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Effects of Low-Dose Rimonabant on Locomotor Endurance After Spinal Cord Injury: A Translational Proof-of-Concept

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ABSTRACT

Improving locomotor function is a key goal in spinal cord injury (SCI) rehabilitation. CB1 cannabinoid receptor antagonists enhance neuronal excitability and may support motor recovery. To evaluate the safety and efficacy in improving locomotion of short-term, low-dose Rimonabant (a CB1 receptor antagonist/inverse agonist) in moderate/incomplete SCI, we conducted: A) three sequential crossover, randomized, blind, placebo-controlled preclinical experiments in a rat model of moderate contusive thoracic SCI assessing safety and improvements in locomotor endurance after treatment with low doses of Rimonabant (0.062–0.800 mg/kg); and B) a double-blind, randomized, placebo-controlled crossover clinical trial (RIMOFATSCI-1) involving eight individuals with chronic incomplete SCI testing safety and efficacy after 5-day treatment with Rimonabant (2.5 and 5 mg/day). Preclinical analyses did not show treatment-related adverse events but revealed that 0.250 mg/kg Rimonabant significantly improved endurance 24 hours after administration ($p < 0.01$, Cohen's $d = 0.808$), with clinically meaningful improvements in 35.7% of responders. The clinical trial confirmed safety, with no psychiatric or severe adverse events. Although functional outcomes improved over time, no significant treatment-specific effects were detected. Thus, low-dose Rimonabant is safe and may acutely enhance locomotor endurance post-SCI. These findings support further investigation in larger trials.

KEYWORDS

Motor recovery, CB1 cannabinoid receptor, Rimonabant, neuronal excitability, incomplete spinal cord injury, translational evidence.

INTRODUCTION

Restoring and improving locomotion is a primary goal for individuals with traumatic spinal cord injury (SCI), as it is significantly associated with quality of life and personal independence [1]. Effective locomotion requires the coordinated activation of trunk and lower-limb muscles, relying on both muscle strength and endurance. This coordination depends on the precise temporal activation of motoneuron pools [2-5], modulated by continuous sensory feedback [6].

At the spinal level, locomotor activity is organized by neural networks located in the lumbosacral cord, commonly referred to as the central pattern generator (CPG), which coordinates alternating and flexor–extensor muscle activity [7,8]. The function of these networks is governed by the balance between excitatory and inhibitory inputs (E/I balance), primarily mediated by glutamatergic excitation and GABAergic/glycinergic inhibition [9]. Descending inputs from supraspinal centers modulate this spinal circuitry, ensuring appropriate initiation, intensity, and adaptability of locomotor output.

Following incomplete SCI, disruption of descending pathways reduces the excitatory drive reaching the lumbosacral circuitry, leading to impaired motoneuron recruitment, diminished strength, reduced endurance (motor fatigability, a physiological dimension of the multifaceted symptom of fatigue), and altered coordination. Importantly, injury-induced alterations in the E/I balance further compromise CPG function and locomotor output [10].

Experimental evidence suggests that preservation or restoration of excitatory neuronal function is critical for motor recovery [11,12], whereas excessive inhibition or loss of excitatory phenotype negatively impacts functional outcomes [13]. Notably, excitatory neurons appear particularly vulnerable to persistent dysfunction and degeneration after SCI [14], further compromising network excitability and reducing the capacity of spinal circuits to maintain the excitation–inhibition balance, thereby reinforcing the need for strategies that preserve or enhance excitatory drive.

Consistent with this rationale, therapeutic approaches—including pharmacological modulation and electrical stimulation—have shown that restoring excitatory activity within spinal networks can improve motor performance after injury [15-17].

The endocannabinoid system plays a key role in modulating central nervous system (CNS) excitability. CB1 cannabinoid receptor is widely expressed in motor control regions—including cortical, subcortical, and spinal structures—as well as in skeletal muscle [18-21]. CB1 activation inhibits neurotransmitter release via retrograde signaling, affecting both excitatory and inhibitory synapses [22-24]. Preclinical studies have shown that targeting the endocannabinoid system can alleviate motor dysfunction, spasticity, and abnormal reflexes [20, 25-27].

Rimonabant, a blood-brain-barrier-penetrant CB1 antagonist/inverse agonist, modulates neuronal excitability by reducing CB1-mediated inhibition of neurotransmitter release [28-29]. A single 20 mg dose of Rimonabant has been shown to lower motoneuron activation thresholds at both cortical and spinal levels, shifting the E/I balance toward excitation [30]. Although reducing activation thresholds alone do not ensure improved motor performance—and may even exacerbate spasticity—enhancing responsiveness to descending excitatory input may improve motoneuron recruitment.

Rimonabant was withdrawn from the European market in 2009 due to an increased risk of psychiatric and neurological adverse events—such as anxiety, depression, agitation, and suicidal ideation—at a 20 mg dose [28, 31-32]. Gastrointestinal adverse events were also reported at this dose [28, 33]. However, clinical data indicate that lower doses (e.g. 5 mg/day) do not significantly increase adverse event frequency and are generally considered safe [32].

Based on this evidence, we hypothesize that modulating E/I balance with low doses of Rimonabant can restore physiological motoneuron activation dynamics and improve locomotion after SCI without inducing significant adverse effects. To test this hypothesis, we first conducted a series of preclinical experiments in a rat model of moderate contusive SCI evaluating both safety and locomotor outcomes following acute administration of low doses of Rimonabant. Building upon these findings, we subsequently performed a proof-of-concept, randomized, double-blind clinical trial in individuals with chronic incomplete SCI and preserved walking capacity, assessing the safety and short-term functional effects of a 5-day low-dose Rimonabant treatment.

MATERIALS AND METHODS

Preclinical Experiments

Three sequential dose-response experiments were conducted using a rat model of moderate contusive thoracic spinal cord injury SCI to evaluate the safety and effects on running endurance of low doses of Rimonabant. The three experiments used randomized, blinded, placebo-controlled crossover block designs (Figure 1). In total, 60 experimental conditions were evaluated in 15 adult male Wistar rats (n=5 *per* experiment).

All manipulations and treatments were carried out in full accordance with the guidelines on care and management of animals established by the European Union (directive 2010/63/UE), the guidelines on the use of animals for Neuroscience Research of the Society for Neuroscience, the NIH guide for care and use of laboratory animals, and the normative R.D. 53/2013 from the Spanish Ministry of Presidency and the Agriculture Council of the Castilla-La Mancha animal ethics committees. All procedures were approved by the Hospital Nacional de Parapléjicos Animal Care and Use Committee (153BCEEA/2016). All efforts were made to implement the 3Rs principles, minimizing both the number of animals used and their suffering.

After a post-injury recovery period allowing stabilization of spontaneous recovery, animals received placebo or Rimonabant according to each experimental protocol. Safety assessments included daily monitoring of adverse events from the time of surgery onward. Side effects were evaluated through sensory function testing using the Von Frey test on the hind paws, locomotor assessment using the Basso–Beattie–Bresnahan (BBB) scale, stool form and dry weight analyses for intestinal alterations (only in experiment 1), and body weight measurements to monitor general condition.

Endurance capacity was assessed on a 5º inclined treadmill (Cibertec Cinelec 21, Spain) equipped with an electric shock grid. Endurance was defined as the total duration of active running behavior until exhaustion, established as ≥5 consecutive seconds on the shock grid on two occasions. Endurance was measured at baseline, followed by immediate treatment, and re-evaluated 2 and 24 hours after treatment. The primary efficacy endpoint

was ΔE, calculated as the percentage change in endurance relative to baseline: $\Delta E = 100 * \frac{E_X - E_{BL}}{E_{BL}}$, where E_X represents endurance at time X and E_{BL} baseline endurance.

Experiment 1 evaluated the safety and endurance-related effects of placebo and Rimonabant at 0.250, 0.125, and 0.062 mg/kg (equivalent to approximately 3, 1.5, and 0.75 mg in a 70 kg human; [34]) across four treatment blocks. Intestinal effects were additionally monitored through stool form and dry weight measurements. The treadmill protocol consisted of 10 minutes at 0.17 m/s, followed by 5 minutes at 0.25 m/s and 5 minutes at 0.33 m/s.

Experiments 2 and 3 evaluated placebo or Rimonabant at 0.8, 0.25, and 0.125 mg/kg (approximately equivalent to 9, 3, and 1.5 mg human doses, respectively). In Experiment 3, the recovery period was extended to 9 weeks because of early postoperative complications, particularly soiled abdomens. Safety assessments were identical to those in Experiment 1, except that stool analyses were omitted. The treadmill protocol was more demanding and consisted of 5 min at 0.17 m/s, followed by 7.5 min at 0.25 m/s and 7.5 min at 0.33 m/s.

Data Analysis: Data completeness was verified prior to analysis, and no relevant missing values were identified. Endurance outliers were screened before treatment unblinding using Tukey's Fence (k = 1.5), based on baseline endurance (E_{BL}) and changes in endurance (ΔE). Datasets are available in *Supplementary data 1*.

Results are presented as mean ± SD or as violin or box-and-whisker plots, as appropriate. Parametric comparisons were performed using repeated-measures ANOVA and paired or unpaired t-tests. Non-parametric analyses included the Wilcoxon signed-rank test and Friedman test. General linear models were additionally applied when appropriate. Qualitative comparisons (responder analysis) were assessed using Fisher's exact test, and effect sizes were estimated using Cohen's d. Statistical analyses were performed using JASP version 0.19.3 [35].

Detailed experimental procedures, including surgeries, drug preparation, behavioral testing, stool processing, and histological analyses, are provided in *Supplementary Methods 1*.

Clinical Trial

A clinical trial entitled *"Effects of a CB1 receptor antagonist/inverse agonist (Rimonabant) on ambulation ability and endurance in traumatic incomplete spinal cord injury: proof of concept (RIMOFATSCI-1)"* was conducted between 2021 and 2022. The study was prospectively registered in the European Clinical Trials Database (EUDRA_CT_2020-003175-17, see *Supplementary Methods 2*) and the Spanish Clinical Studies Registry (REec, No. 2020-003175-17; available at https://reec.aemps.es/reec/estudio/2020-003175-17, last accessed on November 3rd, 2026) before enrolment of the first participant. A supplementary ClinicalTrials.gov registration (NCT05398913; available at https://clinicaltrials.gov/study/NCT05398913?cond=Spinal%20Cord%20Injury&intr=Rimonabant&viewType=Card&rank=1, last accessed on November 3rd, 2026) was subsequently completed in May 2022. RIMOFATSCI-1 was a prospective, randomized, double-blind, placebo-controlled, crossover proof-of-concept study designed to assess the safety of two low doses of Rimonabant and explore their potential to improve motor endurance. Eight

adult participants (men and women, aged 18–65) with chronic traumatic incomplete SCI (≥12 months post-injury) and preserved walking capacity were included. The study comprised three treatment arms (blocks), each involving 5 consecutive days of treatment with either placebo, 2.5 mg/day Rimonabant, or 5 mg/day Rimonabant (Figure 2). All participants received each treatment in a randomized order, separated by 6-week washout periods.

Efficacy Measures: The primary efficacy outcome was the percentage change in meters walked during the 6-Minute Walk Test (6MWT) relative to baseline. Secondary outcomes included:

- Perceived fatigue (modified Borg Scale post-6MWT)
- 10-Meter Walk Test (10MWT)
- Fatigue Severity Scale (FSS)
- Patient's Global Impression of Change (PGIC)
- Health-related quality of life (EQ-5D questionnaire)
- AIS motor score

Safety Assessments comprised:

- Adverse Events (AEs)
- Vital signs (temperature, blood pressure, heart rate, weight)
- Electrocardiogram (ECG)
- Laboratory tests (blood and urine)
- Pain (Visual Analogue Scale, VAS)
- Spasticity (Modified Ashworth Scale, MAS; Penn Spasm Frequency Scale, PSFS)
- Psychological status (Beck Depression Inventory, BDI; Hospital Anxiety and Depression Scale, HADS)

Ethics and Oversight: The study was approved by the Institutional Review Board (CEIm, Gerencia de Atención Integrada de Talavera de la Reina, Toledo; Ref. 09/2020) and by the Spanish Agency of Medicines and Medical Devices (AEMPS). The trial complied with Good Clinical Practice guidelines and the Declaration of Helsinki. All participants provided written informed consent prior to enrollment. Trial monitoring was performed by Fundación Teófilo Hernando CRO to ensure data integrity. All clinical procedures and data collection were conducted at the National Hospital for Paraplegics (Toledo, Spain), a specialized center in SCI care.

Data Analysis: baseline demographics and clinical characteristics were described per treatment sequence. AEs were coded using MedDRA and tabulated by group. The primary outcome (6MWT) was analyzed using ANOVA to assess treatment effects and temporal progression. Baseline vs. Day 5 comparisons and overall trends were assessed. Secondary outcomes were analyzed similarly. Non-parametric variables were evaluated using the Friedman test. Further details on inclusion/exclusion criteria, randomization and blinding procedures, and assessments are provided in *Supplementary Methods 2*. Datasets are available from the senior authors of this article upon reasonable request.

We used the *Scientific Writing Assistant* (a custom GPT from OpenAI, June 2025 version, available at: https://chat.openai.com/gpts) to support manuscript editing and scientific language refinement.

RESULTS

Preclinical safety: low-dose Rimonabant Is well tolerated in a rat model of Spinal Cord Injury

To evaluate the safety of low-dose Rimonabant in the context of SCI, we analyzed data from 15 animals undergoing surgery and treatment with different doses of Rimonabant in three preclinical experiments. All adverse events (AEs) were recorded and classified as binary variables (present/absent). A total of 23 AEs were documented, primarily consisting of skin ulcers and urinary tract infections (Table 1).

Skin ulcers occurred in 6 animals (40%) during the sub-acute post-injury period, prior to treatment. These lesions, typically located in the flanks or inguinal region, were managed by veterinary staff and did not preclude evaluations. Urinary infections, a common complication in this SCI model, affected 9 animals (60%) in the first days after surgery and were resolved within the first week with antibiotics. In Experiment 3, four animals developed soiled abdomens associated with infections, requiring over two weeks to resolve and delaying treatment initiation. Three animals exhibited transient signs of pain—two associated with ulcers and one with urinary infection—successfully treated with buprenorphine. Importantly, all AEs began before any treatment was administered, although some events persisted following the treatment, indicating no relationship with the investigational product.

Motor and Sensory Safety Outcomes: Sensitivity (Von Frey test) and locomotor function (BBB scale) were assessed 24 hours before and 48 hours after treatment and compared using repeated-measures ANOVA (*Supplementary Results 1*). Neither parameter showed significant differences across the experiments. However, in Experiment 1, a general reduction in sensitivity (Von Frey score) was observed after treatment with vehicle and 0.0625 mg/kg Rimonabant nearly reaching significance ($p = 0.054$), though this trend was not confirmed in subsequent experiments.

Gastrointestinal and Weight Assessments: To evaluate gastrointestinal effects, stool consistency and dry weight were measured at baseline and 24- and 48-hours post-treatment in Experiment 1. No differences in stool form were noted. A minor, non-significant reduction in stool dry weight was observed, particularly in vehicle-treated animals. Body weight was monitored throughout all experiments. No treatment-related differences were found.

In summary, none of the evaluated functional, sensory, physiological, or histological variables showed significant treatment-related effects. These findings support the safety of low-dose Rimonabant (up to 0.8 mg/kg, equivalent to up to ~9 mg in humans) in preclinical models of chronic SCI. Details on the analyses and results are available at *Supplementary Results 1*

Clinical safety: low doses of Rimonabant are safe for treatment of individuals with SCI

To translate preclinical safety findings to a clinical context, we conducted a randomized, double-blind, placebo-controlled, crossover trial (RIMOFATSCI 1) evaluating low-dose Rimonabant in eight individuals with chronic SCI. Participants received placebo, 2.5 mg, and 5 mg of Rimonabant across three treatment blocks. Safety outcomes included

adverse events, clinical and laboratory evaluations, and assessments of neurological and psychological status.

Participant Characteristics: The study population included 6 males and 2 females, with a mean age of 45.0 ± 12.2 years and an average time since injury of 128 ± 96 months. Lesions were primarily cervical (n = 6), with varied etiologies including road traffic accidents, work-related injuries, and falls. Functional independence, measured by the spinal cord independence measure (SCIM), averaged 75.4 ± 15.1.

Adverse Events Profile: A total of 14 AEs were reported by 6 of 8 participants (75%). All but one AEs (93%) were mild and did not require intervention. The only moderate AE —a tendon disorder— occurred during the placebo treatment block and was judged as unrelated to the study drug. No severe adverse events were reported. Related AEs (n = 6) were observed in three participants during active treatment (Table 2).

Nature of Related AEs: Events considered possibly related to Rimonabant included gastrointestinal symptoms (mouth ulceration, nausea, abdominal pain), nervous system events (headache, spasticity), and muscle spasms. All were mild in intensity and transient.

Clinical and Laboratory Assessments: Comprehensive evaluations—including physical examinations, ECG, vital signs, hematology, biochemistry, thyroid function, urinalysis, and validated scales for depression (BDI, HADS-D), anxiety (HADS-A), spasticity (MAS, PSFS), and pain (VAS)—showed no significant worsening during or after treatment. No psychiatric adverse events were reported.

In summary, low-dose Rimonabant (2.5–5 mg/day) for 5 consecutive days was well tolerated in individuals with chronic SCI. The overall AE profile was mild, transient, and consistent with safety data from Rimonabant product information, placebo-controlled trials and post-marketing surveillance (RIO and STRATUS series of clinical trials, see [36, 37], respectively). These findings support the clinical safety of Rimonabant at low doses for SCI individuals and warrant further investigation in larger trials and with longer duration of the treatment.

Preclinical efficacy: single low-dose Rimonabant enhances locomotor endurance in rats with spinal cord injury

To evaluate the efficacy of low-doses of Rimonabant to improve locomotor performance in SCI, we measured running endurance in a rat model of moderate contusion. In three crossover placebo-controlled experiments, 15 adult rats sequentially received a single dose of each treatment with a 6-days washout period between treatments (Figure 1). Endurance, defined as active running time on the treadmill, was assessed at baseline, followed by treatment and re-evaluation 2 h and 24 h later (Figure 3A). The primary efficacy end-point is the change in endurance relative to baseline measurement, expressed as $\Delta E = 100 * \frac{E_X - E_{BL}}{E_{BL}}$ (E_X and E_{BL} correspond to endurance at time X or at baseline (BL), respectively).

Placebo effects (Figure 3B)**.** In the placebo-treated animals, endurance values exhibited a transient decline at 2 h post-administration (mean ΔE = −3.0 ± 6.0%) that returned to baseline levels by 24 h (mean ΔE = 0.39 ± 3.37%). Recovery between the 2 h and 24 h time points was statistically significant (Wilcoxon signed-rank test, W = 5.5, p = 0.016).

Efficacy of low doses of Rimonabant. Placebo effects were compared to values after single doses of 0.062, 0.125, 0.250, and 0.800 mg/kg Rimonabant (approximately equivalent to 0.75, 1.5, 3, and 9 mg human doses, respectively) to assess their efficacy.

As illustrated in Figure 3C, at **2 h post-treatment**, none of the tested doses of Rimonabant (0.062, 0.125, 0.250, and 0.800 mg/kg, equivalent to 0.75, 1.5, 3, and 9 mg human doses, respectively) improved endurance relative to baseline. Indeed, values across all treatment groups showed average reductions comparable to those of the placebo. Interestingly, data distribution reveals that the average reductions results from a notable minority (30% of all values) displaying decreases exceeding 5% while most animals maintained baseline endurances.

In contrast, at **24 h post-treatment**, consistent increases in endurance supported the efficacy of low doses of Rimonabant (Figure 3D). Mean ΔE progressively increased with dose, showing a rapid increase up to 0.125 mg/kg followed by a more gradual increase toward the highest dose tested. Paired comparisons against placebo demonstrated that ΔE was significantly larger for the 0.250 mg/kg dose (paired t-test: $t(11) = -2.799$, $p = 0.009$), with a large effect size (Cohen's $d = 0.808$). The 0.800 mg/kg group also reached nominal significance ($t(7) = -1.974$, $p = 0.044$).

Responder analyses (Figure 3E) were performed using the upper 99% confidence limit of placebo variability (mean + 2.576 × SD = 9.06) as the threshold criterion. Under this definition, responder frequencies exceeded 20% at doses ≥0.125 mg/kg and peaked at 35.7% in the 0.250 mg/kg group (Figure 3E). Fisher's exact test confirmed the significance of the increase in responder frequency at 0.250 mg/kg relative to placebo (35.7% vs 0%, $p = 0.046$).

Responder distribution was strongly associated with motor performance, measured as baseline BBB locomotor scores. Positive responses were concentrated exclusively in animals with BBB ≤13, particularly within BBB classes 11–12. Consistently, animals with BBB >13 remained within a narrow fluctuation range (−2.33% to +2.86% relative to baseline), whereas the BBB ≤13 subgroup accounted for nearly all high-amplitude responses observed in the study. Tissue preservation analyses showed a broadly similar pattern, although the association was less pronounced. Higher white matter preservation (%55%) within the penumbra region was associated with highly stable endurance and minimal treatment responsiveness, and all but one responder individual preserved less than the 55% white matter.

Taken together, these findings support the existence of endurance-enhancing effects induced by low doses of Rimonabant following spinal cord injury and identify 0.250 mg/kg Rimonabant as the optimal dose within the tested range, combining significant population-level efficacy, large effect size, and the highest frequency of clinically meaningful responders. The 0.800 mg/Kg dose was not considered optimal, despite significant effects, because it almost duplicates the doses demonstrated to be secure in RIO and STRATUS clinical trials (5 mg, [36,37], respectively) and did not outperform the 0.250mg/Kg dose (equivalent to 3 mg in humans).

Detailed descriptions of the analyses and results on the efficacy of treatments are available at *Supplementary Results 2*.

Clinical Evidence: No Clear Efficacy Signal in Human Trial

In the RIMOFATSCI-1 clinical trial, eight participants with chronic SCI were randomized to receive 2.5 mg, 5 mg Rimonabant, or placebo in a crossover design. The primary efficacy endpoint was the 6-minute walk test (6MWT). Overall, participants improved their 6MWT performance during the study (Figure 4, left), but these gains were not associated with any specific treatment arm. Repeated-measures ANOVA confirmed the significant effect of time ($p = 0.014$), but no treatment ($p=0.974$), sequence ($p=0.976$), or interaction effects (treatment x sequence: $p=0.279$; treatment x time: $p=0.913$; sequence x time: $p=0.243$; treatment x sequence x time: $p= 0.3$). Similar results were observed analyzing the modified Borg scale (secondary outcome measure, Figure 4, right). Repeated-measures ANOVA confirmed the significant effect of time ($p = 0.025$), but no treatment ($p=0.805$), sequence ($p=0.295$), or interaction effects (treatment x sequence: $p=0.143$; treatment x time: $p=0.839$; sequence x time: $p=0.454$; treatment x sequence x time: $p= 0.955$).

Secondary outcomes—including the Borg scale, 10-meter walk test, AIS motor score, Fatigue Severity Scale (FSS), Patient Global Impression of Change (PGIC), and EQ-5D quality of life—showed no significant differences between treatment groups. Improvements were observed across time in all groups, suggesting a general benefit from participation, but not a specific drug effect.

In summary, while preclinical evidence supports a beneficial effect of 0.250 mg/kg Rimonabant on locomotor performance in rats, this efficacy was not replicated in the small, short-duration human trial. Further studies with larger sample sizes and extended treatment duration are needed to conclusively assess clinical efficacy.

DISCUSSION

This study investigated the safety and efficacy of low-doses of Rimonabant—a selective CB1 receptor antagonist/inverse agonist— for enhancing locomotor endurance following spinal cord injury. Using a translational approach, we first established preclinical evidence in a well-characterized rat model of moderate contusive SCI. We then extended these findings to a first randomized, double-blind, placebo-controlled crossover proof-of-concept clinical trial in individuals with chronic incomplete SCI. Our results support the safety of short-term low-dose Rimonabant and provide preliminary preclinical evidence for its efficacy in improving locomotor performance.

In the preclinical studies, a single administration of low-dose Rimonabant produced delayed but measurable improvements in treadmill-based locomotor endurance. No beneficial effects were observed 2 hours after treatment; however, clear efficacy signals emerged at 24 hours post-administration. Endurance improvements increased progressively across doses, with 0.250 mg/kg identified as the optimal dose. This dose significantly increased endurance relative to placebo (paired t-test, $p = 0.009$) and produced a large effect size (Cohen's $d = 0.808$). Importantly, treatment effects were not uniformly distributed across the cohort but were driven by a distinct responder subpopulation. Using a placebo-derived threshold for clinically meaningful improvement, 35.7% of animals treated with 0.250 mg/kg met responder criteria compared with 0% in the placebo condition (Fisher's exact test, $p = 0.046$).

In the human trial (RIMOFATSCI-1), treatment with Rimonabant (2.5 and 5 mg/day) over five days was well tolerated, with no severe or psychiatric adverse events observed. All eight participants completed the study as per protocol. Although 6-Minute Walk Test (6MWT) performance improved across time, no significant differences between treatment conditions were detected. Secondary outcomes (fatigue, gait speed, quality of life) mirrored this pattern, showing time-related improvements without clear treatment effects. While the clinical improvements did not reach statistical significance, in humans, between treatments, the overall upward trend suggests a possible learning or training effect across blocks. The use of a crossover design, while statistically efficient for rare conditions, may introduce carryover effects despite washouts, particularly when evaluating neuroplasticity-based interventions. Additionally, the relatively high baseline function of participants may have limited the observable treatment window.

There are also two other key considerations that we deem important in comparing human and animal data. First, most of the individuals with SCI had cervical lesions (6/8), while all the animals had thoracic lesions. Second, humans are bipedal, which may suggest that more time is needed for the modification of motor system excitability to translate into improved performance. Future studies will help clarify this difference.

To our knowledge, this is the first translational study to assess the motor-enhancing effects of a CB1 receptor antagonist in the context of SCI. Previous work has primarily focused on CB1 agonists to manage spasticity or pain [38-39]. Strengths of this study include the dual-level (preclinical-clinical) design, use of validated locomotor outcomes, blinded assessments, and complete follow-up. The integration of endurance metrics with safety readouts strengthens translational validity.

Limitations include a small clinical sample size, short treatment duration, and high inter-individual variability. The study may have been underpowered to detect modest between-treatment differences. The treadmill-based endurance measure used preclinically differs in construct from the 6MWT, although both assess sustained locomotor output. Finally, our study remains exploratory, and the findings require evaluation in larger, powered trials.

In summary, short-term, low-dose Rimonabant is safe and well tolerated in both preclinical and clinical settings of moderate/incomplete spinal cord injury (SCI), showing no psychiatric or relevant adverse events. While a single dose significantly improves locomotor endurance preclinically, the clinical trial demonstrates its safety profile and suggests potential functional benefits. These findings support the repurposing of Rimonabant or related CB1 antagonists as adjunctive therapies to enhance motor recovery. Future research in larger cohorts is warranted to define optimal dosing and timing, as well as to improve patient selection based on residual motor function. Furthermore, prospective studies should evaluate extended treatment durations and the combination of this pharmacological modulation with task-specific training or other rehabilitation strategies.

REFERENCES

1. Ferber, G. A., Anderson, K. D. Recovery Insights Following Spinal Cord Injury: A Consumer's Perspective. *Phys Med Rehabil Clin N Am*. 36, 139-154 (2025).

2. Yakovenko, S., Mushahwar, V., VanderHorst, V., Holstege, G., Prochazka, A. Spatiotemporal activation of lumbosacral motoneurons in the locomotor step cycle. *J. Neurophysiol.* **87**, 1542–1553 (2002).
3. Ivanenko Y.P. et al. Temporal components of the motor patterns expressed by the human spinal cord reflect foot kinematics. *J Neurophysiol*. **90**, 3555-3565 (2003)
4. Cappellini, G., Ivanenko, Y. P., Dominici, N., Poppele, R. E., Lacquaniti, F. Migration of motor pool activity in the spinal cord reflects body mechanics in human locomotion. *J Neurophysiol*. **104**, 3064-3073 (2010).
5. Dominici, N. et al. Locomotor primitives in newborn babies and their development. *Science* **334**, 997-999 (211).
6. Kiehn, O. Locomotor circuits in the mammalian spinal cord. *Annu. Rev. Neurosci*. **29**, 279-306 (2006).
7. Arber, S., Costa, R. M. Connecting neuronal circuits for movement. *Science* **360**:1403-1404 (2018).
8. Ramírez-Jarquín, U. N, Tapia R. Excitatory and Inhibitory Neuronal Circuits in the Spinal Cord and Their Role in the Control of Motor Neuron Function and Degeneration. *ACS Chem Neurosci*. **9**, 211-216 (2018).
9. Ramírez-Jarquín, U. N., Lazo-Gómez, R., Tovar-Y-Romo, L. B., Tapia, R. Spinal inhibitory circuits and their role in motor neuron degeneration. *Neuropharmacol.* **82**, 101-107 (2014).
10. Courtine, G., Sofroniew, M. V. Spinal cord repair: advances in biology and technology. *Nat Med.* **25**, 898-908 (2019).
11. Bertuzzi, M., Chang, W., Ampatzis, K. Adult spinal motoneurons change their neurotransmitter phenotype to control locomotion. *Proc Natl Acad Sci U S A*. 115, E9926-E9933 (2018).
12. Chen, B. et al. Reactivation of Dormant Relay Pathways in Injured Spinal Cord by KCC2 Manipulations. *Cell* **174**, 521-535 (2018).
13. Bertels, H., Vicente-Ortiz, G., El Kanbi, K., Takeoka, A. Neurotransmitter phenotype switching by spinal excitatory interneurons regulates locomotor recovery after spinal cord injury. *Nat Neurosci*. **25**, 617-629 (2022).
14. Li, Q. et al. Reduction of prolonged excitatory neuron swelling after spinal cord injury improves locomotor recovery in mice. *Sci Transl Med*. **16**, eadn7095; 10.1126/scitranslmed.adn7095 (2024)
15. Zuo, Y. et al. Controlled delivery of a neurotransmitter-agonist conjugate for functional recovery after severe spinal cord injury. *Nat Nanotechnol*. **18**, 1230-1240 (2023).
16. Hingorani, S. et al. Transplantation of dorsal root ganglia overexpressing the NaChBac sodium channel improves locomotion after complete SCI. *Mol Ther*. **32**, 1739-1759 (2024).
17. Zhou, K. et al. Dual electrical stimulation at spinal-muscular interface reconstructs spinal sensorimotor circuits after spinal cord injury. *Nat Commun*. **15**, 619 (2024).
18. Herkenham, M. et al. Cannabinoid receptor localization in brain. *Proc Natl Acad Sci U S A*. **87**, 1932-1936 (1990).
19. Herkenham, M. et al. Characterization and localization of cannabinoid receptors in rat brain: a quantitative in vitro autoradiographic study. *J Neurosci*. **11**, 563-583 (1991).
20. El Manira, A., Kyriakatos, A., Nanou, E., Mahmood, R. Endocannabinoid signaling in the spinal locomotor circuitry. *Brain Res Rev*. **57**, 29-36 (2008).
21. Mendizabal-Zubiaga, J. Cannabinoid CB1 Receptors are localized in striated muscle mitochondria and regulate mitochondrial respiration. *Front Physiol*. **7**, 476; 10.3389/fphys.2016.00476 (2016).

22. Freund, T. F., Katona, I., Piomelli, D. Role of endogenous cannabinoids in synaptic signaling. *Physiol Rev*. **83**, 1017-1066 (2003).
23. Heifets, B. D., Castillo, P. E. Endocannabinoid signaling and long-term synaptic plasticity. *Annu Rev Physiol*. **71**, 283-306 (2009).
24. Kano, M., Ohno-Shosaku, T., Hashimotodani, Y., Uchigashima, M., Watanabe, M. Endocannabinoid-mediated control of synaptic transmission. *Physiological Rev* **89**, 309–380 (2009).
25. Baker, D. et al. Cannabinoids control spasticity and tremor in a multiple sclerosis model. Nat. **404**, 84-87 (2000).
26. Fernández-Ruiz, J. The endocannabinoid system as a target for the treatment of motor dysfunction. *Br J Pharmacol*. **156**, 1029-1040 (2009).
27. Richardson, J. D., Kilo, S., Hargreaves, K. M. Cannabinoids reduce hyperalgesia and inflammation via interaction with peripheral CB1 receptors. *Pain* **75**, 111-119 (1998).
28. Padwal, R. S., Majumdar, S. R. Drug treatments for obesity: orlistat, sibutramine, and rimonabant. *Lancet* **369**, 71-77 (2007).
29. Moreira, F. A., Crippa, J. A. The psychiatric side-effects of rimonabant. *Braz J Psychiatry*. **31**, 145-153 (2009).
30. Oliviero, A. CB1 receptor antagonism/inverse agonism increases motor system excitability in humans. *Eur. Neuropsychopharmacol*. **22**, 27-35 (2012).
31. Christensen, R., Kristensen, P. K., Bartels, E. M., Bliddal, H., Astrup, A. Efficacy and safety of the weight-loss drug rimonabant: a meta-analysis of randomised trials. *Lancet* **370**, 1706-1713 (2007).
32. Chavez-Tapia, N. C. et al. Systematic review and meta-analysis on the adverse events of rimonabant treatment: considerations for its potential use in hepatology. *BMC Gastroenterol*. **9**, 75; 10.1186/1471-230X-9-75 (2009).
33. Fabisiak, A., Włodarczyk, M., Fabisiak, N., Storr, M., Fichna, J. Gastrointestinal Adverse Events of Cannabinoid 1 Receptor Inverse Agonists suggest their Potential Use in Irritable Bowel Syndrome with Constipation: A Systematic Review and Meta-Analysis. *J. Gastrointestin. Liver. Dis*. 28, 473-481 (2019).
34. Nair, A. B., Jacob, S. A simple practice guide for dose conversion between animals and human. *J. Basic. Clin. Pharma.* **7**, 27-31 (2016).
35. JASP Team. JASP (Version 0.19.3) [Computer software] (2025).
36. van Gaal, L., Pi-Sunyer, X., Despres, J.-P., McCarthy, C., Scheen, A. Efficacy and safety of rimonabant for improvement of multiple cardiometabolic risk factors in overweight/obese patients: pooled 1-year data from the Rimonabant in Obesity (RIO) program. *Diabetes Care* **31** (Suppl 2), S229–S240 (2008).
37. Robinson, J.D. Pooled analysis of three randomized, double-blind, placebo controlled trials with rimonabant for smoking cessation. *Addict Biol*. **23**, 291-303 (2018).
38. Hagenbach, U. et al. The treatment of spasticity with Delta9-tetrahydrocannabinol in persons with spinal cord injury. *Spinal Cord* **45**, 551-562 (2007).
39. Karst, M., Wippermann, S., Ahrens, J. Role of cannabinoids in the treatment of pain and (painful) spasticity. *Drugs* **70**, 2409-38 (2010).

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AUTHOR CONTRIBUTIONS

Conceptualization: MND, VSL, and AO; Methodology: MND, VSL, and AO; Preclinical experiments were designed and performed by MND, RMM, DR, ASN and MABM. Clinical trial design, participant recruitment, clinical assessments, and data collection were conducted by VSL, FGH, CRG, YAPB, SFC, ABC, MABM and AO. Formal Analysis: MND, VSL and AO. Data Curation: MND, VSL, MABM and AO.; Resources and funding Acquisition: MND, VSL, and AO; Project Administration: MND, VSL, MABM, and AO. Supervision: MND, VSL and AO.; Visualization: MND and VSL; Writing – Original Draft: MND, VSL and AO.; Writing – Review & Editing: All authors. All authors critically reviewed the manuscript, approved the final version, and agreed to be accountable for all aspects of the work.

DATA AVAILABILITY STATEMENT

The preclinical datasets generated and analyzed during the current study are provided as Supplementary Material, together with detailed descriptions of the methodology and results of the corresponding analyses. A summary of the Investigational Plan of the RIMOFATSCI-1 clinical trial is also available as in the Supplementary Material. The clinical trial results, including individual de-identified participant data underlying the results reported in this article, are available from Dr. Antonio Oliviero (antonioo@sescam.jccm.es) upon reasonable request and subject to a data-sharing agreement and signing of a Non-Disclosure Agreement (NDA). The Clinical Study Report (CSR) has been submitted to the AEMPS/EMA.

CONFLICTS OF INTEREST

The Fundación Hospital Nacional de Parapléjicos is the owner of a patent family related to the use of Rimonabant for neurological disorders. Dr. Antonio Oliviero is the owner of Samos Medical Enterprise SL, which holds the rights (license) to exploit patents related to the use of Rimonabant for neurological disorders. Samos Medical Enterprise SL (Dr. Antonio Oliviero), Dr. Vanesa Soto León, and Dr. Manuel Nieto Díaz are co-founders of Sinfatin SL, a company focused on the development of Rimonabant for the treatment of neurological diseases.

TABLES

Table 1

Event Type	Exp 1	Exp 2	Exp 3	Total
Number of animals	5	5	5	15
Animals with any AE (%)	4 (80%)	2 (40%)	4 (80%)	10 (66%)
Severe AEs (%)	1 (20%)	1 (20%)	1 (20%)	3 (20%)
Treatment-related AEs (%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Withdrawals due to AE (%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Event Counts	**Exp 1**	**Exp 2**	**Exp 3**	**Total**
Urinary tract infections	3	2	4	9
ed abdomen	0	0	4	4
Skin ulcers	3	1	3	7
Pain	1	1	1	3
Total AEs	7	4	12	23

Table 1. Summary of Adverse Events recorded across preclinical experiments. AE=adverse event; Exp: experiment.

582 **Table 2**

Overall incidence of AEs

Treatment	Subjects with AE (%)	Total AEs	Related AEs (n)
Placebo	2 (25%)	3	0
2.5 mg/day	4 (50%)	6	3
5 mg/day	4 (50%)	5	3
Total	6 (75%)	14	6

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AEs Possibly Related to the Study Medication

System Organ Classes (SOCs)	Adverse Event	No. of AEs	Dose (mg/day)
Nervous system disorders	Spasticity	1	2.5
Musculoskeletal and connective tissue disorders	Muscle spasms	1	2.5
Gastrointestinal disorders	Mouth ulceration	1	2.5
Gastrointestinal disorders	Abdominal pain	3	5
Gastrointestinal disorders	Nausea	4	5
Nervous system disorders	Headache	1	5

584 **Table 2. Summary of the Adverse Events recorded in RIMOFATSCI-1 clinical trial.**

FIGURES

Figure 1

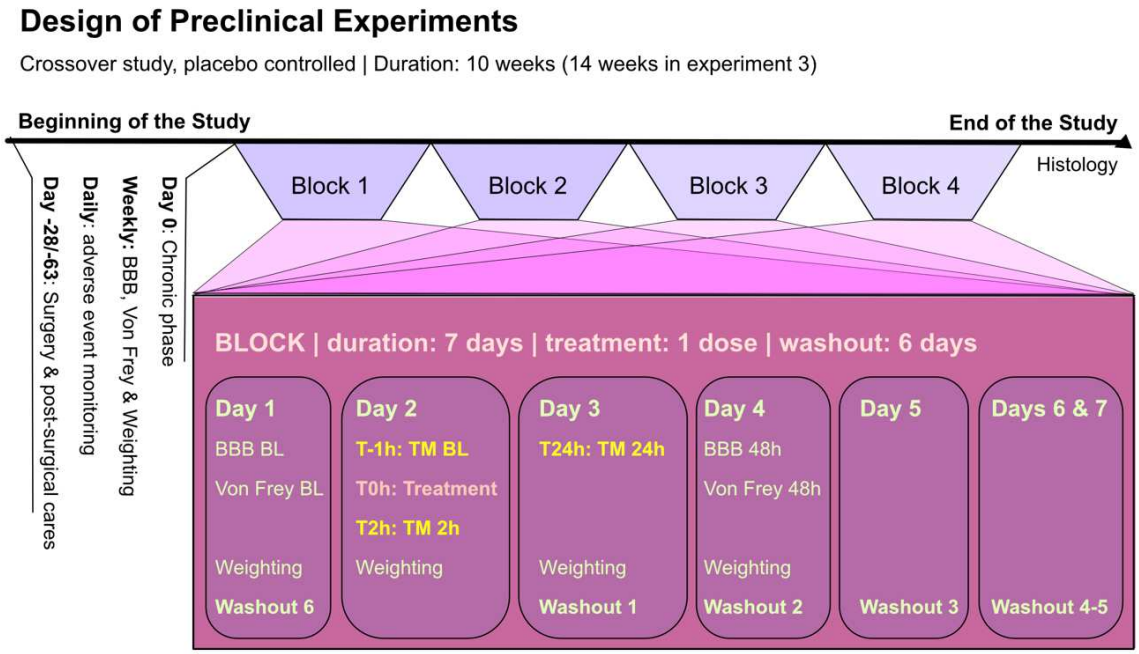


Figure 1. General design of the placebo-controlled, single-doses, crossover, preclinical analyses. The study consisted of four 7-day treatment blocks (placebo plus three Rimonabant doses). Each block began with baseline assessment of motor and sensory function on Day 1. Baseline endurance was evaluated on Day 2, followed by administration of placebo or Rimonabant and reassessment of endurance 2 h later. Endurance was reevaluated 24 h after treatment (Day 3), whereas BBB and Von Frey assessments were repeated 48 h after treatment (Day 4). Washout periods lasted 6 days (Days 3–7 of each block plus Day 1 of the subsequent block). Surgeries were performed 28 days before baseline measurements in Experiments 1 and 2, and 63 days before baseline measurements in Experiment 3, to ensure establishment of chronic SCI. At the end of Block 4, all animals were euthanized and spinal cord tissue was collected for histological analysis. The daily timeline for each experiment is detailed in *Supplementary Methods 1*.

Figure 2

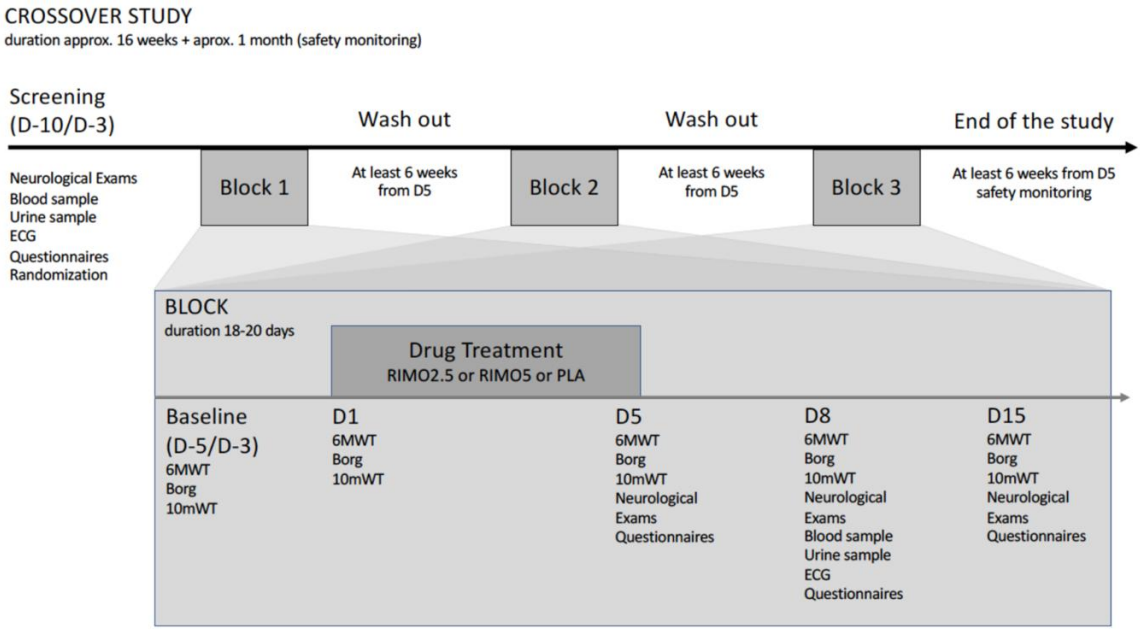


Figure 2. Study Design and Visits. The study was structured in three 12-week blocks (placebo, 2.5 mg/day, 5 mg/day). Each block comprised five visits. V1–V2: Phone screening and in-person screening (consent and eligibility; one subject excluded); V3/V8/V13: Baseline assessments; V4/V9/V14: Treatment start (Day 1); V5/V10/V15: End of treatment (Day 5); V6/V11/V16 and V7/V12/V17: Follow-up at 3 and 15 (±2) days post-treatment. V18: Final study visit approximately 30 days after block 3. Each day, participants received two tablets: 5 mg/day dose: two tablets of Rimonabant (2.5 mg each); 2.5 mg/day dose: one Rimonabant + one placebo; Placebo dose: two placebo tablets. Placebo and Rimonabant tablets were identical in appearance. Vital signs were obtained at each visit.

Figure 3

A. Summary statistics

	ΔE 2h					ΔE 24h				
	Placebo	0.062	0.125	0.250	0.800	Placebo	0.062	0.125	0.250	0.800
Valid	14	5	13	13	10	13	5	14	14	9
Missing	1	0	2	2	0	2	0	1	1	1
Mean	-2.997	-2.870	-1.872	-1.828	-5.142	0.386	2.096	3.641	3.758	4.042
Std. Deviation	5.981	8.370	3.178	5.806	5.472	3.368	4.238	5.966	6.091	5.121
Minimum	-17.740	-11.980	-9.930	-15.190	-13.480	-5.280	-3.120	-4.190	-5.790	-0.590
Maximum	2.920	9.900	0.760	8.090	3.290	7.320	7.990	16.030	13.700	13.290

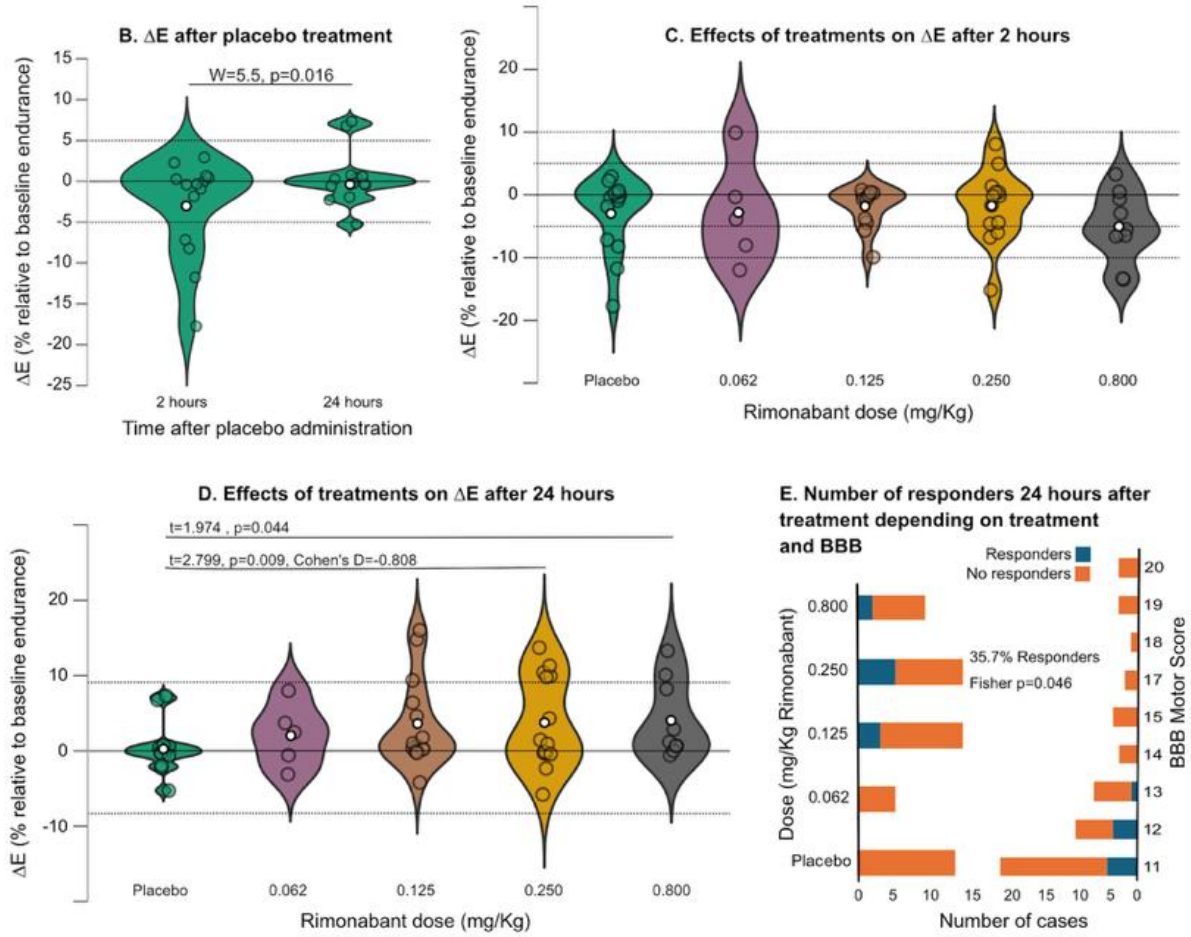


Figure 3. Efficacy of low doses of Rimonabant on increased endurance. Changes in endurance relative to baseline (ΔE) were determined 2 and 24 hours after treatment with single doses of Rimonabant or placebo. **A.** Summary statistics. **B**. Violin plots showing the distribution of ΔE in placebo-treated animals at 2 and 24 hours. W denotes the statistic from a paired Wilcoxon signed-rank test comparing ΔE between the two times. n = 12 (3 individuals were excluded). **C-D**. Violin plots illustrating the effects of treatments on ΔE at 2 hours (C) and 24 hours (D). In D, t denotes the statistic from paired t-tests comparing ΔE between placebo and 0.250 mg/kg and 0.800 mg/kg Rimonabant, 24 hours after treatment (n = 9 paired comparisons). In B–D, white circles indicate the mean value for each condition. **E**. Bar plots comparing the number of responders and non-responders according to treatment and BBB motor score. Fisher's p denotes the significance level after Fisher Exact Test comparing ΔE between placebo and 0.250 mg/kg Rimonabant.

Figure 4

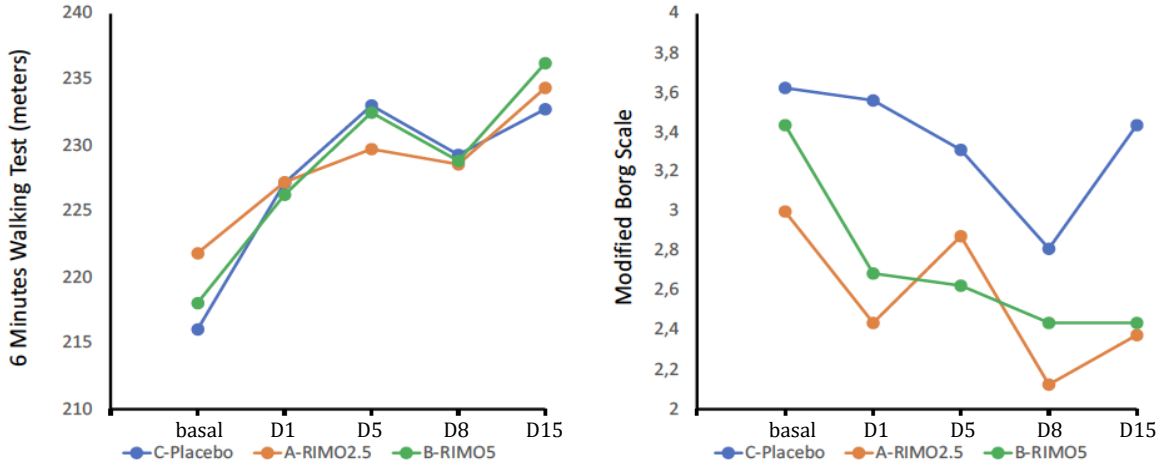


Figure 4. Effects of Rimonabant on 6MWT performance. Left. 6MWT Performance Over Time. Groups: A = 2.5 mg, B = 5 mg, C = Placebo. Error bars are not displayed for graphical reasons. The SEM was in a range from 60 to 69. **Right. Modified Borg Scale Over Time**. Groups: A = 2.5 mg, B = 5 mg, C = Placebo. Error bars are not displayed for graphical reasons. The SEM was in a range from 0.77 to 1.41. N=8.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- NIETORimonabantSuppMatvs2.pdf