Brief Communication

Aphrodisiac Activity of *Dactylorhiza hatagirea* (D.Don) Soo in Male Albino Rats

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Introduction

The bulbous roots of *Dactylorhiza hatagirea* (D.Don) Soo (Fam. Orchidacea) which are synonymous to the tubers of *Orchis macula* (Orchidaceae) and serve as source of Salep, are used traditionally in Indian subcontinent specially in the Northern region and Nepal as aphrodisiac and sexual stimulant. It is considered as a nutritive and restorative tonic and also as an alternative source of Salep used very commonly in Europe (1).

Materials and Methods

Lyophilized aqueous extract of roots was studied for effect on sexual behavior and spermatogenesis in male albino rats. Wistar strain albino rats of either sex weighing 220-225 g were fed on standard pellet diet and water ad libitum. The animals were housed at room temperature $(24 \pm 2^{\circ}C)$ on a reversed day-night cycle (06:00 h to 18:00 h). Male rats were divided into groups of six each and the extract tested for sexual and aphrodisiac behavior (2). All the animal experimentation was carried out after prior permission from the institutional ethical committee of Dr H.S. Gour University, Sagar (MP), India. Group I (no treatment) served as control. Group II was administered with daily dose of aqueous extract 200 mg kg⁻¹ b.w. orally. Group III was given 0.5 mg kg⁻¹ b.w of suspension of testosterone propionate in arachis oil twice weekly intramuscularly. Twenty-eight days after treatment, body weights of all groups were recorded and animals were sacrificed by cervical dislocation. The testes, prostate and seminal vesicles were removed and weighed. Thin sections of testes of all the groups of animals were cut with the help of a microtome. The sections were fixed in Bouins fixative, dehydrated by varying percentage of ethanol and stained with hemotoxylin and eosin. The sections were studied microscopically for changes in histo architecture or morphology (2,3).

Effect of extracts on behavioral aspects were gauged by evaluation of three different parameters namely, selfexploratory behavior which involved rearing, self-licking and anogenital sniffing. Environmental exploration which comprised of exploration, roaming, climbing and non-self exploratory behavior which included mounting over female, licking, anogenital sniffing. The methodology for scoring was the same as followed by Malmnas and Meyerson (4). Scoring was made giving a value of 0 (no sexual activity), 1 (no interaction, rears and climbs on chamber), 2 (sniffs other rat), 3 self exploratory behavior i.e. grooming and sniffing of genitals, 4 (grooms female rat anywhere), 5 (rears and climbs sexually), 6 (pursues and sniffs other rat, 7 (tries to mount but easily discouraged), 8 (mounts with an integrated deliberate manner, not easily discouraged), 9 (reflex and almost involuntary mount).

The hesitation time was recorded as the time (in seconds) required by the male rat before making an attempt to cross the barrier. In the same way, a scoring for attraction towards female was recorded by a score between 0–5 during an observation period of 15 min. A complete cross of the partition by the male rat each time was given a score of 5 while an attempt to climb was given a score of 2 and disinterest to climb was rated as 0 (3).

There were other sexual parameters like mount, intromission and ejaculatory frequency as well as latency

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which were evaluated, along with penile erection index (PEI) and copulatory rate.

Penile Erection Index = % rats exhibiting erection × Mean number of erections

Coapulatory rate

 $= \frac{Number\ of\ mounts + Number\ of\ intromissions}{Time\ from\ first\ mount\ till\ ejaculation}$

Measurement of testosterone level in blood was undertaken using HPLC (Shimadzu, Japan) with ESD. Male rats were sacrificed by cervical dislocation and blood samples were collected. The method of Bonilla-Jaime (5) was followed with slight modifications.

Results, Discussion and Conclusion

The results of studies undertaken suggest that aqueous extract of *D. hatagirea* causes significant anabolic effect which is comparable to testosterone treatment. Genesis of steroids is one of the causes of increased body and sexual organ weights and an increase in this parameter could be regarded as a biological indicator for effectiveness of the herbal drugs in improving the genesis of steroidal hormones.

Treatment with lyophilized extract of *D. hatagirea* remarkably influenced the behavior of the treated animals, which were more attracted towards female. A 2.5-fold increase in attraction towards female was noticed (P < 0.01) compared to nearly doubled increase in attraction (P < 0.05) in testosterone treated group.

The indulgence of treated animals in copulation increased and the number of bouts increased significantly as well. Standard testosterone-treated group also exhibited greater indulgence than control group and three times increase in bout frequency was observed.

PEI indicates the involvement of nitrous oxide (NO)-based mechanism. An increase in PEI was observed in treated groups; D. hatageria treated animals were superior in this regard (P < 0.01) compared to control group while testosterone treated animals were the next to follow (P < 0.05). Percentage of ejaculating animals was also increased after treatment as compared to control group animals.

The mount intromission and post-ejaculatory latency (PEL) were significantly reduced in extract treated groups. Mount latency time which is an indicator of physical exhaustion during a sexual act was reduced by 36% in D. hatagirea treated and 34% in testosterone-treated group as compared to control group. Intromission (IL) and PEL time was reduced by 36% in D. hatagirea-treated group (P < 0.05) whereas only 17% reduction was observed in testosterone treated group. Reduced IL and PEL have

been correlated with invigoration of endocrine system thereby, resulting in enhanced sexual performance and motivation.

A marked effect of administration of extract as well as testosterone (T) could be observed in male rats. The control level of T in untreated group males was $2.33 \,\mathrm{ng}\,\mathrm{ml}^{-1}$. In extract-treated males, T increased significantly. The mean levels of T increased approximately to $9 \,\mathrm{ng}\,\mathrm{ml}^{-1}$ in the experienced males, and this increase was significantly larger (P < 0.05) than the enhancement observed in the control group.

Dactylorhiza hatagirea is considered as an important aphrodisiac plant in Ayurvedic, Unani literature and is employed to enhance performance as well as to increase vigor and vitality (1). The usage of this herb by practitioners of traditional medicine is frequent, although very little scientific evidence is available for its purported aphrodisiac properties. The observed anabolic activity evidenced by gain in body and organ weights is suggestive of testosterone intervention of the drug extracts. It is likely that the extracts help in improving the testosterone availability to gonads.

Increase in testosterone level has been associated with a moderate but significant increase in sexual desire as well. Clinical data on testosterone also suggest that a slightly increased level of testosterone in adult males results in an increased sexual desire and arousability. There is also sufficient evidence that for peripheral responses in nervous system an increased testosterone level is a must which is not the case in case of CNS activity (6). Therefore, an improved serum testosterone level after administration of extracts could be considered as one of the contributing factors responsible for an overall incremented sexual performance in treated groups. Etiology of erectile dysfunction has also been correlated with low serum testosterone levels. Enhanced levels of testosterone after treatment could be foreseen as a possibility for usage of the herb in treatment of erectile dysfunction as well.

Conclusively, the study validates the effectiveness of herb in improving as well as preventing the functionality of sexual organ as well as substantiates the hype that these plants have aphrodisiac activity and may be helpful in improving the sexual behavior and performance. The results also corroborate the hype that the plant is capable of being nominated as herbal cure for sexual dysfunction. Further studies underpinning the chemical characterization of the herb and their effect on testicular functions could provide substantial evidence in future.

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