

# Increase of the Ejaculatory Potency by the Systemic Administration of Aqueous Crude Extracts of Cihuapatli (*Montanoa* Genus) Plants in Spinal Male Rats

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## Abstract

In the present study, evidence on the aphrodisiac activity of *Montanoa frutescens* and *Montanoa grandiflora* and a comparison with the aphrodisiac activity of *Montanoa tomentosa* is presented. By using the fictive ejaculation model in spinal male rats, electro-myographic recordings of the genital motor pattern of ejaculation were obtained in the bulbospongiosus muscles and analyzed after the intravenous injection of aqueous crude extracts of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora*. Results showed that the systemic administration of the aqueous crude extracts of *Montanoa* plants elicits a significant increase in the ejaculatory capacity of spinal male rats with very robust ejaculatory motor patterns that included the expression of tonic penile erections and penile movements and the potent expulsion of urethral contents. In conclusion, *Montanoa frutescens* and *Montanoa grandiflora* increase the ejaculatory potency with aphrodisiac activity similar to *Montanoa tomentosa*.

## Keywords

*Montanoa tomentosa*, *Montanoa frutescens*, *Montanoa grandiflora*, ejaculation, aphrodisiac plant, male rat

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## Introduction

Ejaculatory dysfunction is considered one of the most prevalent sexual complaints in sexually active men and arises due to disorders in the expression of normal ejaculation.<sup>1</sup> Many pharmaceutical agents including antidepressant agents have been used to control ejaculatory response though these compounds produce considerable side effects.<sup>2</sup> Recent evidence has shown that medicinal plants with aphrodisiac activity are viable alternatives in the management of ejaculatory disorders.<sup>3</sup> Potent natural aphrodisiacs that influence ejaculation include the Mexican plant cihuapatli.<sup>4</sup>

Cihuapatli is the nahuatl name assigned to a group of Mexican medicinal plants (ie, *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora*) of the *Montanoa* genus. These plants have been used for centuries in Mexican traditional medicine as a remedy for reproductive impairments including mood disorders.<sup>5-8</sup> Ancient descriptions of the use of Cihuapatli as a traditional remedy are contained in the *Badianus Codex* or *Libellus de Medicinalibus Indorum Herbis*<sup>9</sup> where botanical determinants, traditional recipes, and prescriptions are provided. Cihuapatli extract was described as a remedy to favor parturition and during the puerperium but the outstanding use was as a contraceptive agent.<sup>10-12</sup>

Experimental studies with laboratory animals have confirmed that plants included in the *Montanoa* genus prepared as aqueous crude extracts or its purified fractions have contraceptive effects that are provoked by inhibition of implantation, cervical dilatation, and uterine bleeding<sup>7,10-12</sup> without influencing the endocrine status. Aqueous crude extracts of Cihuapatli plants do not modify the hematological, blood lipid, protein, and electrolytic status or the function of the liver, kidney, and thyroid gland.<sup>10</sup> A partial mechanism to describe the effects of the Cihuapatli aqueous crude extract on the female reproductive tract suggests an oxytocinergic profile exerted on the peripheral organs<sup>8</sup> and at the level of the central nervous system.<sup>13,14</sup>

Recent studies reported that the extract of Cihuapatli prepared with *Montanoa tomentosa* can cross the blood-brain

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barrier to exert their actions directly on the central nervous system,<sup>13,14</sup> and in the case of copulating male rats, administration of this extract promotes an aphrodisiac effect by improving both sexual motivation and performance.<sup>13</sup> Aphrodisiac effects of Cihuapatli extracts are mainly exerted on the ejaculatory function where a significant reduction in the ejaculatory latency and a noteworthy robustness in the genital motor pattern of ejaculation are observed.<sup>13,14</sup>

Cihuapatli plants are still used today in México, and to obtain the medicinal effect midwives mostly prepare aqueous crude extracts from *Montanoa tomentosa* as the starting material, but this decoction may also be prepared from equal quantities of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora*. Experimental studies on Cihuapatli performed up to date have been made using *Montanoa tomentosa* as the starting material, but evidence shows that extracts prepared from *Montanoa frutescens*, *Montanoa grandiflora*, or even *Montanoa leucantha* elicit similar effects to that obtained by *Montanoa tomentosa* alone,<sup>15</sup> and in some batches a mixture of *Montanoa* plants is more potent. Throughout Mexico, more than 21 varieties of *Montanoa* species have been collected and examined, and 9 of them were screened in an experimental pilot showing that *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* are the more effective.<sup>15</sup>

The present study was intended to evaluate the pro-ejaculatory properties of Cihuapatli aqueous crude extracts prepared from *Montanoa frutescens*, *Montanoa grandiflora*, and *Montanoa tomentosa*. To this aim, we analyzed the effect of intravenous administration of the aqueous crude extracts prepared from these plants in the fictive ejaculation model in spinally transected and anesthetized male rats, which mainly permits the quantitative evaluation of the genital motor pattern of ejaculation and the visualization of penile events accompanying expulsion of seminal material. We hypothesized that the aqueous extracts prepared from *Montanoa frutescens* and *Montanoa grandiflora* exhibit pro-ejaculatory activity similar to that evoked by the *Montanoa tomentosa* extract.

## Methods

### Animals

Sexually experienced male Wistar rats (300-350 g body weight) were used. Animals were housed in groups (4 rats per cage), under an inverted light-day cycle, 12:12 hours, at 22°C and with free access to food (Harlan, México, S.A. de CV, Mexico City) and water. The Local Committee of Ethics on Animal Experimentation approved all experimental procedures, which followed the regulations established in the Mexican official norm for the use and care of laboratory animals: "NOM-062-ZOO-1999."

### Sexual Behavior Observations

In this study, male rats received 5 sexual behavior tests with receptive females. Female receptivity was induced by the sequential subcutaneous administration of oestradiol valerianate (4 µg/rat, subcutaneously) followed 44 hours later by progesterone (2 mg/animal, subcutaneously). Behavioral observations were conducted 4 hours

after progesterone administration and 2 hours after the onset of darkness. Males were introduced into a cylindrical observation cage, and a 5-minute adaptation period was allowed. Thereafter a receptive stimulus female was introduced and sexual behavior was recorded during 20 minutes. The sexual behavior parameters monitored were the following: intromission latency—time from introduction of the female until the first mount with pelvic thrusting and vaginal intromission; and ejaculation latency—time from the first intromission until ejaculation. The animals were classified as sexual experienced when ejaculation latencies of the first ejaculatory series recorded in the last 3 sessions occurred before 15 minutes after introduction of the female stimulus.

### Groups

Animals were divided into 4 groups. The first group (n = 4) served as the control group and was used to elicit the genital motor pattern of ejaculation by the mechanical stimulation of the urethra. Group 2 was employed to elicit the fictive ejaculation response by urethral stimulation and to receive saline solution used as a vehicle to prepare plants extracts. Groups 3 to 5 (n = 4 each) were used to analyze the effects of the systemic administration of *Montanoa tomentosa* (G3; 50 mg/kg), *Montanoa frutescens* (G4; 50 mg/kg), and *Montanoa grandiflora* (G5; 50 mg/kg) aqueous crude extracts, respectively, on the expression of the genital motor pattern of ejaculation. After the administration of individual treatments the animals received repeated mechanical stimulation of the urethra at 3-minute intervals, as described below, until the inhibition of the ejaculatory motor pattern.

### General Surgical Procedures

All animals were anesthetized with urethane (0.7 g/kg, intraperitoneal) and the adequacy of anesthesia was assessed by the absence of a withdrawal reflex after noxious paw pinch. The bulbospongiosus genital muscles were identified after a surgical incision on the perineum, and 2 platinum wires (Grass) were inserted into the muscles to record electromyographic activity, which was registered on a polygraph (Grass M7). An additional surgery was performed to expose the bulbar portion of the penis and its anatomical connections with the striated bulbospongiosus muscles for a better visualization of the genital rhythmic motor pattern of ejaculation and its associated genital events. The right femoral vein was cannulated for the administration of the plant extracts. At the end of the surgery the spinal cord was blunt transected at T6 spinal cord level.

### Activation and Recording of the Fictive Ejaculation Response

After the spinal cord transection, the genital motor pattern of ejaculation is immediately and spontaneously expressed with a mean latency of 1 to 3 minutes. To assess the capacity of the spinal cord to produce the genital motor pattern of ejaculation, after spinalization, one spontaneous genital motor pattern of ejaculation was allowed to be expressed and then recorded in the bulbospongiosus muscles. This muscle was selected as a monitor of the genital muscles activity since it discharges during ejaculation in synchrony with all genital muscles and given its superficial position on the perineum. Immediately after the expression of a spontaneous genital motor pattern of ejaculation, we evoked the ejaculatory response by sensory stimulation of the urethra, which consisted in its distension produced by the injection of saline solution with a syringe pump (200 µL/min) during 10 seconds,

through a PE-50 catheter (0.965 mm OD) inserted into the pelvic urethra via a bladder incision, while occluding the penis meatus to achieve an intraurethral stimulation by increasing its pressure that ranged from 20 to 30 mm Hg. Once the motor pattern of ejaculation was obtained by physiological-like (by urethral mechanical stimulation) means, the plant extracts were administered and the responses under its influence were analyzed. Finally, the motor pattern of ejaculation was repeatedly activated at 3-minute intervals by urethral stimulation until its inhibition. The criterion used to consider the inhibition of this response was the absence of the genital pattern of ejaculation expression on 2 consecutive stimulation periods following its repeated elicitation. At this moment, the stimulation protocol was completed.

### Activation of Fictive Ejaculation by the Systemic Administration of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* Aqueous Crude Extracts in Spinal Male Rats

To establish the pro-ejaculatory properties of *Montanoa frutescens* and *Montanoa grandiflora* on ejaculation in spinal male rats, in animals from groups 4 and 5, 2 consecutive genital motor patterns were elicited after spinalization by urethral stimulation to establish the capacity of the spinal cord to produce the ejaculatory motor response. The ejaculations obtained occurred within a 10-minute period. Immediately after the expression of the first urethrally induced ejaculatory pattern, the extracts of *Montanoa tomentosa* (G3), *Montanoa frutescens* (G4), and *Montanoa grandiflora* (G5) were injected intravenously, and the muscular genital responses observed under their influence were registered and graphed. After the injection of the plant extracts and to verify the expression of ejaculation by the spinal cord, an additional urethral stimulation was applied and its resulting genital responses, if present, were observed. When no response was obtained, the experiment was ended. Data were compared to those obtained in control (G1) animals and to those obtained in vehicle-treated animals (G2).

### Quantification of Genital Motor Pattern of Ejaculation Variables

To analyze the pro-sexual properties of *Montanoa* plants, we used electromyographic techniques to register the rhythmic activity of the bulbospongiosus muscles. In the present study, we defined a genital motor pattern of ejaculation as the expression of the first motor train of a genital pattern of ejaculation (which could be accompanied or not by an after-discharge component) obtained in response to mechanical stimulation of urethra or after the systemic administration of the aqueous extracts. The parameters considered for the analysis of the genital motor patterns registered were the latency to the expression of the motor patterns of ejaculation after mechanical stimulation of the urethra or after the administration of the extracts, the number of discharges in the genital motor patterns of ejaculation and its frequency, as well as the number of genital motor patterns expressed prior to its inhibition. The after-discharge component of the motor pattern of ejaculation is only observed in urethrally evoked responses. Since this type of sensorial stimulation exerts an almost immediate cumulative inhibitory effect on the expression of the after-discharge component, we determined to exclude the after-discharge component from the quantitative analysis of both the urethrally induced and plant extracts-induced motor patterns of ejaculation.

### Preparation of Cihuapatli Extracts

*Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* were all collected in its habitat in Tlaxcala during October 2012 and were authenticated by personnel from the Universidad Autónoma de Tlaxcala Herbarium, where voucher specimens are preserved and cultivated (Serial Numbers: MT UATX10, MF UATX11, and MG UATX12). Leaves of *Montanoas* were collected and prepared to be dried during 20 days. Doses of aqueous crude extracts of *Montanoas* were selected on the basis of previous studies<sup>8,13,14</sup> and confirmed with midwives and traditional healers. In brief, to select the convenient doses, we enquired about the quantity of *Montanoas* employed by a healer in Tlaxcala, México. The traditional healer recommended us to boil equal parts (20 leaves/plant) of the dried plants to obtain an aqueous crude extract with pro-sexual effects in adult men. Once dried, the materials were ground into a fine powder, 100 g of which were mixed with 100 mL of distilled water. These mixtures were warmed for approximately 10 minutes, just before boiling. The obtained infusions were filtered and oven dried at a temperature of 55°C, and the brownish residues of the extract yield were calculated to be 130 mg for *Montanoa tomentosa*, 80 mg for *Montanoa frutescens*, and 65 mg for *Montanoa grandiflora*. The dried extracts of the plants were maintained at 3°C and then used to prepare the stock solutions, 50 mg/mL. Infusions and solutions were prepared 40 minutes prior to its administration to avoid modifications in the chemical properties of the extracts.

### Drugs

Urethane was purchased from Sigma Chemical Co (Lot 38H5236; St Louis, MO) and dissolved in bidistilled water and administered at 20%.

### Data Analysis

Bulbospongiosus electromyographic activity was recorded differentially, amplified, and filtered (1000×, 0.1-1 kHz bandpass) (Poliview Data Acquisition System; Grass Astro-Med Inc, Warwick, RI). Quantitative comparisons among groups were calculated from means of genital motor patterns of ejaculation and statistically analyzed by using a one-way ANOVA, followed by the Tukey test. The variables analyzed from the genital motor pattern of ejaculation included the latency of response, the number and frequency of discharges, and the total number of evoked genital motor patterns of ejaculation.  $P < .05$  was considered to be statistically significant. The Sigma Stat program (version 3.5) was used for all statistical analyses.

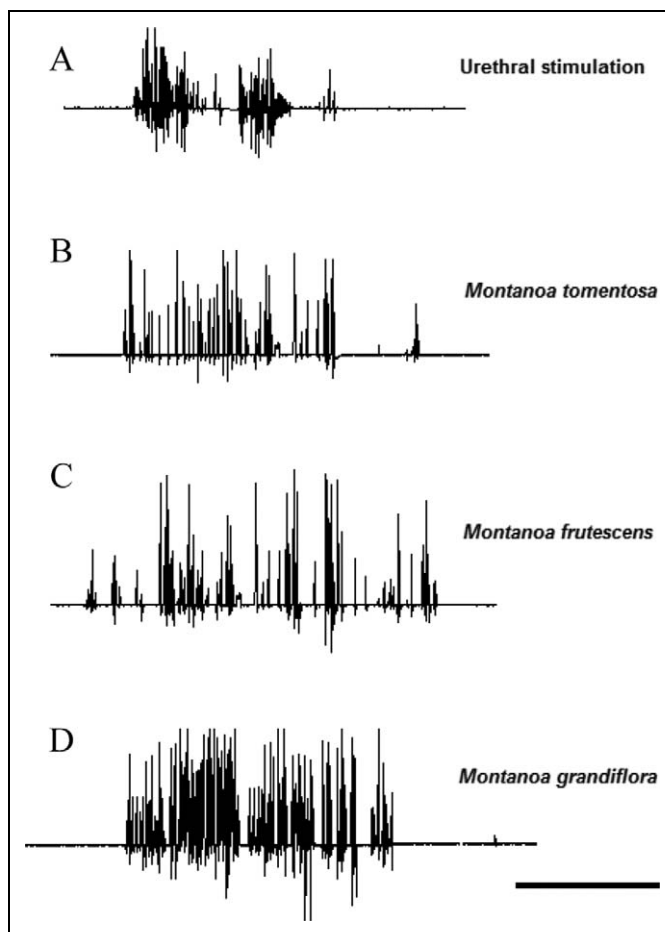
### Results

#### General Observations on the Activation of the Fictive Ejaculation by *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* Aqueous Crude Extracts

As previously reported, in all the studied control animals the genital motor pattern of ejaculation elicited by urethral stimulation were registered as highly rhythmic motor patterns of bulbospongiosus muscle activity and included a first ejaculatory motor train and an after-discharge component. Rhythmic contractions elicited by urethral stimulation were always

accompanied by the potent expulsion of seminal secretions and always coincided with phasic penile erections including penile movements such as flaring, flips, and cups. Consecutive repeated stimulation of the urethra induced an inhibitory effect on the genital motor pattern of ejaculation, which was gradually evidenced in the parameters of the first ejaculatory motor train and in its after-discharge component, and a progressive reduction in the number of motor discharges and its frequency was observed in successive motor patterns of ejaculation. In these animals, the first sensory elicited ejaculatory phase was the most potent and the last one, previous to its inhibition, the weakest. The ejaculatory capacity of control animals consisted of the expression of a mean number of 6 genital motor patterns of ejaculation. Once the ejaculatory capacity maximum level was accomplished, no further genital motor patterns of ejaculation including its penile movements or expulsion of seminal contents occurred. At this moment, the ejaculatory ability was considered as inhibited. Vehicle-treated animals were able to produce ejaculatory motor patterns until its inhibition after urethral stimulation, but given that these animals lacked genital and ejaculatory reactions after the administration of vehicle solution, they were discarded for statistical comparisons.

Besides, *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* were all able to activate the genital motor pattern of ejaculation in spinal male rats. Thus, systemic administration of individual *Montanoas* elicited motor patterns of ejaculation that always consisted of the expression of highly rhythmic motor patterns registered in the bulbospongiosus muscles, very similar to those registered in control animals. All aqueous crude extracts of *Montanoas* elicited ejaculatory motor responses always accompanied by penile movements including flaring, flips, and cups, but without the after-discharge component. General visual observations of penile erections and movements elicited by the aqueous crude extracts of the *Montanoas* analyzed permitted us to notice that the expression of these sexual responses was significantly more potent as compared to that elicited by urethral stimulation (Figure 1). Thus, while penile erections and movements elicited by urethral stimulation were described as epiphenomena in our model, animals receiving crude extracts of *Montanoas* expressed penile cups, flips, and flaring very pronounced similar to that exhibited by male rats during copulation (our laboratory observations). In contrast to control animals where penile erections were displayed as phasic genital responses, similar to those exhibited by male rats during copulation and during ex-copula penile tests, male rats treated with the *Montanoa* extracts displayed only tonic penile erections. Repeated urethral stimulation also induced the inhibition on the genital motor pattern of ejaculation after the administration of the *Montanoa* extracts but significant differences among treatments were noticed. The main difference in the repeated activation of genital motor patterns of ejaculation in male rats treated with *Montanoa* extracts was a general increase in the ejaculatory capacity (Figures 2 and 3).

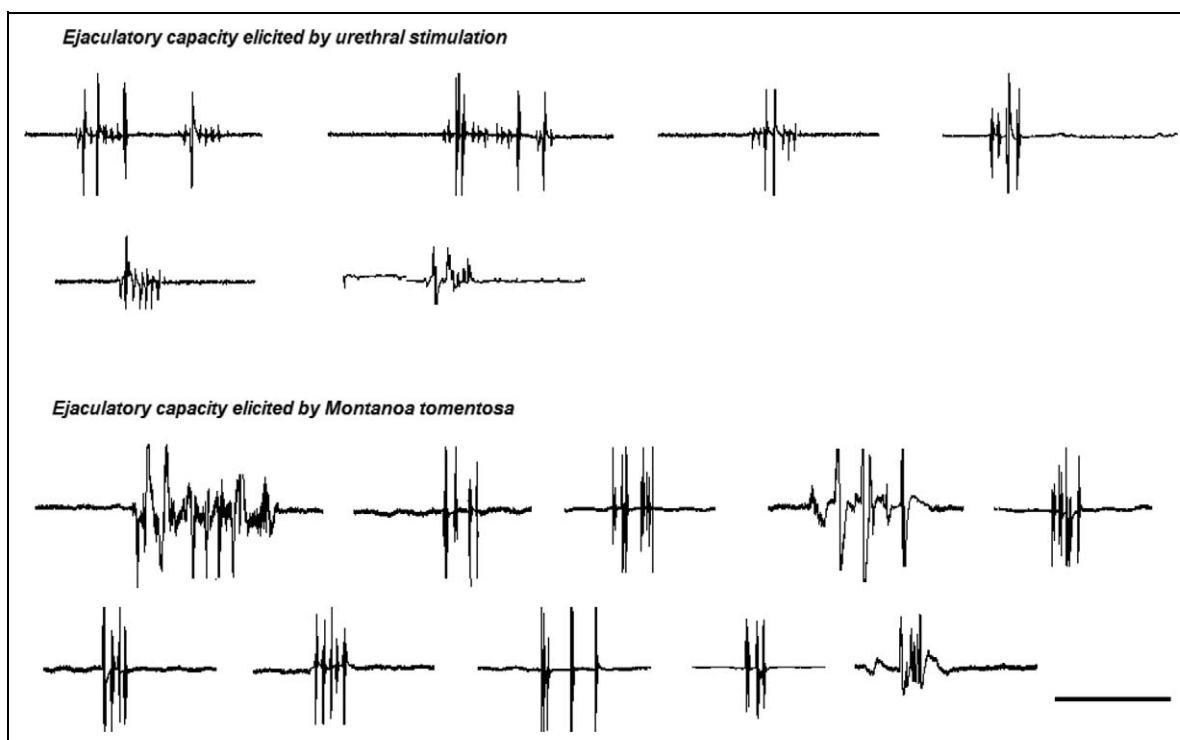


**Figure 1.** Activation of ejaculatory rhythmic motor patterns in sexually experienced spinal male rats by urethral stimulation (A) and by the intravenous injection of *Montanoa tomentosa* (50 mg/kg) (B), *Montanoa frutescens* (50 mg/kg) (C), and *Montanoa grandiflora* (50 mg/kg) (D) aqueous crude extracts. Calibration bar = 10 seconds.

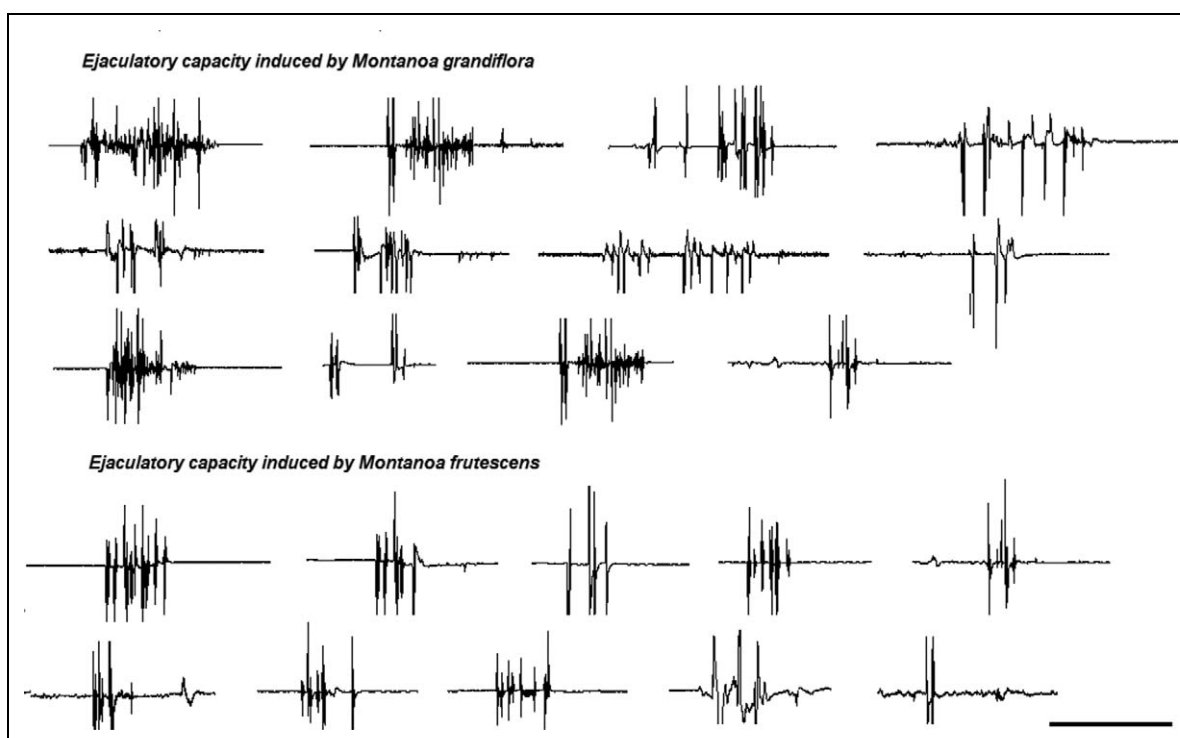
### Effects of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* Aqueous Crude Extracts on the Genital Motor Pattern of Ejaculation Parameters

When compared to control animals, administration of *Montanoa tomentosa* (50 mg/kg) produced the immediate expression of genital motor patterns of ejaculation, with a statistically significant reduced latency of response ( $P \leq .001$ ). The motor patterns of ejaculation elicited by the extract of this plant exhibited an increased number of discharges ( $P = .004$ ) but the frequency of discharge was not modified ( $P = .70$ ; Table 1). In addition, a significantly increased number of genital motor patterns of ejaculation ( $P = .005$ ) were elicited after the injection of *Montanoa tomentosa*.

Systemic administration of *Montanoa frutescens* (50 mg/kg) significantly reduced the latency of response ( $P \leq .001$ ) and provoked statistically significant increases in the number of discharges ( $P = .004$ ) and in the number of motor patterns of ejaculation ( $P = .005$ ) expressed, when compared to control



**Figure 2.** Sample electromyography tracings showing the increase of the ejaculatory capacity elicited by the systemic injection of 50 mg/kg of *Montanoa tomentosa* in spinal male rats. Calibration bar = 10 seconds.



**Figure 3.** Electromyography tracings depicting the increased ejaculatory capacity promoted by the systemic injection of 50 mg/kg of *Montanoa frutescens* and *Montanoa grandiflora* in spinal male rats. Calibration bar = 10 seconds.

**Table 1.** Comparison Among the Parameters of the Genital Motor Patterns of Ejaculation Registered in Response to Urethral Stimulation and Those Obtained After Systemic Administration of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora*.

Treatment	Latency	Number of Discharges	Frequency (Hz)	Ejaculatory Motor Patterns
Urethraly induced	15.33 ± 3.08	6.25 ± 0.49	1.02 ± 0.04	6.25 ± 0.62
<i>Montanoa tomentosa</i> (50 mg/kg)	0 <sup>a</sup>	17.27 ± 2.04 <sup>b</sup>	0.99 ± 0.03	10.50 ± 0.64 <sup>c</sup>
<i>Montanoa frutescens</i> (25 mg/kg)	0 <sup>a</sup>	23.14 ± 3.8 <sup>b</sup>	1.07 ± 0.04	11.25 ± 1.03 <sup>c</sup>
<i>Montanoa grandiflora</i> (25 mg/kg)	0 <sup>a</sup>	17.60 ± 2.61 <sup>b</sup>	1.05 ± 0.03	11.75 ± 1.25 <sup>c</sup>

Tukey test: latency of discharge versus control animals <sup>a</sup> $P \leq .001$ , number of discharges versus control animals, <sup>b</sup> $P = .004$ , number of motor patterns versus control animals <sup>c</sup> $P = 0.005$ .

animals. The frequency of discharge of the genital motor patterns of ejaculation elicited by *Montanoa frutescens* was not significantly modified ( $P = .70$ ). The pro-ejaculatory effect obtained after administration of *Montanoa frutescens* to spinal rats elicited more robust motor patterns than *Montanoa tomentosa* and *Montanoa grandiflora* (see Table 1).

Besides, when compared to control animals, intravenous injection of *Montanoa grandiflora* (50 mg/kg) exerted a statistically significant facilitatory effect on the latency of response ( $P \leq .001$ ), on the number of discharges ( $P = .004$ ), and in the number of genital motor patterns of ejaculation ( $P = .005$ ) evoked, but not in the frequency of discharges of the motor patterns of ejaculation ( $P = .70$ ; see Table 1).

## Discussion

Mexican Cihuapatli as a traditional remedy for reproductive impairments has been extensively documented.<sup>6,7</sup> Experimental evidence has shown that Cihuapatli can cross the blood-brain barrier and promotes an aphrodisiac effect.<sup>14</sup> Present findings are in line with previous observations and show that (a) the systemic administration of *Montanoa frutescens* and *Montanoa grandiflora* aqueous crude extracts promotes the expression of genital motor patterns of ejaculation with similar electromyographic features to those elicited by *Montanoa tomentosa* and (b) the pro-ejaculatory effects promoted by these extracts are exerted on the circuits that control ejaculation located at the spinal cord level. Thus, the data suggest that the aqueous crude extracts of *Montanoa frutescens* and *Montanoa grandiflora* have aphrodisiac properties comparable to the *Montanoa tomentosa* aqueous crude extract.

Aphrodisiacs are substances that may increase the libido (ie, sexual desire), sexual potency (ie, effectiveness of erection and ejaculation), and sexual pleasure.<sup>16</sup> Aphrodisiacs actions of *Montanoa tomentosa* are exerted on the ejaculatory function by significantly decreasing its latency<sup>13</sup> and also by positively influencing the electromyographic variables of the genital motor pattern of ejaculation.<sup>14</sup> In the present study we used the fictive ejaculation model to evaluate the pro-ejaculatory potential of *Montanoa frutescens* and *Montanoa grandiflora* after the analysis of the latency of response, the number and frequency of discharges of the ejaculatory motor pattern, and the number of motor patterns of ejaculation evoked. In this animal model, the latency to express ejaculation after the application of physiological-like stimulation (urethral stimulation) or after the

administration of standard drugs or plants extracts mirror the threshold of ejaculation that could be seen in conscious male rats and reflects the responsiveness of the spinal cord to the sexual stimuli.<sup>17</sup> Previous studies on the pro-ejaculatory effect of *Montanoa tomentosa* in spinal animals did not report the effects of this plant extract on the latency of ejaculation. In the present study, we observed that *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* all promote the rapid activation of the genital motor pattern of ejaculation. Thus, after systemic administration of the extracts, it was found that the latencies to express the genital motor pattern of ejaculation in rats are drastically reduced, suggesting that the compounds contained in these plants can act on the spinal ejaculation circuits to modify the ejaculatory threshold and hence the rapid activation of the ejaculatory response, in all probability after decreasing the intraspinal inhibitory tone of the circuit of ejaculation. In line with this notion, it has been demonstrated that *Montanoa tomentosa* promotes the rapid activation of the ejaculatory response in behaving male rats,<sup>13</sup> and thus, based on the present data, it is proposed that *Montanoa frutescens* and *Montanoa grandiflora* could provoke a similar significant reduction in the ejaculation latency in copulating male rats. Further behavioral experiments are necessary to test this proposal.

The number of discharges in the genital motor pattern of ejaculation reflects the robustness of the rhythmic motor pattern<sup>18</sup> given its similarities with the electromyographic response seen in copulating rats<sup>19</sup> and an increase in its number has been paralleled to a facilitated ejaculatory response.<sup>20,21</sup> *Montanoa tomentosa* promotes a significantly increased number of ejaculatory rhythmic discharges.<sup>14</sup> Findings on the number of discharges obtained in the present study using *Montanoa tomentosa* are in line with previous data and in addition show that *Montanoa grandiflora* and *Montanoa frutescens* both exhibit pro-ejaculatory profiles that allow significant increases in the number of ejaculatory discharges. Besides, genital motor patterns of ejaculation produced by the administration of *Montanoa frutescens* show to be the most robust in the number of discharges when compared to control values. Thus, it seems that the aqueous crude extract of *Montanoa frutescens* has the most potent aphrodisiac capability to act on the spinal circuits to generate robust ejaculatory responses. The frequency of discharge in the ejaculatory motor train was maintained constant in response to the 3 extracts evaluated here, and no statistically significant differences were found when compared to control

animals. This is a striking result, and we propose that all the aqueous crude extracts of the *Montanoas* used in this study induce a steady ejaculatory rhythm, comparable to the physiological ejaculatory rhythms of the genital motor pattern of ejaculation, despite significant changes in the other parameters of this response. In line with this notion, traditional healers and midwives reveal that, at parturition, all *Montanoas* promote a very rhythmic labor (our unpublished data). Additionally, experimental studies have described that in uterine strips *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* induce very rhythmic and organized uterine electromyography, in contrast to other uterotonic agents.<sup>12,15</sup>

In the fictive ejaculation model the number of genital motor patterns of ejaculation evoked by urethral or pharmacological stimulation is related to the ejaculatory capacity of male rats.<sup>22</sup> A long-lasting inhibitory state in the expression of the genital motor patterns of ejaculation can be induced as a result of repeated stimulation of the urethra. Thus, once the supraspinal inhibitory influences are removed by spinal cord transection, a spinal intrinsic rhythm of activation of the motor pattern of ejaculation, which occurs at intervals of 3.5 minutes, is turned on<sup>17,23</sup> and the urethral stimulation activates a sensory feedback mechanism that exerts both facilitatory and inhibitory influences on the ejaculatory trains.<sup>24</sup> The excitatory influences of genital sensory inputs provided by urethral stimulation are observed only at the initial period of the stimulation protocol, and as a result, robust genital motor patterns of ejaculation are registered, but sustained urethral stimulation rapidly commences to inhibit the expression of ejaculatory trains. Inhibition of ejaculatory trains is considered as a spinal component of the sexual satiation phenomenon.<sup>17,21</sup> In a previous study, we analyzed the pro-ejaculatory effect of *Montanoa tomentosa* without including the analysis of changes in the ejaculatory capacity acquired after by the administration of its extract.<sup>14</sup> Data of the present study show that the systemic administration of *Montanoa tomentosa*, as well as the *Montanoa frutescens* and *Montanoa grandiflora* extracts, increases the ejaculatory capacity of spinal rats. We previously have suggested that aphrodisiac plants can increase the ejaculatory capacity as a result of sustained increases in the activity of the spinal generator for ejaculation<sup>18</sup> targeted by the compounds of plant extracts. In copulating animals, the overexpression of genital motor patterns of ejaculation might improve ejaculatory capacity not only by the reduction of the ejaculatory latency but also by heightening the sexual experience permitting subsequent copulatory series. This notion could apply to the augment in the ejaculatory capacity seen after the systemic administration of *Montanoa* extracts described here and it could be thought that the ejaculatory capacity can be positively driven by the compounds contained in these plants.

The exact mechanism through which the aqueous crude extract of *Montanoas* exerts its pro-ejaculatory actions is not known, but a partial mechanism involving the oxytocinergic spinal system has been proposed.<sup>14</sup> Evidence shows that oxytocin is actively involved in regulating penile sexual reflexes, including ejaculation and penile erection via spinal mechanisms.<sup>25,26</sup> Systemic or intracerebroventricular OT administration

decreases ejaculation latency and the postejaculatory interval in male rats<sup>27</sup> and facilitate the expression of the genital motor pattern of ejaculation in anesthetized and spinalized male rats.<sup>24</sup> In the present study, the fact that *Montanoa frutescens* and *Montanoa grandiflora* extracts confirm pro-ejaculatory actions acting with unexpected similarities on all parameters of the genital motor pattern of ejaculation suggest that an oxytocinergic mechanism could be also implicated, at least partially, in the aphrodisiac effect of the 3 *Montanoas* described here. In support with this notion, we noticed after the administration of *Montanoa* extracts sustained penile erections accompanying ejaculatory rhythmic discharges, a response that suggests tonic activation of oxytocinergic receptors. Specific experiments to dissect the effect of the *Montanoas* on ejaculation and penile erection are necessary.

It has been demonstrated that kaurenes that include the kaurenoic, grandiflorenic, kauradienoic, and monoginoic acids are the main biologically active compounds contained in the extracts of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora*.<sup>28-31</sup> At present, the quantities of kaurenes contained in *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* has been not elucidated, but might be speculated that equivalent concentrations of these acids could be contained in all *Montanoas* and that these kaurenes are responsible of the pro-ejaculatory effects of these plant extracts. Further experiments using individual fractions of *Montanoas* should be conducted to test this possibility.

## Conclusion

The present data show that systemic administration of *Montanoa tomentosa*, *Montanoa frutescens*, and *Montanoa grandiflora* exerts aphrodisiac effects in spinal male rats by increasing the sexual potency after a noteworthy facilitation of the genital motor patterns of ejaculation.

## Author Contributions

MCJ designed the experimental study and MAF and MLRP carried out the experiments. Data were analyzed and described by MCJ, MAF, and MLRP. MCJ prepared the article, and MAF and MLRP reviewed the final version of the article.

## Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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## Ethical Approval

This study received authorization from the Comité Interno de Ética de la Escuela de Medicina Veterinaria y Zootecnia de la Universidad Autónoma de Tlaxcala (MVZ-189/12).

## References

1. Wijesinha S, Piterman L, Kirby CN. The male reproductive system—an overview of common problems. *Aust Fam Physician*. 2013;42:276-278.
2. Linton K, Wylie KR. Recent advances in the treatment of premature ejaculation. *Drug Des Dev Ther*. 2010;4:1-6.
3. Singh AP, Singh R. Potent natural aphrodisiacs for the management of erectile dysfunction and male sexual debilities. *Front Biosci (School Ed)*. 2012;1:167-180.
4. Kotta S, Ansari SH, Ali J. Exploring scientifically proven herbal aphrodisiacs. *Pharmacogn Rev*. 2013;7:1-10.
5. Ximenez F. Cuatro libros de la naturaleza y virtudes de las plantas y animales que están recevidos en el uso de medicina en la Nueva España, y la método, y corrección y preperación que para administrarlas se requiere con lo que el doctor Francisco Hernández escribió en lengua latina. Viuda de Diego López Davalos, 1615, México.
6. Levine SD, Hahn DW, Cotter ML, et al. The Mexican plant zoapatle (*Montanoa tomentosa*) in reproductive medicine. Past, present and future. *J Reprod Med*. 1981;26:524-528.
7. Gallegos AJ. The zoapatle VI. Revisited. *Contraception*. 1985;31:487-497.
8. Carro-Juárez M, Rodríguez-Landa JF, Rodríguez-Peña Mde L, Rovirosa-Hernández Mde J, García-Orduña F. The aqueous crude extract of *Montanoa frutescens* produces anxiolytic-like effects similarly to diazepam in Wistar rats involvement of GABAA receptor. *J Ethnopharmacol*. 2012;143:592-598.
9. de la Cruz-Badiano Codex. Libellus de Medicinalibus Indorum Herbis. 1552, Ediciones del Instituto Mexicano del Seguro Social, México DF.
10. Hahnn DW, Ericson EW, Lai MT, Probst A. Antifertility activity of *Montanoa tomentosa* (Zoapatle). *Contraception*. 1981;23:133-140.
11. Gallegos AJ. The zoapatle I. A traditional remedy from Mexico emerging to modern times. *Contraception*. 1983;27:211-225.
12. Ponce-Monter H, Girón H, Lozoya X, et al. The zoapatle III. Biological and uterotonic properties of aqueous plant extract. *Contraception*. 1983;27:239-253.
13. Carro-Juárez M, Cervantes E, Cervantes-Méndez M, Rodríguez-Manzo G. Aphrodisiacs properties of *Montanoa tomentosa* aqueous crude extract in male rats. *Pharmacol Biochem Behav*. 2004;78:129-134.
14. Carro-Juárez M, Lobaton I, Benitez O, Espiritu A. Pro-ejaculatory effect of the aqueous crude extract of cihuapatli (*Montanoa tomentosa*) in spinal male rats. *J Ethnopharmacol*. 2006;106:111-116.
15. Estrada AV, Enríquez RG, Lozoya X, et al. The zoapatle II. Botanical and ecological determinants. *Contraception*. 1983;27:227-237.
16. Sandroni P. Aphrodisiacs past and present: a historical review. *Clin Auton Res*. 2001;11:303-307.
17. Carro-Juárez M, Rodríguez-Manzo G. Sensory and motor aspects of the coital reflex in the spinal male rat. *Behav Brain Res*. 2000;108:97-103.
18. Carro-Juarez M, Rodríguez-Manzo G. The spinal pattern generator for ejaculation. *Brain Res Rev*. 2008;58:106-120.
19. Holmes GM, Chapple WD, Leipheimer RE, Sachs BD. Electromyographic analysis of the male rat perineal muscles during copulation and reflexive erections. *Physiol Behav*. 1991;49:1235-1246.
20. Durán ID, Rojas-Piloni JG, Cueva-Rolón R. Facilitation and inhibition of the urethrogenital reflex in spinal cord-transected rats. *Brain Res*. 1997;775:1-10.
21. Carro-Juárez M, Rodríguez-Manzo G. Participation of endogenous opioids in the inhibition of the spinal generator for ejaculation in rats. *J Sex Med*. 2009;6:3045-3055.
22. Carro-Juárez M, Alcazar C, Ballesteros-Polvo E, Villalobos-Peñalosa P. Increase of ejaculatory capacity by systemic administration of the oquichpatli (*Senecio cardiophyllus*) aqueous crude extract in male rats. *J Ethnopharmacol*. 2009;126:506-511.
23. Carro-Juárez M, Cruz SL, Rodríguez-Manzo G. Evidence for the involvement of a spinal pattern generator in the control of the genital motor pattern of ejaculation. *Brain Res*. 2003;975:222-228.
24. Carro-Juárez M, Rodríguez-Manzo G. Evidence for the presence and functioning of the spinal generator for ejaculation in neonatal male rats. *Int J Impot Res*. 2005;17:270-276.
25. Burri A, Heinrichs M, Schedlowski M, Kruger TH. The acute effects of intranasal oxytocin administration on endocrine and sexual function in males. *Psychoneuroendocrinology*. 2008;33:591-600.
26. Corona G, Jannini EA, Vignozzi L, Rastrelli G, Maggi M. The hormonal control of ejaculation. *Nat Rev Urol*. 2012;9:508-519.
27. Arletti R, Bazzani C, Castelli M, Bertolini A. Oxytocin improves male copulatory performance in rats. *Horm Behav*. 1985;19:14-20.
28. Enríquez RG, Escobar LI, Romero ML, Chávez MA, Lozoya X. Determination of grandiflorenic (kauradienoic) acid in organic and aqueous extracts of *Montanoa tomentosa* (zoapatle) by reversed phase high performance liquid chromatography. *J Chromatogr*. 1983;258:297-301.
29. Bejar E, Enríquez R, Lozoya X. The in vitro effect of grandiflorenic acid and zoapatle aqueous crude extract upon spontaneous contractility of the rat uterus during estrus cycle. *J Ethnopharmacol*. 1984;11:87-97.
30. Valencia A, Wens A, Ponce-Monter H, et al. Zoapatle. XII. In vitro effect of kaurenoic acid isolated from *Montanoa frutescens* and two derivatives upon human spermatozoa. *J Ethnopharmacol*. 1986;18:89-94.
31. Villa-Ruano N, Betancourt-Jiménez MG, Lozoya-Gloria E. Biosynthesis of uterotonic diterpenes from *Montanoa tomentosa* (zoapatle). *J Plant Physiol*. 2009;166:1961-1967.