -0.44$, $P = 0.0002$) (Fig. 5A). This correlation was observed at 6 months although it was not significant ($r = -0.42$, $P = 0.19$) (Fig. 5A insert). An inverse correlation between AHI at baseline and AHI change from baseline to M6 has been shown ($r = -0.72$, $P = 0.0004$) (Fig. 5B).
Seven adverse events were given as a reason for withdrawal (hypersensitivity, ECG QTc interval prolonged, status epilepticus, insomnia, severe constipation, and motor dysfunction in 2 patients) (Fig. 1). Six serious adverse events were recorded in 5 patients (3 patients in high DMI, 1 patient in low DMI, and 1 patient in the placebo group). Among them, 2 serious adverse events were related to study treatment (1 hypersensitivity among a patient with high DMI and 1 status epilepticus among a patient in low DMI) (Table 3).

A total of 94 adverse events were reported, of which 88 occurred during the treatment period. These events occurred in 27 patients (79.4%): 11 patients in the placebo group, 9 patients in the high DMI group, and 7 in the low DMI group. Forty five (51.3%) of adverse events were deemed by the investigator to be doubtfully or possibly related to the treatment. And 11 of the 45 events
with a doubtful or possible relationship to the treatment were severe.

No significant differences were observed between treatment groups in the changes from baseline to month 6 on occurrence of epileptic seizures. A total of 166 ECG were performed and judged as normal in 89.8% and 9% were assessed as abnormal and not clinically significant. Two prolonged QT intervals were observed for two consecutive visits of one patient (in the high DMI group) after 2 weeks of treatment, leading to her withdrawal from the study (Table 4).

**Discussion**

To our knowledge, this is the first randomized and controlled clinical trial assessing the noradrenaline-uptake blocker DMI on breathing disorders in RTT girls. This is also the first to specifically assess the effects of a drug on breathing patterns in RTT. Our results did not show any significant difference between the effects of placebo, low DMI, and high DMI on the AHI of awake patients at 6 months. Neither did we show significant effects for any secondary outcome measurements, including other respiratory parameters. Several reasons could explain the lack of clinically significant effects of DMI on breathing patterns in RTT girls. First, as described by several authors, breathing dysfunction in RTT is very complex and highly variable. Second, in addition to the absence of potential efficacy, our trial might have been underpowered to detect a significant effect, due to the lack of previously standardized data available on monthly changes in respiratory patterns of RTT girls used to estimate the number of subjects needing to be included. Third, the rationale for our study was based on results from two relevant studies in mouse RTT model. As previously summarized in a recent review, the construct validity, reproducibility, and robustness of mouse models for the study of RTT has clearly been established. One of the most robust symptoms that has been found in many RTT mouse models, in both males and females, is the breathing phenotype. This fact, together with the genetic homogeneity of the males compared to the females that are subjected to X-chromosome inactivation and its subsequent phenotypic variability, are part of the reasons why it is acceptable to use male mice for the study of RTT. Moreover, the knockout male mouse models also recapitulate other key RTT symptoms such as motor dysfunctions, osteoporosis and epilepsy. At the neurochemical level, the amount of available NE in the CNS is reduced in both RTT patients and knockout male mouse, which is evidence that the biogenic amines stimulation is a high-potential target for the pharmacological treatment in RTT. Although the pre-clinical data showed a

<table>
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<th>System organ class</th>
<th>Preferred term</th>
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<th>Low dose (n = 12)</th>
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</table>

Data are the number of treatment-emergent adverse events (TEAE). In brackets the number of patients with at least one adverse event and the number of serious adverse events (SAE).
clear beneficial effect of DMI on RTT phenotype, the efficacy of the DMI treatment in RTT patients was lower than expected. This could be due to the wide age range of the RTT patients as well as the heterogeneity in their MECP2 mutation. Other limiting factors could be the administration route (ip vs oral) that limited the DMI bio-availability and/or the metabolic rate of DMI that seemed to greatly vary among RTT patients.

Furthermore, due to the pilot phase of the study, two doses of DMI were assessed in order to best discriminate a potential pharmacodynamic effect among the range of DMI tested. In this context, one important point of our study is the exploratory pharmacological analysis performed to test the correlation between AHI and DMI plasma concentrations. In several studies, inter-individual differences in side effects and treatments responses have been associated with variability of tricyclic drug plasma concentrations.27 Our clinical trial also showed the inter-variability of DMI plasma concentrations between treatment groups (DMI low and high dose) and the intravariability of DMI within each treatment group. Due to this pharmacological variability, we tested whether there was a relationship between plasma DMI concentration and AHI, whatever the treatment allocation and the study visits (Fig 4). Interestingly, and even if this result should be interpreted cautiously, a correlation was found, suggesting a possible relationship of concentration effect between desipramine and AHI. In other words, a higher level of DMI concentration would be associated to a greater improvement of AHI. Similarly, we also found an inverse correlation between AHI at baseline and AHI change from baseline to M6, suggesting a greater improvement in patients with high levels of breathing disorders at baseline. Although all of these findings require further studies, the analyses on relationship between systemic exposure levels and AHI provide additional useful clinical information and relevant reasons for testing the noradrenergic pathways in RTT.

One strength of our study was the careful consideration taken to establish inclusion and exclusion criteria and the methodological design assessing two DMI doses in comparison with placebo, DMI plasma concentrations and the rigorous assessment of breathing patterns, using the same device and a centralized blinded analysis. One limitation may include the use of a respiratory monitor for the assessment of AHI rather than the gold standard polysomnography. However, the use of this device has been previously validated, and it is more practical than polysomnography for use in a clinical trial including several and repeated measurements in ambulatory patients. We have also validated in this study the device for measuring AHI in awake children with RTT. However, the ApneaLink® does not include an automated measurement of hyperventilation events, a measurement lacking in this trial. Nevertheless this measurement was not considered to be essential because this trial was constructed to test whether the drug target reported in animal models was valid in patients.

One other important point was to ensure the safety of participants. Even if DMI had previously been used in humans for several conditions (major depression, pain) or in children for enuresia or attention-deficit hyperactivity disorder28,29, a careful cardiac and epileptic monitoring was performed on selection (excluding uncontrolled cardiopathy) and throughout the study. RTT girls were assessed for cardiac safety following the American Heart Association’s recommended guidelines: (1) before starting DMI administration (2) during dose adjustments and (3) periodically during maintenance.30 Among the 166 ECG performed during the study, two prolonged QT intervals for one patient in the high dose DMI were judged to be clinically significant, leading to her withdrawal from the study visits (Fig 4). Interestingly, and even if this result should be interpreted cautiously, a correlation was found, suggesting a possible relationship of concentration effect between desipramine and AHI. In other words, a higher level of DMI concentration would be associated to a greater improvement of AHI. Similarly, we also found an inverse correlation between AHI at baseline and AHI change from baseline to M6, suggesting a greater improvement in patients with high levels of breathing disorders at baseline. Although all of these findings require further studies, the analyses on relationship between systemic exposure levels and AHI provide additional useful clinical information and relevant reasons for testing the noradrenergic pathways in RTT.

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trial (Table 4 and Fig. 1). Even if this adverse event is well described with tricyclic drugs, the occurrence required particular attention in RTT due to reduced cardiac vagal tone with a higher risk of cardiac arrhythmias and sudden death.\textsuperscript{31} DMI is also known to decrease the seizure threshold. For this reason, we carefully monitored the occurrence of seizures and did not find any significant worsening of epilepsy during the study. Other adverse events related to the atropinic effect of DMI were noted, such as constipation and motor dysfunction. These adverse events, although well-known and usually reversible, led to premature discontinuation of the drug in 7 patients and also affected the trial performance.

In conclusion, we carried out the first randomized, double blind, placebo controlled trial to assess the effects of desipramine on breathing patterns in girls with RTT. We did not show any clinical efficacy whatever the dose tested. Nevertheless, a non-significant decrease of AHI in awake patients in the DMI groups at 6 months and the significant and positive relationship between DMI systemic exposure levels and improvement of AHI provide scientific and pharmacological confirmation for the implication of noradrenergic pathway in RTT. We also took repeated recordings of AHI and other respiratory parameters, which may be useful for designing future clinical trials on breathing abnormalities in RTT.

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Author Contribution

JMa, JCD, EJ, JCR, LA, LV, OB, and JMic contributed to the design of the study. JMa was the principal investigator and EL, PC, CC, CCor, YC, RJC, VP, PF, LV, and NRB participated in the study as investigators. JMa, LA, OB, and JMic participated in the implementation, the planning, and the coordination of the trial. JCD centralized and analyzed breathing data. EJ and RT wrote the statistical analysis plan of the protocol and analyzed the data. JMa and JMic did the literature research. EJ and RT drew up tables and figures for the paper. JMa and JMic wrote the first draft of the paper. EJ and RT wrote the section analysis. All authors interpreted the data, contributed to the subsequent versions of the article, and approved the final version.

Conflicts of Interest

The authors have no conflicts of interest.

References


Supporting Information
Additional Supporting Information may be found online in the supporting information tab for this article:

Appendix S1. Respiratory parameters (AHI, Respiratory rate, Apneas, Hypopneas, O2 desaturation and SpO2 below 90%) and O2 Saturation.
Appendix S2. Symptom Severity Index (SSI).
Appendix S3. Type of mutations.