The effect of cannabinoids on the stretch reflex in multiple sclerosis spasticity

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The aim of this observational study was to assess the efficacy of a tetrahydrocannabinol–cannabidiol (THC : CBD) oromucosal spray on spasticity using the stretch reflex in patients with multiple sclerosis (MS). Numeric rating scale (NRS) for spasticity, modified Ashworth scale (MAS), and the stretch reflex were assessed before and during treatment in 57 MS patients with spasticity eligible for THC : CBD treatment. A significant reduction in stretch reflex amplitude as well as significant reductions of NRS and MAS scores were observed. There was a low concordance between the three measures (stretch reflex, NRS, and MAS), likely related to the different aspects of muscle hypertonia assessed. Stretch reflex responders were taking a significantly higher number of puffs, whereas no differences were found in the responders by the other scales, suggesting that a higher dosage would add benefit if tolerated. The present study confirms the efficacy of cannabinoids in reducing spasticity in patients with MS, suggesting a higher sensitivity and specificity of the stretch reflex compared with other measures. As an objective and quantitative measure of spasticity, the stretch reflex is particularly useful to assess the effects of cannabinoids on spinal excitability and may play a role in future pharmacological studies. *Int Clin Psychopharmacol* 31:232–239 Copyright © 2016 Wolters Kluwer Health, Inc. All rights reserved.

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Introduction

Spasticity may affect up to 80% of patients with multiple sclerosis (MS), determining disability in at least one-third of cases, as shown by an epidemiological survey based on questionnaires (Rizzo *et al.*, 2004). Spasticity is often associated with a variety of symptoms including fatigue, pain, and the presence of muscle spasms. In patients with MS, the use of drugs such as baclofen, dantrolene, and tizanidine is difficult because of the side effects (drowsiness, weakness, dizziness) increasing the burden of the pre-existing symptoms related to the disease. The tetrahydrocannabinol : cannabidiol (THC : CBD) oromucosal spray (USAN name: nabiximols, trademark: Sativex) is a combination of delta-9-tetrahydrocannabinol and cannabidiol administered by oral puffs and adsorbed through a transmucosal route, active on CB1 and CB2 cannabinoid receptors. Large studies showed its efficacy in relieving symptoms related to spasticity in MS patients showing no significant benefit from other antispastic drugs (Zajicek *et al.*, 2003; Collin *et al.*, 2007; Wade *et al.*, 2010; Flachenecker *et al.*, 2014; Paolicelli *et al.*, 2015).

Spasticity is traditionally defined as a motor disorder characterized by an exaggeration of the stretch reflex (Lance, 1980), resulting in a velocity-dependent increase in muscle tone during passive limb movements. From a clinical point of view, the examiner perceives a resistance that can be measured semi-quantitatively using scales such as the modified Ashworth scale (MAS), one of the most widely used scales. Although the neurophysiological recording of the stretch reflex would be the ideal parameter to quantitatively assess spasticity, its use is limited in a clinical setting. Most of the studies involving the stretch reflex measurement in fact use robotic devices that are expensive, bulky, and often designed for a single joint. In many studies, spasticity was assessed using clinical scales often based on the patient’s own experience. In particular, the numeric rating scale (NRS) (Farrar *et al.*, 2008; Anwar and Barnes, 2009), the MSSS-88 (Hobart *et al.*, 2007; Wade et al., 2009), the MSSSS-88 (Hobart *et al.*, 2006), and the spasms frequency scale (Collin *et al.*, 2007; Farrar *et al.*, 2008) are basically subjective measures directly provided by the patients.
The assessment of spasticity directly performed by a clinician usually includes MAS, which, although related to the NRS (Anwar and Barnes, 2009), is very imprecise and operator dependent (Collin et al., 2007; Thaera et al., 2009) and not always correlates with the stretch reflex amplitude, especially with respect to the intermediate scores (Damiano et al., 2002). Indeed, in most of the studies assessing the effect of cannabinoids on spasticity, MAS failed to show a significant variation, probably because of its intrinsic limitations and lack of sensitivity (Killestein et al., 2002; Zajicek et al., 2003; Collin et al., 2007; Thaera et al., 2009). Conversely, NRS appeared to be much more effective in detecting subjective changes in spasticity, becoming the gold standard in large pharmacological trials on the effect of cannabinoids and specifically THC : CBD (Zajicek et al., 2003; Collin et al., 2007). Nonetheless, NRS remains a subjective estimation of a phenomenon with a precise neurophysiological characterization on the basis of the patient’s understanding and often including other clinical expressions of upper motor neuron damage. In this view, many studies advise the need for spasticity assessment tools that are sensitive and validated (Zajicek et al., 2003; Collin et al., 2007; Anwar and Barnes, 2009; Thaera et al., 2009).

Neurophysiological methods suitable to measure the stretch reflex have been underused in clinical research. Our group has recently validated a method to elicit and measure the stretch reflex in a clinical setting with the use of surface electromyography (EMG) and without robotic devices (Marinelli et al., 2013). The method is effective on major joints (wrist, elbow, knee, and ankle) and enables a reproducible and quantitative evaluation of the stretch reflex in patients with spasticity.

The present study measures the effect of THC : CBD in spastic MS patients using the stretch reflex and compares it with other spasticity scales.

Patients and methods

Patients

We recruited 57 consecutive MS patients with spasticity, eligible for THC : CBD treatment in the Department of Neuroscience, Rehabilitation, Ophthalmology, Genetics, Maternal and Child Health, of the University of Genova, Italy. An unrestricted grant from Almirall S.A. was obtained to fund the study after submission of the project protocol and evaluation of an experts board external to the funder. The patients whom we consecutively recruited had already been prescribed THC : CBD and were ready to start the treatment; thus, we had no time to organize a randomized-controlled study. We therefore decided to parallel the standard clinical assessment with the stretch reflex evaluation limiting to an exploratory observational study.

Patients were 26 (46%) men and 31 (54%) women, mean age was 52 years (range: 27–79 years), mean expanded disability status scale (EDSS) was 6.9 (range: 5–8), and mean disease duration was 194 months (range: 48–456 months). MS type was primary progressive in 11 (19%), secondary progressive in 43 (76%), and relapsing-remitting in three (5%). The inclusion criteria were as follows: (i) presence of spasticity with MAS lower than 4 in at least one of the following muscle groups: flexor muscles of the wrist, flexor muscles of the forearm, extensor muscles of the leg, foot plantiflexors, (ii) absence of significant peripheral nervous system pathology detectable on clinical basis, (iii) absence of concomitant parkinsonism, (iv) no exposure to oral or smoked cannabinoids in the 30 days before starting THC : CBD, (v) no botulinum toxin injections and no dosage variation of other drugs potentially affecting spasticity and pain in the 30 days before starting THC : CBD, and (vi) nabiximols approved label requirements.

No limitations related to age and degree of disability were applied for patient selection. The patients with questionable spasticity were rated 0 at the MAS and included only if a stretch reflex was well recognizable at angular velocities less than 180° (Thilmann et al., 1991).

The following sociodemographic data were collected: sex, age, years with MS, and muscles affected by spasticity. The present study was carried out in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans; a written informed consent was obtained from all participants. The study was approved by the local ethics committee.

Experimental procedure

Preliminary setting and clinical evaluation

The patients were evaluated in a quiet room with a temperature between 21 and 23°C. The muscle group with the highest level of spasticity but preserved range of motion (MAS < 4) was selected among flexor muscles of the wrist, flexor muscles of the forearm, extensor muscles of the leg, or foot plantiflexors. All patients underwent a complete neurological examination and the range of motion of the selected joint was determined along with MAS. To assess the stretch reflex, the patients rested in a seated position with the arms leaning on a pillow for flexor muscles of the wrist or flexor muscles of the forearm, whereas for the extensor muscles of the leg, the patients were lying supine on a comfortable examination table with the head and shoulders slightly elevated, both legs over the end of the couch. To assess foot plantiflexors, the patients were lying prone with the feet protruding from the examination table to enable a full ankle range of movement.

The subjective perception of spasticity was assessed using NRS for spasticity, along with visual analogue scale (VAS) for pain. All measures were collected by medical
doctors who specialized in neurology or rehabilitative medicine; in each patient, the same physician performed the stretch reflex procedure before and during the treatment with THC: CBD.

**Stretch reflex technical setup**
With the patients in the same position described above, the examiner perceived the tones produced by a software emulated metronome through headphones and was seated near the patient to move the chosen limb comfortably. The EMG activity produced by the stretched muscle was recorded by surface preamplified electrodes with a fixed 20 mm interelectrode distance (TSD150B; Biopac Systems Inc., Goleta, California, USA) placed over the muscle belly following SENIAM guidelines (SENIAM, 2016) and acquired by a Biopac MP150 Unit (Biopac Systems Inc.). Joint angle during the movements was measured during movements determining elongation (SENIAM, 2016) and acquired by a Biopac MP150 Unit connected to the same MP150 Unit.

**Stretch reflex procedure**
The method has been described in detail in a previous validating work (Marinelli et al., 2013). During the procedure, the patients were instructed to remain relaxed and to avoid resisting or facilitating the movements performed by the examiner. Initially, the optimal audio tone frequency was decided and set on the metronome: for instance, when choosing a frequency of 60 beats per minute (BPM), the interval between consecutive tones is 1 s. As the examiner is required to move the limb throughout the full range of motion in the time corresponding to the interval between two consecutive metronome tones, the movement velocity increases linearly with BPM. The choice of the BPM was made taking into account the fact that low values may not elicit a stretch reflex (especially in patients with a low degree of spasticity), whereas high values could produce discomfort to the patient and excessive fatigue in the examiner. Once the optimal BPM were set, the examiner started performing consecutive flexion and extension movements following the rhythm reaching the extreme limb positions in synchrony with consecutive metronome tones. Performing about five of these continuous (or ‘sinusal’) movements enabled the examiner to learn the appropriate velocity and therefore perform the movements also interposing a few tones interval between the movements, thus obtaining discontinuous (or ‘linear’) movements. The procedure was repeated to obtain 40 flexion and 40 extension movements. The stretch reflex was measured during movements determining elongation of the spastic muscle. The procedure lasted about 15 min and was performed on a single muscle group.

**Tetrahydrocannabinol: cannabidiol effect evaluation**
The stretch reflex procedure and the clinical evaluation were performed twice: at the baseline condition (T0) before starting the treatment and 4 weeks later, the medication label required a trial period, when the optimal THC: CBD dose was reached (T1). During this trial period, the number of puffs was titrated according to the medication label scheme, adding one or two puffs each day up to the highest tolerated number (no more than 12 puffs in a day; five in the morning and seven in the afternoon or evening). Between T0 and T1, the patients were asked to maintain concomitant medications unchanged. During T1 evaluation, the number of sprays/day and the possible adverse events were also recorded. To obtain a reproducible electrode positioning between T0 and T1, a picture of the electrode and its relation with nearby anatomical landmarks was taken in each patient. To reduce all possible sources of variability, the entire setup was the same at T0 and T1, including the patient’s position, the examiner, and obviously the metronome BPM to obtain reproducible velocity (Marinelli et al., 2013).

**Data analysis**
The main endpoint of the study is the reduction of spasticity assessed with the stretch reflex after 4 weeks of treatment with THC: CBD at the maximal tolerated individual dose. To this aim, the EMG recordings were filtered, rectified, and the mean amplitude (‘mean function of the analysis software) of the bursts during lengthening duration was calculated and defined as ‘meanEMG’ (Marinelli et al., 2013) using the dedicated AcqKnowledge Analysis Software (Biopac Systems Inc.). The baseline meanEMG values are highly variable among patients because of many factors such as thickness of subcutaneous tissue, muscle trophism, level of spasticity, etc. A normalization procedure using a parameter such as maximal M wave amplitude following nerve stimulation and maximal voluntary contraction would be indicated; however, both appeared unfeasible. In fact, the majority of MS patients showed spasticity in proximal limb muscles (such as biceps brachii and quadriceps femorii), where nerve stimulation is difficult to perform and would have caused considerable discomfort. However, maximal voluntary contraction could not be performed because of variable muscle weakness associated with spasticity. To measure the meanEMG variation between T0 and T1, we therefore compared the average meanEMG using a paired t-test.

We also distinguished between responders and non-responders to the treatment as assessed by the neurophysiological procedure. To this aim, in each patient, an unpaired t-test was performed between the 40 meanEMG values at T0 and T1: if the meanEMG values at T1 were significantly lower (P < 0.05) compared with T0, the patient was considered a responder.

As secondary endpoints, NRS for spasticity, MAS, and VAS for pain were compared between T0 and T1 using the Wilcoxon test for semiquantitative data. The
numbers of responders for NRS and MAS were also calculated as the number of patients with an NRS reduction of at least 20% (Farrar et al., 2008) and an MAS reduction of at least one point at $T_1$. For the purpose of analyses, MAS values were transformed to obtain values ranging from 0 to 6 (1+ becomes 2, 2 becomes 3, etc.) (Anwar and Barnes, 2009). Transformed MAS values are reported in the results section and in the figures. The numbers of THC:CBD puffs between responders and nonresponders were compared using an unpaired $t$-test.

The sponsor of this study played no role in the study design, data collection, data analysis, data interpretation, and writing of the report, providing support only for English language revision.

Results

Five of the 57 recruited patients could not perform the stretch recording at $T_0$ because of discomfort related to the position required to perform the evaluation. Out of the remaining 52 patients, 15 (26%) stopped THC:CBD before the re-evaluation 4 weeks later ($T_1$) because of side effects, mostly dizziness, sleepiness, and nausea. One patient could not remain relaxed and could not be re-evaluated. Therefore, stretch recordings were collected both at $T_0$ and $T_1$ in 36 patients (mean age: 54 years, range: 31–79 years, 15 men) (Table 1); the remaining patients were excluded from the analysis also for non-neurophysiologic measures. Five patients were tested on flexor carpi radialis, six patients on biceps brachii, one patient on triceps brachii, and 24 patients on quadriceps, with metronome pacing ranging between 40 and 120 BPM.

A reproducible stretch reflex could be elicited in all patients (Fig. 1). Despite the high variability of baseline meanEMG values among patients, related to the lack of normalization (see the Patients and methods section), the meanEMG reduction at $T_1$ in the 36 patients using a paired $t$-test was indeed statistically significant ($P = 0.026$). On comparing the single stretch in each patient, a significant reduction in the meanEMG was detected in 20 out of 36 (56%) patients, defined here as ‘neurophysiological responders’ (Fig. 2). No significant difference was found in eight (22%) patients and a significant increase was found in another eight (22%) patients.

Baseline spasticity in the selected muscle ranged between 0 and 4 (mean ± SD: 2.4 ± 1.2, MAS transformed values) and decreased significantly ($P = 0.0012$) at $T_1$ (1.8 ± 1.4). Two patients (#3 and #19) had questionable spasticity at $T_0$ scoring 0 at the MAS, but showed a clearly recognizable stretch reflex; these two patients were excluded from MAS analysis. The number of responders considering MAS (clinical responders) was 15 out of 34 (44%) patients (Fig. 2) (3 points reduction in two patients, 2 points in four patients, 1 point in nine patients); in 18 patients, MAS was unaltered, whereas in one patient, MAS increased by 1 point.

All 36 patients scored at least 3 on NRS for spasticity at $T_0$. On average, NRS for spasticity was 6.8 ± 1.7 at $T_0$ and 5.7 ± 2.1 at $T_1$, consistent with a statistically significant decrease ($P = 0.0007$). NRS for spasticity showed a reduction of at least 20% in 14 (39%) patients (subjective responders) (Fig. 2), whereas in another nine patients, the reduction was lower than 20%. In 11 patients, the NRS was unchanged and in two patients, it increased by at least 1 point.

In the first nine recruited patients, VAS for pain was not collected. In the remaining 27 patients, VAS for pain was reduced by at least 1 point in 12 (44%) patients, whereas it remained unchanged in the other 15 patients (seven had no pain at baseline, VAS = 0). No patients reported increase in pain at $T_1$. In 10 of the 12 (83%) patients, pain was reduced by at least 20%. In the 27 patients analyzed, the mean values at $T_0$ were 3.9 ± 3.1 and 2.9 ± 2.6 at $T_1$, VAS for pain reduction was statistically significant ($P = 0.0048$).

The number of neurophysiological responders ($n = 20$; 56%) was higher than subjective (NRS) ($n = 14$; 39%) and clinical (MAS) responders ($n = 15$; 44%). This observation, along with the presence of a stretch reflex also in two patients who had no spasticity detectable with MAS at baseline, suggests a higher sensitivity of the neurophysiological measure compared with the clinical assessment. We found only a partial agreement in the definition of responders using the three methods: comparing two methods at a time, the sum of responders (white background) and nonresponders (black background) in both methods ranged between 53 and 61% of the patients (Fig. 2).

To further assess the effect of THC:CBD on spasticity, we compared the number of puffs between responders and nonresponders using subjective, clinical, and neurophysiological measures. Considering the subjective NRS measure, responders were taking 7.0 ± 2.9 puffs, whereas nonresponders were taking 5.7 ± 2.2 puffs. Considering clinical MAS evaluation instead, responders were taking 6.3 ± 3.1 puffs and nonresponders were taking 6.2 ± 2.1 puffs. Using the neurophysiological stretch reflex assessment, responders were taking 6.9 ± 2.4 puffs and nonresponders were taking 5.4 ± 2.5 puffs. Despite the similar number of THC:CBD puffs in the three groups, the number of puffs was significantly higher in responders versus nonresponders only using the stretch reflex recording ($P = 0.047$), whereas on comparing the number of puffs between NRS responders and nonresponders ($P = 0.07$) and also between MAS responders and nonresponders ($P = 0.45$), no significant difference could be found, although a trend toward significance was present for NRS responders.
Discussion

This is the first study using stretch reflex to assess the effect of cannabinoids on spasticity. A significant reduction in spasticity during treatment was found on assessing stretch reflex amplitude (meanEMG) in 36 patients with MS when the examiner performed passive elongations of the spastic muscle. Each patient was considered a responder when the comparison of 40 stretches indicated a significant reduction during the treatment. This neuropsychological assessment was compared with the other two methods generally used: NRS for spasticity and MAS.

In the 36 patients included in the analysis, the stretch reflex elicited the highest number of responders compared with the other standard procedures; in more than half of the patients, the stretch reflex decreased significantly during THC:CBD treatment. Given the low concordance in responder definition between the three methods, an obvious question arises about what they are measuring and which method is actually measuring spasticity. Interestingly, only the stretch reflex assessment could distinguish between responders and nonresponders, defining two groups where the number of puffs was significantly different, despite the reduced sample size. The use of an objective instrumental neuropsychological measure is particularly interesting when assessing spasticity as the previous works were limited by subjective and clinical scales providing surrogate endpoints lacking objective and quantitative information (Killestein et al., 2002; Zajicek et al., 2003; Wade et al., 2004, 2010; Collin et al., 2007; Farrar et al., 2008; Flachenecker et al., 2014; Paolicelli et al., 2015). The present study shows that assessment of spasticity with the described stretch reflex procedure is feasible, not particularly time consuming, and well tolerated by the patients. Most importantly, the described stretch reflex measure is sensitive and provides quantitative data that closely reflect both the mechanisms underlying spasticity and the assessment modality used during clinical practice (Marinelli et al., 2013).

In our group, eight patients showed a significant increase in the stretch reflex at \( T_1 \). Many reasons could be discussed to explain this finding; however, the most important is that stretch reflex, like other objective

| Patient | Sex | Age | Muscle | Side | BPM | Baseline MAS (6 points) | Baseline spasticity NRS | EDSS | Months with MS | MS type | Treatment |
|---------|-----|-----|--------|------|-----|------------------------|-------------------------|------|---------------|---------|-----------|
| 1       | F   | 60  | QF     | L    | 120 | 3                      | 8                       | 6.5  | 242           | SP      | Gabapentin, amitryptilin |
| 2       | M   | 48  | FCR    | R    | 100 | 2                      | 7                       | 6    | 160           | PP      | None      |
| 3       | M   | 64  | BB     | R    | 120 | 0                      | 2                       | 8    | 240           | SP      | Backlofen |
| 4       | F   | 68  | BB     | L    | 100 | 2                      | 6                       | 7.5  | 252           | SP      | Venlafaxine, trazodone   |
| 5       | F   | 45  | QF     | R    | 40  | 4                      | 8                       | 6.5  | 228           | PP      | None      |
| 6       | F   | 49  | QF     | R    | 40  | 3                      | 6.5                     | 6.5  | 158           | SP      | Backlofen |
| 7       | F   | 45  | QF     | L    | 120 | 3                      | 7                       | 7    | 180           | SP      | None      |
| 8       | F   | 66  | FCR    | L    | 120 | 2                      | 7                       | 7    | 144           | SP      | Backlofen |
| 9       | M   | 60  | QF     | R    | 120 | 1                      | 5                       | 6.5  | 276           | SP      | None      |
| 10      | F   | 48  | QF     | R    | 60  | 4                      | 7                       | 6.5  | 120           | SP      | Backlofen |
| 11      | M   | 57  | QF     | L    | 120 | 2                      | 7                       | 6    | 268           | PP      | None      |
| 12      | F   | 52  | QF     | L    | 120 | 1                      | 6                       | 6.5  | 94            | SP      | Baclofen, escitalopram  |
| 13      | F   | 42  | QF     | R    | 60  | 3                      | 6                       | 7.5  | 96            | SP      | Tizanidine |
| 14      | F   | 64  | FCR    | R    | 120 | 2                      | 8                       | 7.5  | 186           | SP      | Baclofen, amitryptilin, bromazepam |
| 15      | F   | 49  | QF     | L    | 120 | 3                      | 9                       | 6.5  | 52            | SP      | 4-Aminopyridine, baclofen, duloxetine, pregabalin |
| 16      | M   | 40  | QF     | L    | 120 | 3                      | 6                       | 7    | 210           | SP      | Prednisone |
| 17      | M   | 42  | QF     | L    | 60  | 2                      | 8                       | 6.5  | 260           | SP      | Baclofen, modafinil     |
| 18      | F   | 67  | FCR    | L    | 120 | 1                      | 2                       | 8    | 180           | SP      | Pregabalin |
| 19      | F   | 80  | QF     | L    | 120 | 0                      | 4                       | 7    | 106           | SP      | Gabapentin |
| 20      | M   | 60  | QF     | R    | 90  | 4                      | 8                       | 7    | 168           | SP      | None      |
| 21      | F   | 50  | QF     | L    | 120 | 1                      | 6                       | 6    | 288           | SP      | None      |
| 22      | M   | 56  | QF     | R    | 40  | 4                      | 7                       | 6.5  | 306           | SP      | None      |
| 23      | M   | 43  | QF     | L    | 90  | 3                      | 6                       | 6.5  | 48            | SP      | None      |
| 24      | M   | 53  | BB     | L    | 120 | 2                      | 9                       | 6.5  | 48            | PP      | Naltrexone |
| 25      | F   | 50  | QF     | R    | 120 | 3                      | 8                       | 6    | 94            | SP      | None      |
| 26      | M   | 47  | TB     | R    | 60  | 3                      | 7                       | 7.5  | 212           | SP      | Baclofen, clonazepam   |
| 27      | M   | 74  | QF     | L    | 60  | 2                      | 5                       | 6.5  | 48            | SP      | Baclofen |
| 28      | F   | 53  | QF     | R    | 40  | 4                      | 7                       | 6.5  | 456           | SP      | Baclofen |
| 29      | F   | 53  | QF     | L    | 120 | 4                      | 9                       | 7    | 214           | SP      | Backlofen |
| 30      | F   | 67  | QF     | L    | 60  | 3                      | 7                       | 7.5  | 254           | SP      | Fentanyl, sertraline   |
| 31      | F   | 79  | BB     | L    | 60  | 4                      | 8                       | 7.5  | 132           | PP      | Diazepam |
| 32      | F   | 31  | QF     | R    | 120 | 1                      | 7                       | 6.5  | 72            | RR      | None      |
| 33      | M   | 44  | FCR    | R    | 120 | 3                      | 7                       | 7    | 220           | SP      | None      |
| 34      | M   | 53  | QF     | R    | 120 | 1                      | 6                       | 6.5  | 180           | SP      | None      |
| 35      | F   | 50  | BB     | L    | 120 | 2                      | 10                      | 7    | 154           | SP      | Baclofen |
| 36      | M   | 47  | BB     | R    | 100 | 2                      | 8                       | 7.5  | 180           | PP      | 4-Aminopyridine |

BB, biceps brachii; BPM, beats per minute; EDSS, expanded disability status scale; FCR, flexor carpi radialis; L, left; MAS, modified Ashworth scale; MS, multiple sclerosis; NRS, numeric rating scale; PP, primary progressive; QF, quadriceps femoris; R, right; RR, relapsing-remitting; SP, secondary progressive; TB, triceps brachii.

Table 1 Demographic and clinical features of the 36 patients included in the analysis. In the treatment column, only drugs potentially affecting spasticity and pain have been reported.
measures such as MAS, takes a snapshot of the situation in a specific moment, without the possibility of an average estimation over a larger time span. If, during the evaluation, the patients for example experience more discomfort, pain, or cold, spasticity might temporarily increase and sensitive tools such as the stretch reflex can probably detect this increase. Of course, subjective measures such as NRS overcome this intrinsic limitation of objective measures.

The poor concordance between subjective, clinical, and neurophysiological responders could be related not specifically to spasticity, but to different phenomena related to upper motor neuron syndrome measured by the three methods. A relevant MAS variability between examiners has often been discussed; moreover, it cannot distinguish between stiffness resulting from reflex muscle contraction (properly considered the equivalent of spasticity) and nonreflex components of muscle hypertonia related to the modification of the intrinsic viscoelastic properties of muscular tissue (such as fibrosis and sarcomeres shortening). The stretch reflex conversely measures the electric activity produced by the contracting muscle in response to a passive stretch and is considered the most specific measure of spasticity (Trompetto et al., 2014). As a secondary endpoint, the present study confirms a significant reduction in the NRS for spasticity during THC:CBD treatment. The NRS for spasticity is a subjective estimation based solely on the individual interpretation of spasticity and is currently considered the gold standard to measure the effects of cannabinoids on spasticity (Wade et al., 2004; Hobart et al., 2006; Collin et al., 2007; Farrar et al., 2008; Anwar and Barnes, 2009). NRS acquisition is easy and quick, taking into account what spasticity means for the patient and the impact on daily life over a 24 h time span (Anwar and Barnes, 2009). A good test–retest reliability has been shown (Farrar et al., 2008; Anwar and Barnes, 2009); however, keeping in mind that spasticity is an exaggeration of the stretch reflex (Lance, 1980), the use of NRS raises certain perplexity. Given an acceptable patient’s understanding of spasticity as an ‘increased stiffness in the limbs’, the patient probably cannot assess the amount of stiffness in his/her own paretic and reasonably hypoesthetic limbs. During discussions with the patients, our perception is that at least some patients misunderstand the concept of ‘stiffness’ with those of ‘decreased strength’. Indeed, NRS for spasticity is strongly related to the motricity index: patients who perceive higher spasticity actually have more objective difficulties in performing movements (Anwar and Barnes, 2009). The important relation of NRS for spasticity with the subjective measure of other symptoms related to spasticity such as the Spasms Frequency Scale and the Patient Global Impression of Change may support the reliability and validity of NRS (Farrar et al., 2008), but also underlines its limited...
The beneficial effect of cannabinoids on pain has been known for millennia and previous studies in patients with MS reported a significant effect of nabilimol on pain (Zajicek et al., 2003), even though a relevant placebo effect was also detected (Wade et al., 2004). In our case series, VAS for pain was significantly reduced during treatment, even though this finding has limited relevance considering that VAS for pain was available only in 27 of the analyzed patients and seven patients had no pain at baseline. The relation between pain and spasticity is, however, very interesting and, in our opinion, deserves further investigation; in fact, spasticity itself may be the cause of pain. However, pain increases the levels of spasticity (Trompetto et al., 2014).

The neurophysiological responders were taking more THC : CBD puffs compared with the nonresponders. It is likely that responders benefit from higher THC : CBD levels in the central nervous system, with a higher beneficial effect on spasticity than nonresponders, who were taking fewer puffs. This could be an indirect demonstration that THC : CBD is effective in reducing spasticity in MS and we may also hypothesize that the threshold between responders and nonresponders is related to the extent of side effects induced by the drug and therefore the number of puffs that each patient is able to take. For example, a nonresponder might become a responder if he/she is able to take and tolerate a higher number of puffs. An interesting outcome is that if side effects were fewer, probably more patients could be able to take more puffs and may become responders. The concept of ‘responders’ could therefore be associated with the capability of tolerating THC : CBD side effects in the context of a pharmacogenetically related response to the drug (Onaivi, 2010).

The majority of patients (23/36) were taking drugs potentially modulating pain and spasticity, in most cases baclofen, which were maintained stable during the study. Most of the associated drugs such as baclofen and diazepam can indeed modulate GABA transmission and thus potentially interact with the study drug. To our knowledge, only animal studies are currently available (Méndez-Díaz et al., 2013). Considering the high variability of the stretch reflex among patients, we believe that the effects of an interaction should be negligible in this study. However, we believe that this issue deserves specific studies in humans.

Apart from stretch reflexes, other neurophysiological parameters have been used to assess the effect of cannabinoids on spasticity. Spinal cord excitability using the ratio between H-reflex and M-wave amplitudes (H/M ratio) showed no modification following THC : CBD treatment in a group of 20 patients with MS (Centonze et al., 2009). The H/M ratio, however, measures only some of the mechanisms underlying spasticity, estimating spinal cord neural circuits; excitability by electrical

specificity. Some authors discussed that other symptoms such as pain and fatigue might influence the perception, as well as obvious pitfalls in patients with cognitive impairment and communication difficulties (Anwar and Barnes, 2009). The low agreement between subjective and objective indicators of spasticity must be interpreted keeping in mind that one method is not necessarily less reliable or valid then the other, especially in the short-term follow-up period studied; rather, they measure complementary outcomes that combine together for a thorough understanding of the phenomenon (Hobart et al., 2006).

A significant reduction in MAS scores during THC : CBD treatment is a secondary endpoint of the study. Previous studies did not show a significant reduction in clinically assessed spasticity (using traditional or MAS) both with THC : CBD and with other compounds containing cannabinoids (Killestein et al., 2002; Zajicek et al., 2003; Collin et al., 2007). It is likely that the absence of a placebo arm and the unblinded status of the examiners could have affected this finding. Many studies, however, underline the unreliability of MAS as a measure of spasticity (Collin et al., 2007; Thaera et al., 2009).
stimulating a peripheral nerve. Different from the stretch reflex, this technique does not take into account all the neural systems involving peripheral and central connections that constitute the network where spasticity develops (Lance, 1980). Another study on the effects of cannabinoids on MS spasticity involving neurophysiological measures is actually focused on painful stimulus perception and tremor attenuation following smoked cannabis, but not spasticity itself (Meinck et al., 1989). A more recent double-blind placebo-controlled study on 44 patients with MS did not find modifications of the H/M ratio as well as measures of cortical excitability by means of transcranial magnetic stimulation (Leocani et al., 2015).

The main limitation of this study is related to the lack of a placebo arm. Given the observational and exploratory nature of this study, the results are innovative and introduce an undersized sensitive and specific measure of spasticity, thus supporting the feasibility of future double-blind placebo-controlled studies. The decision to evaluate the stretch reflex only on one muscle group is justified by the need to limit the duration of the assessments and patients’ discomfort. Moreover, we believe that it is unlikely that the drug may affect spasticity differently depending on muscle groups; further studies may properly address this issue.

The stretch reflex recording deserves more attention as it fulfills the urgent need for sensitive and validated spasticity assessment tools. Larger and longer studies would be advisable to track the clinical evolution of stretch reflex responders and nonresponders through time.

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Conflicts of interest
There are no conflicts of interest.

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