

Garden Cress Seed Could be A Factual Galactagogue

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ABSTRACT:

BACKGROUND:

Trivial number of books, concerning traditional medicine, had mentioned a galactagogal role of Garden cress seeds. Others ignore that. This controversy, in addition to the steroid (family of sex hormones) contents of the seeds, directed us to evaluate the role of this herb in mammogenesis and lactogenesis.

METHODS:

Twelve parameters were used to assess the effect of Garden cress seeds on the mammary gland of young adult virgin rats. These parameters comprise gross assessment, histological examination (routine/haematoxylin and eosin stain and special stain/ PAS), enzymatic histochemical study (alkaline phosphatase, acid phosphatase and lipoprotein lipase cytochemical localization), biochemical estimations (hormonal assay of FSH, LH, prolactin, estrogen and progesterone) and morphometrical measurements.

RESULTS:

All parameters significantly exhibited a strong mammotrophic and lactogenic effects of Garden cress seeds on the non-primed mammary gland of adult virgin rat.

CONCLUSION:

Garden cress seeds are most probably a real galactagogue and might be useful in induction of lactation.

KEY WORDS: Garden cress (*Leptidium sativum*) , Galactagogue , Mammary gland.

INTRODUCTION:

Garden cress (Family name = Cruciferae, Scientific name = *Leptidium sativum*, Arabic name = Rhishad) is a 20 – 40 cm high herb with a glabrous bluish bloom. The stem is erect, round and branched.

The leaves are light green, thin and have dentate to prickly segments⁽¹⁾.

The medicinal parts are the seeds and the fresh or dried herb harvested during or shortly after flowering season^(1,2). The herb is grown worldwide^(1, 3,4). Their main chemical constituents are glucosinolate (chief components glucotropeolin), ascorbic acid (vitamin C), vitamin B, and cardiac steroids (cardenolides). Seeds are known, by the practitioners of traditional medicine, as tonic, aphrodisiac and stimulant. They are given as a decoction or infusion in hiccup, dysentery, diarrhea, skin diseases and as a galactagogue^(1,2,3,4).

No health hazards or side effects are known in conjunction with the proper administration of designated therapeutic dosage^(1,4).

Regarding the lactogenic activity of the Garden cress seeds, some references mentioned it⁽²⁾ but others didn't^(1, 3,4). Therefore, this investigation was directed to cast a light on the capability of these seeds in mammogenesis and /or lactogenesis induction.

MATERIALS AND METHODS:

Twenty-six adult female virgin Norway rats (*Rattus norvegicus*) were used in this study.

Their age was two months \pm one week and their weight (140 – 200 g).

Animals were kept at a constant temperature between 20 – 24 °C. They were housed in wire-meshed stainless steel cages. The light / dark cycle was kept (12 hr. /12 hr.). Rats were fed ordinary rat's pallet diet and they had continuous free access to food and water. Care was taken to avoid unnecessary stress as noise and cage crowding.

Rats were divided into twenty experimental (Garden cress-treated) and six controls.

Garden cress seeds were cleaned from artifacts and cleared from other seeds, and then standardized in the "Iraqi National Herbarium" and in the "Dept. of Pharmacognosy/ College of Pharmacy/ Univ. of Baghdad". Seeds were ground in a coffee grinder.

The seeds powder was suspended in distilled water and given via oro-gastric tube.

Each experimental rat was given 1.6 mg seeds powder /gm body weight /day (the whole dose was suspended in 4ml distilled water) for fourteen days.

Control rats received 4 ml distilled water as a placebo, under identical conditions.

After fourteen days, animals were sacrificed. Mammary gland and liver (to be used as positive

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control for enzyme histochemical procedures) samples were obtained.

Blood samples were taken, via intra-cardiac puncture (to be used for hormonal assay).

Mammary gland specimens, of each sacrificed rat, was processed for haematoxylin and eosin (HE) stain⁽⁵⁾, PAS stain^(6,7) and histochemical localization of alkaline phosphatase (AKP)⁽⁸⁾, acid phosphatase (ACP)⁽⁹⁾ and lipoprotein lipase (LPL)⁽¹⁰⁾.

Following examination and morphometrical assessment, tissue sections were photographed (using SC 35 camera attached to Olympus light microscope).

Data collected were analyzed using the computer facility with the available software statistical packages of "Statistical Packages for Social Sciences", version 10.0 (SPSS 10.0).

The significance of differences among the quantitative variables of three subgroups was assessed using one-way analysis of variance (ANOVA). Results between two groups were analyzed using "Student's T-test"⁽¹¹⁾.

RESULTS:

During the treatment course, rats were maintaining good general health and well-being as assessed by their normal activities and rather sufficient food intake. However, changes in body weight, between experimental and control rats, were not significant. Grossly, controls' mammary glands were small-sized with small nipples. Cross section of mammary gland revealed white colored tissue. On the contrary, Garden cress treated rats' mammary gland exhibited a great enlargement in its size with deep purple cross sections. Histological sections (HE stained sections) of mammary gland of controls showed small lobules scattered among huge amount of adipose and connective tissues. Each lobule consisted of few secretory tubules (Fig. 1-A). Interlobular ducts were found within interlobular connective tissue.

On the other hand, experimental rats' mammary gland arrayed relatively very large lobules, which were seen fully packed by a large size and widely dilated acini. Acinar lumen was filled with pink homogenous material that contains lipid droplets (milk secretion) (Fig. 1-B). Interlobular ducts were filled with milk secretion too.

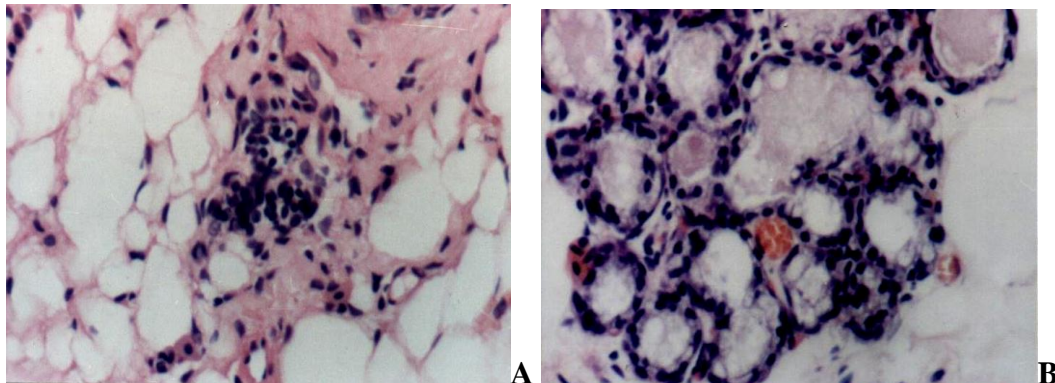


Figure 1: HE stained mammary gland sections of control (A) and Garden cress-treated (B) rats. Note the great dilatation of the acini in the experimental group. (x400)

PAS stained sections of mammary gland of control group demonstrated histological features similar to those stained with HE stain.

However, various cell population of mammary tissue and their cytoplasm, especially those of connective tissue and related residents such as macrophages, were easily visualized by this stain.

Likewise, experimental rat's mammary glands gave identical features of experimental rat's mammary gland stained by HE stain, except the appearance of strong positive PAS stain in the homogenous part of milk secretion within acinar and ductal lumens (Fig. 2-A and 2-B).

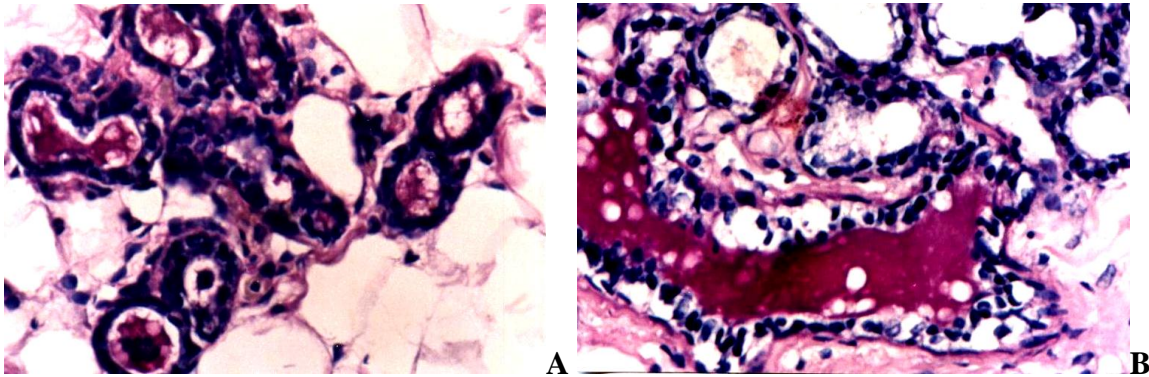


Figure 2: PAS stained sections of Garden cress-treated rat's mammary gland, exhibiting strong positive reaction in milk secretion within acini (A) and duct (B). (x400).

Generally, mammary glands of all control groups disclosed a weak to trivial enzymatic activities (Fig. 3-A, 4-A and 5-A). While, mammary glands of all

experimental rats exposed striking increase in the intensity of their final reaction products (Fig. 3-B, 4-B and 5-B).

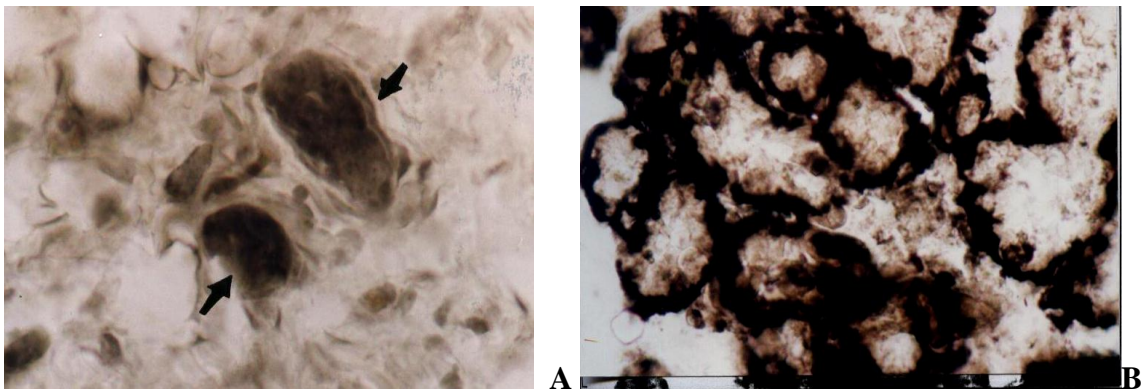


Figure 3: AKP histochemical activity of control (A) and experimental (B) rat's mammary gland. (x400)

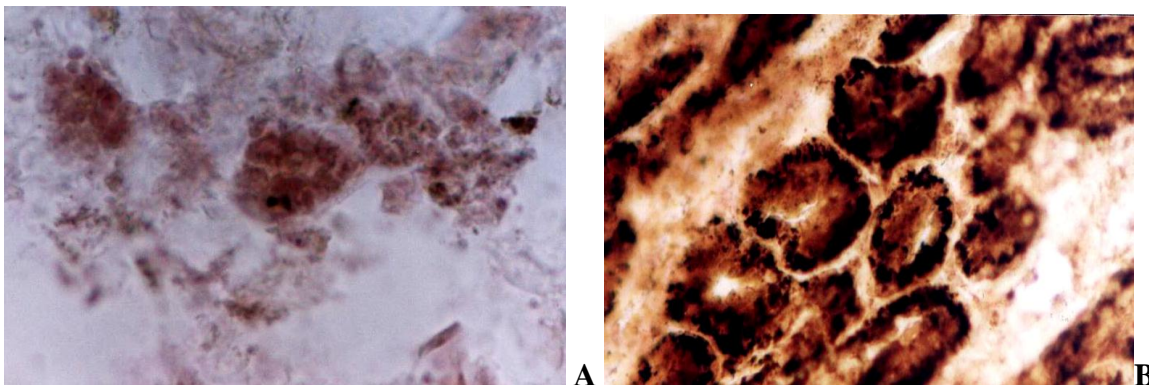


Figure 4: ACP final reaction product in rat's mammary gland of control (A) and Garden cress-treated (B) rats. Note the intense ACP activity in the Acinar epithelium of the experimental rat's mammary gland. (x400)

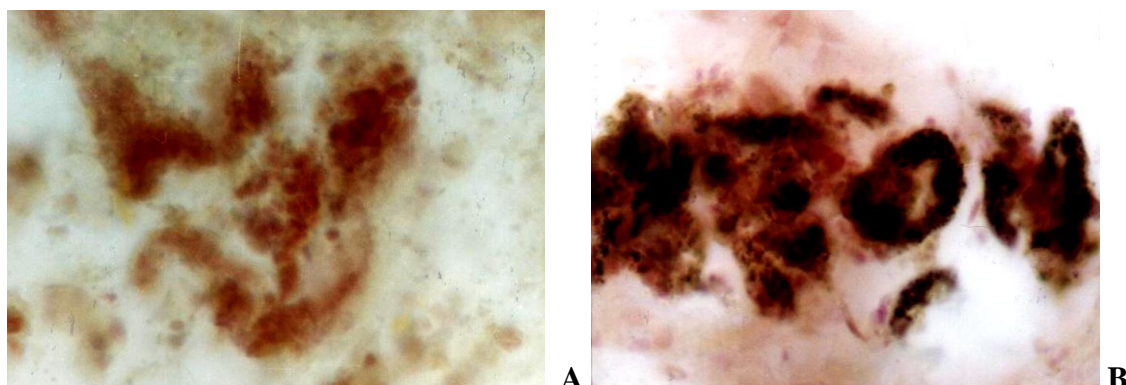


Figure 5: LPL activity in the control (A) and Garden cress-treated (B) rat's mammary gland. (x400)

Data of hormonal studies were obtained after synchronization of estrus cycle. They are summarized in table (1), while those of morphometrical measurements are listed in table (2).

Table 1: Hormonal assay in control and experimental rats.

Hormones	Control	Garden cress
FSH	4.63±0.36	4.78±7.52
LH	6.00±0.00	10.36±0.13*
Prolactin	4.50±0.31	9.00±0.14*
Progesterone	7.11±0.19	7.75±0.13*
Estrogen	72.90±89.30	78.65±0.35*

Data are expressed as mean ± standard deviation

All hormones are measured by ng / ml

* = significant at P < 0.01

FSH = follicular stimulating hormone, LH = luteinizing hormone

Table 2: Morphometrical analysis of mammary gland items in control and experimental rats, using eye piece microtome (10x40).

Item	Control	Garden cress
Acinar diameter (μm)	23.01±1.49	44.35±4.31*
Acinar epith. Thickness (μm)	12.16±1.82	7.71±0.31*
No. of nuclei / acinus	14±1.16	16±2.19**

Data are listed as mean ± standard deviation

* = significant at P < 0.01

** = significant at P < 0.05

epith. = epithelium

Acinar diameters were measured by assessment of the greatest dimension of the active lobes in the mammary gland i.e. resting lobules were ignored in all sections.

DISCUSSION:

Maintenance of good general health and well being without weight gain in experimental rats, seen in this study, could be attributed to their vitamin B and C content, antimicrobial influences, antiviral effects and / or diuretic action^(1,12).

Gross examination changes noticed in Garden cress seeds-fed rats declare signs of augmentation of mammary gland activity. Histologically, control rat's mammary gland revealed a resting state.

This finding parallels the age and physiology of an adult virgin rat described elsewhere^(13,14).

On the contrary, Garden cress-treated rat's mammary glands (HE stained) exhibited features of an active (lactating) gland, i.e. this study showed that garden cress seeds have the ability of activation of mammary gland and enhancement of milk secretion into acinar lumen. Such investigation was not previously worked out (Medline reviewed).

PAS stained sections of control and experimental rats paralleled and confirmed those stained by HE stain. However, the strong positive PAS reaction in the milk secretion (seen within the lumen of acini and ducts of mammary glands of experimental rats) could be attributed to the presence of glycoprotein and / or 1,2-glycols of high molecular weight in it⁽¹⁵⁾. In addition, the glycoprotein together with the large lipid droplets, visualized in milk secretion, may point to the fact that there is a synthesis of the three main constituents of milk (carbohydrates, proteins and lipids). Consequently, this outcome discloses the occurrence of real and complete process of lactogenesis. The weak enzymatic activity, recorded in the mammary glands of control rats, also parallels their physiological states.

While the expressive increment in the enzymatic (AKP, ACP and LPL) activities, in Garden cress seeds-treated rat's mammary gland, indicated the occurrence of lactogenesis too. It had been found that AKP increase late in pregnancy and early lactation

⁽¹⁶⁾. Moreover, it had been suggested that this enzyme participates in the milk synthetic process in the mammary gland, especially in lactose synthesis⁽¹⁷⁾. Concerning ACP, it had been recorded that the activity of this enzyme was significantly higher during lactation than pregnancy⁽¹⁸⁾.

Furthermore, it had been assumed that it may play a role in controlling the secretory processes in the mammary epithelium, storage functions and autophagic digestion⁽¹⁹⁾.

Lastly, the intense LPL activity, elicited in this study in the experimental rat mammary gland, coincides with its function of provision of fatty acid from the plasma in preparation for milk fat synthesis^(20,21). Hormonal outcome, specially the highly significant rise in serum prolactin in experimental rats, accords the histological and histochemical results, found in this investigation, and agrees with a state of lactating mammary gland^(14, 22).

Finally, morphometrical analysis approves all earlier parameters. The highly significant increase of acinar diameter, in experimental rats, is most probably reflecting the active condition of mammary gland. Similar changes were recorded during pregnancy and lactation⁽¹⁷⁾. The significant rise in number of nuclei per one acinus accords the former outcome. It had been found that the number of nuclei in the average acinus cross section was relatively constant during pregnancy and increased approximately by two folds at the time of parturition⁽¹⁷⁾.

In conclusion, all used parameters reflect the clear lactogenic role of Garden cress seeds.

Further studies should be directed towards ultrastructural studies on mammary tissue after induction of lactation using these seeds and comparison of these seeds with the harmful hormones (estrogen and progesterone) and drugs (metochlopramide and chlorpromazine) which are used commonly in induction of lactation.

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