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Maca (*Lepidium meyenii*) supplementation increase the sexual capacity of low but not high sexual performance rams (*Ovis aries*)

[La suplementación con maca (*Lepidium meyenii*) incrementa la capacidad de servicio en carneros (*Ovis aries*) de bajo pero no de alto desempeño sexual]

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Abstract: To evaluate the effect of maca (*Lepidium meyenii*) supplementation on the sexual capacity of males, 40 rams were classified as high (HP) and low performers (LP). Within each category (n = 20), ten rams were supplemented with 233 mg of dry maca/kg of body weight/day during four weeks, followed by four weeks of a control diet (residual), while the rest of the animals remained as controls during the eight-weeks period. LP rams increased (P < 0.05) all sexual behaviors during maca supplementation up to HP levels, decreasing to control planes one week after the supplementation ceased. In contrast, in HP rams, maca supplementation only increased genital sniffs and nudging, while mounts and ejaculations remained unaffected during both supplementation and residual phases. It was concluded that maca supplementation affects males differently, according to their original sexual capacity.

Keywords: Sexual behavior, supplementation, sheep, aphrodisiac, breeders

Resumen: Con el propósito de evaluar el efecto de la suplementación con maca (*Lepidium meyenii*) en la capacidad de servicio de los machos, 40 carneros se clasificaron como de alto (HP) y bajo desempeño sexual (LP). Dentro de cada categoría (n = 20), diez carneros se suplementaron con 233 mg de maca seca/kg de peso corporal/día durante cuatro semanas, seguidos de cuatro semanas de dieta control (residual), mientras que el resto de los animales permanecieron como testigos durante las ocho semanas. Los carneros LP incrementaron (P<0.05) todas sus conductas sexuales durante la suplementación hasta niveles similares a los mostrados por carneros HP, disminuyendo a niveles del grupo control una semana después de terminada la suplementación. En contraste, en los carneros HP, la suplementación com maca sólo incrementó las conductas de olfateo genital y cortejo, sin embargo montas y eyaculaciones permanecieron sin cambios tanto durante la fase de suplementación como la residual. Se concluye que la suplementación con maca actúa diferente en machos de acuerdo a su capacidad de servicio.

Palabras clave: Conducta sexual, suplementación, ovinos, afrodisíaco, reproductores

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INTRODUCCION

Maca (Lepidium meyenii) is an Andean plant that belongs to the brassica (mustard) family (Edzard et al., 2010). In the past, maca has been described to improve sexual behavior in experimental (Zheng et al., 2000; Cicero et al., 2001; Cicero et al., 2002) and domestic animals (Clément et al., 2010; Lavana et al., 2013), which is likely due to the phytosterols or phytoestrogens present in this plant (Wang et al., 2007), although conflictive results have been observed (Lentz, 2007; Edzard et al., 2010). Several possibilities have been mentioned in an attempt to explain these dissimilarities, for example, the use of diverse maca ecotype supplementation (Gonzales et al., 2006), different doses (Chung et al., 2005) or methodologies used (Chung et al., 2005), dissimilar number of trials or sample sizes, among others (Edzard et al., 2010).

In a recent study, Lavana et al. (2013) found a significant increase in the sexual behavior of rams associated with a large variation in the individual response. This lack of homogeneity might have contributed to mask potentially clearer effects of maca. With this in mind, it is feasible to think that at least another reason for the heterogeneous results obtained with maca supplementation could be that animals might respond differently, at least in this case, according to their sexual capacity. Similarly as the results obtained by Zheng et al. (2000) who stated that oral administration of maca, decreased the latent period of erection in rats, but only in those with erectile dysfunction. In any case, the achievement of large improvements in rams with high mating proficiency would be difficult to expect. For this reasons, differentiation between effects in rams with high and low serving capacity appears helpful.

The high variation in mating proficiency of rams has been previously reported (Blockey & Wilkins, 1984) and serving capacity tests have been used to identify individual differences in the mating competence of rams (Snowder *et al.*, 2002), which in addition can be used to predict ram mating performance during pasture breeding (Perkins *et al.*, 1992). Therefore, the purpose of the present study was to compare the effect of maca supplementation on the libido of low vs high sexual capacity hair sheep rams.

MATERIALS AND METHODS

Animals and General Management

The study was carried out at the Experimental Station

of the University of the State of Morelos, Mexico, 18° 56" N and 99° 13" W, situated at 2160 m asl and with an average annual rainfall and temperature of 1243 mm and 20° C, respectively. Animals were 40 sexually naive Saint croix rams, 11-15 months old, weighing 48.6 \pm 8.5 kg at the beginning of the experiment, that were housed isolated from ewes in an open-sided barn under natural lighting. Throughout the research, the animals were fed 500 g/ram/day of a commercial concentrate diet formulated for breeding rams, with 16% protein (Nutres®), and free access to water and hay.

The experimental animals were kept, maintained and treated in adherence to accepted standards for the humane treatment of animals.

Maca (Lepidiummeyenii) supplement

The black maca hypocotyl meal used for the experiment was purchased directly from Macandina® (Peru) who commercializes the product as an organic powder milled food supplement from black hypocotyls only. Maca supplementation consisted in 233 mg of dry maca/kg of body weight/day during four weeks. With this amount of maca, Clément *et al.* (2010) and Lavana *et al.* (2013) found an increase in mounting behavior in cattle and sheep, respectively.

The daily individual ratio was dissolved in 250 ml of water and with the help of a plastic bottle, an oral intake was forced to ensure that each ram consumed the amount of maca provided. This procedure was conducted daily during the supplementation period, before the regular food was offered. The product was generally consumed with only minimal amounts of refusals if any.

Experimental design

One week before maca supplementation, all rams were tested on the same days with a variation of a sexual capacity (SC) test (Kilgour, 1985). The tests were repeated on Monday, Wednesday and Friday. According to the results of these tests, the rams were classified as high performers (HP; averaging > 2ejaculations) or low performers (LP; averaging < 1ejaculation).

At the beginning of the experiment, animals within each category (LP and HP) were randomly allocated to either a control diet for eight weeks or a maca supplemented diet for four weeks, followed by four weeks of a control diet (residual phase).

Evaluations

Sexual capacity tests consisted in the individual assessment of all rams in which each animal was exposed for 20 min to a single restrained estrousinduced female in a portable service crate attached to the floor of the test pen. Each male was visually isolated from other members of the flock and the order of testing was at random, alternating treated and non-treated animals to control for testing sequence effects and variation due to time of day and over test days. The ewe was changed after each test to avoid odors from the previous tested male on the females while tested with subsequent rams. Sexual tests were performed between 8:00 and 14:00 h in two contiguous 4 x 5 m test pens. Two rams were tested simultaneously by one observer. Rams were separated by solid walls precluding visual contact between them. A single observer monitored both pens, and manually recorded behavioral data.

During the SC tests, the observer recorded whether or not the rams investigated and courted the stimulating ewe; recorded when the ram sniffed the genital region or when the ram performed nudging. In addition, the observer recorded the number of mounts (the ram becomes firmly planted on the ewe's rump) and ejaculations (services) characterized by the ram tossing his head upwards while arching his back and thrusting his hips forward.

Statistical analysis

Recorded variables were compared with a two way anova for repeated measures, considering the treatments (treated vs. control), and (HP vs. LP) as well as their interaction. The frequencies of the behaviors recorded were previously normalized by: log [x+1] transformation, as suggested by Fiol & Ungerfeld (2011). Data are presented as mean \pm SEM.

RESULTS AND DISCUSSION

In LP rams, genital sniffs, nudgings and ejaculations increased (P < 0.05) during maca supplementation up to frequencies observed in HP rams, while mounts increased even at higher levels. The latter, suggests even though ejaculation frequencies were similar in LP and HP, the latter were more efficient in terms of mounts/ejaculation. Either way, the increases observed are in accord with previous studies in which oral administration of maca improved the sexual function of mice (Zheng *et al.*, 2000), rats (Cícero *et al.*, 2001; Cícero *et al.*, 2002), livestock (Clément *et* *al.*, 2010; Lavana *et al.*, 2013) and men (Stone *et al.*, 2009). As evidenced by an increase in the number of ejaculations and improvement of self-rated sexual desire, respectively. Contrary, the lack of effect observed in HP rams confirms that not all animals respond equally, and that HP males might need higher doses of maca or longer supplementation periods to attain significant increases. On the other hand, it is also possible that the levels of sexual behaviors assessed cannot go higher.

During the residual phase, all sexual behaviors in LP rams decreased (P < 0.05; genital sniffs, mounts and ejaculations) or tended to decrease (P > 0.10; nudging), not to baseline levels, but to controls, which supports the idea that maca supplementation is not only necessary to improve sexual capacity, but also to keep it at high levels.

In contrast, in HP rams, genital sniff and nudging frequencies increased (P < 0.05) during maca supplementation, remaining at high levels during the residual phase, while mounts and ejaculations remained unaffected during the whole experiment (Table Nº 1). The number of ejaculations in HP rams remained similar to their pre-treatment levels and controls during the supplementation (Figure 1) and residual phases (Figure 2). These findings suggests that in HP rams, maca supplementation might act only as a trigger, favoring a response in some sexual behaviors that will remain high, even four weeks after maca supplementation ceased. Mounts and ejaculations which were already at high levels before the experiment remained unaffected during both phases.

Previous studies in domestic animals (Lavana et al., 2013) have established that the effect of maca supplementation is evident eight weeks after supplementation, and disappeared after another eight weeks of residual phase, when animals were evaluated. Another interesting contribution of the present study is that the length of time after treatment that the behavior can be affected and the length of the residual effect were determined. Low performance animals displayed more ejaculations than their pretreatment levels starting three weeks after maca supplementation (Figure 1), and decreased to control levels two weeks after supplementation. In addition, these rams displayed a similar number of ejaculations to the HP rams from week two until the end of the supplementation period (Figure 1). Furthermore, during the first week of the residual period, no difference (P > 0.05) was found between HP and LP

rams. However, after week five, the number of ejaculations in LP rams decreased in comparison with HP rams, but remained above pre-treatment levels until the end of the study (Figure 2). It might be possible that ejaculation frequencies in treated LP animals did not fall to basal levels during the residual phase, due to at least three possibilities: First, a learning process that subjects might have achieved, predicting a sexual episode after more than six weeks of performing continuous SC tests. A second nonopposed possibility would be that, as time went on, rams were more sexually mature. Even though the experiment took over eight weeks, certain maturation accomplished during this could be period. particularly due to the high sexual activity involved and the limited sexual experience that these animals had at the beginning of the experiment. It has been reported that young rams during their first exposure to females do not display full libido, implying that the SC tests performed on inexperienced rams may lead to unreliable results. In addition, Price et al. (1991) suggested that prior to testing virgin ram-lambs; they need to gain some sexual experience. With this in mind, it is important to note that all values analyzed corresponded to the average of the three SC tests conducted during the pre-treatment period and the 4th week of the respective phase. Pooling the results of several tests reduced the effect of first exposure to females and learning and maturation of the rams. In addition, a third possibility that could not be discarded might be that the residual effect of the treatment could take longer to disappear than the four-week evaluation residual period observed during the present experiment.

In summary, our results corroborate previous findings that maca consumption improves some mating behaviors in rams. But above all, it demonstrates that this effect is achieved particularly by low sexual capacity rams that will reach high sexual capacity levels, and that maca supplementation has a short residual effect.

CONCLUSIONS

It was concluded that maca supplementation affects males differently, according to their sexual capacity.

IMPLICATIONS

The use of HP rams allows reduction of the percentage of males used in field service and increase the lambing rate of the ewes exposed, which also implies a reduction of the related costs (Lindsay, 1986). According to the results of the present experiment, treated LP rams might increase their ejaculations to the level of HP rams after a short twoweek supplementation period, which represents a low cost and fast way to transform LP to HP animals. Even though it remains to prove that LP maca supplemented rams will performed as well as HP rams in the field, as they did during the SC tests, based on the high correlation between the results of the SC tests and field performance of the rams (Perkins et al., 1992), the applicability of the treatment remains highly promising.

Mean (± SEM) sexual behaviors recorded during sexual capacity tests from hair sheep rams while never exposed to Maca¹, supplemented with 233 mg of dry maca/kg of body weight/day for a four week period², or not supplemented with Maca during four weeks after last supplementation³

| | Sexual capacity | | | | | |
|----------------|----------------------|-------------------|-----------------------|----------------------|-------------------|-----------------------|
| | Low performers | | | High performers | | |
| | Control ¹ | Maca ² | Residual ³ | Control ¹ | Maca ² | Residual ³ |
| Genital sniffs | 4.6 ± 1.3a | 6.4 ± 0.9b | 4.1 ± 1.1a | 3.4 ± 1.0a | 6.8 ± 0.9b | 5.1 ± 1.0ab |
| Nudging | 7.0 ± 3.3a | 16.3 ± 3.4b | 13.3 ± 3.2b | 6.6 ± 3.4a | 18.2 ± 2.3b | 20.6 ± 4.0b |
| Mounts | 9.4 ± 3.4a | 16.7 ± 3.3b | 8.4 ± 0.8a | 11.0 ± 4.1a | 11.1 ± 1.9a | 10.7 ± 1.7a |
| Ejaculations | 1.6 ± 0.2a | 3.0 ± 0.4b | 2.0 ± 0.4a | 3.0 ± 0.4a | 3.0 ± 0.3a | 3.2 ± 0.1a |

^{a, b, c} different superscripts indicate that within the row, values differ (P < 0.05) significantly All values correspond to the average of the three tests conducted at the 4th week of the respective treatment

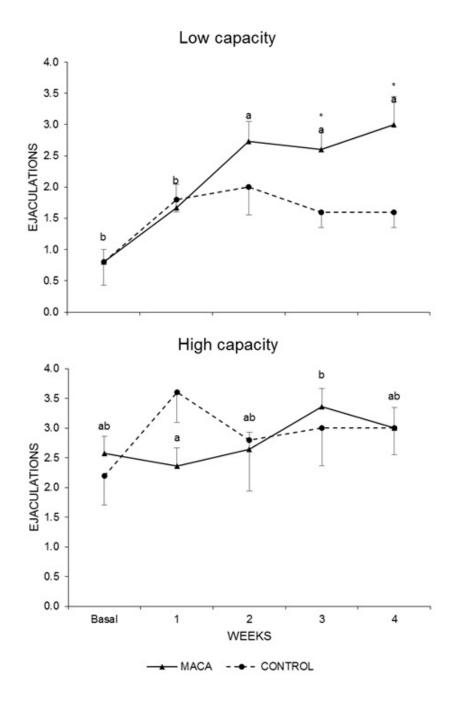


Figure 1 Average (± SEM) number of ejaculations performed weekly by low (LP) and high (HP) performer rams while supplemented with maca in comparison with their controls.

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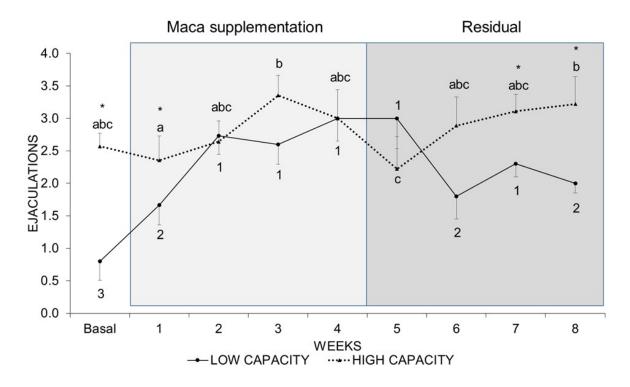


Figure 2

Mean (± SEM) ejaculation pattern from low (LP) and high (HP) performer rams beginning at basal level, during maca supplementation and residual four-week period.

^{a, b, c} different letters indicate weekly values differences (P < 0.05) within High capacity rams; ^{1, 2, 3} different numbers indicate weekly values differences (P < 0.05) within low capacity rams; * represent statistical differences (P < 0.05) at weekly points between low and high performer rams</p>

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