Journal of Ethnopharmacology 122 (2009) 579–582

Contents lists available at ScienceDirect

Journal of Ethnopharmacology

journal homepage: www.elsevier.com/locate/jethpharm

Ethnopharmacological communication

# Aphrodisiac properties of *Allium tuberosum* seeds extract

Hu Guohua[a,*], Lu Yanhua[b], Mao Rengang[a], Wei Dongzhi[b], Ma Zhengzhi[a], Zhang Hua[a]

[a] College of Life and Environment Science, Shanghai Normal University, Guilin Road 100#, Shanghai 200234, PR China
[b] State Key Laboratory of Bioreactor Engineering, East China University of Science & Technology, Meilong Road 130#, Shanghai 200237, PR China

ARTICLE INFO

*Article history:*
Received 4 October 2008
Received in revised form 8 December 2008
Accepted 15 January 2009
Available online 7 February 2009

*Keywords:*
*Allium tuberosum*
Seed
Herbal medicine
Aphrodisiac
Sexual behavior

ABSTRACT

*Aim of the study:* In the present study, we examined the effect of *Allium tuberosum* seeds extract upon the expression of male rat sexual behavior, in order to know whether *Allium tuberosum* seeds extract possess aphrodisiac property.
*Materials and methods:* The aphrodisiac activity of *Allium tuberosum* seeds *n*-BuOH extract was investigated in male rats. The extract (500 mg/kg body weight/day) and L-dopa (100 mg/kg body weight/day) were administered orally by gavages for 40 days. Mount latency (ML), intromission latency (IL), ejaculation latency (EL),mounting frequency (MF), intromission frequency (IF), ejaculation frequency (EF) and post-ejaculatory interval (PEI) were the parameters observed before and during the sexual behavior study at day 0, 10, 20, 30 and 40.
*Results:* The *n*-BuOH extract reduced significantly ML, IL, EL and PEI ($p < 0.05$). The extract also increased significantly MF, IF and EF ($p < 0.05$). These effects were observed in sexually active and inactive male rats.
*Conclusions:* Present findings provide experimental evidence that the *n*-BuOH extract preparation of *Allium tuberosum* seeds possesses aphrodisiac property.


## 1. Introduction

Since ancient time, many *Allium* species, such as onion, garlic, leek, and chives, have been used as foods, spices, and herbal remedies in widespread areas of the world, especially in the northern hemisphere. It is well known that the *Allium* genus, with about 500 species, is a rich source of steroidal saponins, alkaloids, as well as sulfur-containing compounds (Hostettmann and Marston, 1995). The scientific name of Chinese chive is *Allium tuberosum* Rottl. ex Spreng (Chinese chive, Liliaceae). Chinese chive is one of the daily edible green vegetables for Chinese. *Allium tuberosum* is distributed all over mainland China and used not only as food but also as medicine. Chinese chive is widely cultivated in China, whose seeds have been reputedly used as a traditional Chinese medicine for treating both impotence and nocturnal emissions (Jiangsu New Medicinal College, 1979). With regard to the constituents of the *Allium tuberosum* seeds, many new and known steroidal saponions, alkaloids and amides have been reported from the seeds (Sang et al., 1999a,b, 2000a,b,c, 2001a,b, 2002, 2003; Zou et al., 2001). In order to clarify its bioactive compounds, the chemical constituents of the seeds of *Allium tuberosum* were analyzed and studied at our research systematically. We reported the fatty acid compositions of the oil of ten kinds of *Allium tuberosum* seed (Guohua Hu et al., 2005). We also reported the chemical compositions of *Allium tuberosum* seed (Guohua Hu et al., 2006a,b, 2009). However, no experimental investigations have been carried out on the diverse pharmacological properties of this traditional remedy.

There are numerous reports of aphrodisiac activity attributed to plants (Emilia Nocerino et al., 2000; Islam et al., 1990; Subramoniam et al., 1997; Noumi and Zollo, 1998; Bo Lin Zheng et al., 2000; Gauthaman et al., 2002; Carro-Juaĭreza et al., 2004; Ramachandran et al., 2004). *Allium tuberosum* seeds have been reputedly used as a traditional Chinese medicine for treating both impotence and nocturnal emissions (Jiangsu New Medicinal College, 1979). These claims are based largely on subjective opinion rather than scientific observation. In the present study, we examined the effect of *Allium tuberosum* seeds extract upon the expression of male rat sexual behavior, in order to evaluate the possibility that this plant extract might possess aphrodisiac property. Experimental investigations on the diverse pharmacological properties of this plant seed are being actively pursued in our laboratory. The present study was aimed to search the active constituents from *Allium tuberosum* for treating both impotence and nocturnal emissions. The study evaluated the effect of this plant seed *n*-BuOH extract on the male sexual behavior.

* Corresponding author. Tel.: +86 21 64322933; fax: +86 21 64322142.
*E-mail address:* hgh114226@sina.com (H. Guohua).

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doi:10.1016/j.jep.2009.01.018

## 2. Materials and methods

### 2.1. Preparation of Allium tuberosum seeds extract

*Allium tuberosum* Rottl. seed ("*791*", "*791*" is furthest widely cultivated in China in all variety) were purchased from Shanghai Shun-Feng-Tang Group, Shanghai, China in July 2004. A voucher specimen (No. 200305), identified by professor Dr. Dongzhi Wei, was deposited at Institute of Biochemistry, East China University of Science and Technology, Shanghai, 200237, China.

The powdered seeds of *Allium tuberosum* (10.0 kg) were extracted successively with petroleum ether (petroleum ether: seed, 10:1, v/w) 2× and 95% EtOH (EtOH: seed, 10:1, v/w) 3×. The combined EtOH extracts were filtered and evaporated to dryness at a low temperature under reduced pressure in a rotary evaporator. After evaporation of ethanol, the residue was suspended in water and then extracted successively with petroleum ether (petroleum ether: extract, 1:4, v/v) 3×, EtOAc (EtOAc: extract, 1:4, v/v) 3× and *n*-BuOH (*n*-BuOH: extract, 1:4, v/v) 3×. The *n*-BuOH extract was evaporated to dryness at a low temperature under reduced pressure in a rotary evaporator. The *n*-BuOH extract (0.8% (w/w) yield) was suspended in distilled water and used for studying bioactivity.

### 2.2. Animals

Inbred adult Wistar albino rats of either sex were used for the study. The animals were fed with standard animal feed and water. The animals were housed at a temperature of 25 ± 1 °C with a reversed light dark cycle and relative humidity of 50–55%. The study was performed as per the protocols and recommendation of the Institutional Animal Ethics Committee, Science and Technology Commission of Shanghai Municipality. All animal experiments were conducted in accordance with the NIH guide for the care and use of laboratory animals (NIH Publication No. 80-23; revised 1978).

Male rats ($n = 5$/group) were trained for sexual experience. To provide sexual experience, each male rat was allowed 30 min exposure to a female rat in behavioral estrous, several days before testing for copulatory performance in a transparent arena. The animals were tested three times over a 10-day period for copulatory behavior and divided into active and inactive groups (Ramachandran et al., 2004). Sexually active animals were divided into control, L-dopa-treated (100 mg/kg body weight) and *Allium tuberosum seeds*-treated (500 mg/kg body weight) groups. The animals that did not show any sexual interest during training were considered the inactive group that was administered *Allium tuberosum seeds* (500 mg/kg body weight). Female rats were ovariectamised using a standard procedure (Bures et al., 1983). They were allowed to recover from the surgery for 10 days. They were brought into estrous by the administration of a single subcutaneous dose of 2 μg/kg body wt. of estrogen benzoate (Sigma Chemical Co., St. Louis, MO, USA) and 500 μg/kg body wt. progesterone (Sigma Chemical Co., St. Louis, MO, USA) 48 and 6 h before the copulatory study.

### 2.3. Sexual behavior study

The following guidelines (Hart et al., 1983; Islam et al., 1990) were followed in the study; males were kept individually whereas females were kept in groups; training of each male for 15 min at a time was performed until sexual behavior was elicited and when the behavior was noticed, males were exposed to receptive females (one male with five females); repeated training to overcome the lack of sexual response in the presence of observers; the study was conducted in a single observation cage (0.8 m × 1.0 m × 1.2 m) in a silent room under dim red light. Any jerking movement of the mating area was avoided to enable the rats to chase each other; and cleaning of the mating area was performed after each trial, since the urine trails left by one rat might alter the sexual behavior of the next rat. They were fed commercially available food twice each day, and water was always available. All sexual behavior tests were observed for 4 h and performed from 09:00 to 13:00 h.

*Allium tuberosum* seeds (500 mg/kg body weight/day) and L-dopa (100 mg/kg body weight/day) in distilled water were administered for 40 days orally by gavage. L-Dopa served as standard (Greenburg, 1971; Ananthakumar et al., 1994). The control group received 1 ml saline. Each group consisted of six animals (one male and five females). The female was then introduced into the chamber and the following sexual behavior parameters were recorded: (a) mount latency (ML): the time interval between the introduction of the female and the first mount by the male; (b) intromission latency (IL): the interval from the time of introduction of the female to the first intromission by the male (characterized by pelvic thrusting and springing dismount); (c) ejaculation latency (EL): the time interval between the first intromission and ejaculation (characterized by longer, deeper pelvic thrusting and slow dismount followed by a period of inactivity); (d) mount frequency (MF): the number of mounts observed in 30 min; (e) intromission frequency (IF): the number of intromissions observed in 30 min; (f) ejaculation frequency (EF): the number of ejaculations observed in 30 min; (g) post-ejaculatory Interval (PEI): the time interval between ejaculation and the first intromission of the following series.

### 2.4. Toxicity study

Two groups of six male and six female rats each were used. The *n*-BuOH extract (2 g/kg body weight) was administered orally by gavage to the rats in the test group. The rats in the control group each received 1 ml of tap water. Behavioral parameters such as convulsions, sedation, hyperactivity, grooming and accelerated breathing were observed. The animals were observed continuously for 1 h for any behavioral changes or death, then intermittently for 6 h and finally 24 h after drug administration.

### 2.5. Data analysis

Pair-wise statistical comparisons between control and treated groups were done with Student's *t*-test. Mean differences were considered statistically significant if $p < 0.05$.

## 3. Results

The observations of the sexual behavior study are presented in Table 1. The present study confirmed the claims of *Allium tuberosum seeds n*-BuOH extract as an aphrodisiac agent. Sexually active and inactive animals showed increased and improved sexual performance, when *Allium tuberosum seeds* extract (500 mg/kg body weight) was administered for a period of 30–40 days. *Allium tuberosum seeds n*-BuOH extract reduced ML, IL, EL and PEI significantly in both active and inactive male rats. *Allium tuberosum seeds n*-BuOH extract also increased MF, IF and EF significantly in both active and inactive male rats. All these effects were observed on the 30th and 40th days of the study. The MF and EF was significantly altered in *Allium tuberosum* seeds-treated active rats (20th day), whereas L-dopa did not alter these parameters on the 20th day. The MF and IF were significantly altered in *Allium tuberosum seeds*-treated inactive rats (20th day), whereas L-dopa did not alter these parameters on the 20th day. The EL and PEI was significantly altered in L-dopa treated active rats (20th day), whereas *Allium tuberosum* seeds for both active and inactive male rats did not alter these parameters on the 20th day. *Allium tuberosum* seeds for both active and inactive male rats and L-dopa did not alter any of all these parameters on the 10th day. In toxicity test to determine any acute toxicity of *n*-BuOH extract, no mortality or changes in behavior were observed in the