



Formulation, standardization, organoleptic evaluation of natural galactagogue mix and its impact on prolactin levels

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ABSTRACT

To assess the effectiveness of naturally prepared galactagogue mix on breast milk production and prolactin levels in postnatal mothers. Natural Fenugreek and Garlic based Galactagogue mixes were formulated and standardized. Organoleptic evaluation of the same mixes was also conducted by a ten panel members. Followed by this, 30 mother-infant pairs were assigned into 3 groups. Group I (n=10) consisted of mothers who were receiving galactagogue mix containing fenugreek every day, Group II (n=10) consisted of mothers who were receiving galactagogue mix containing garlic every day and Group 3 (n=10) were assigned as control group. The intervention was carried out for seven days, the baseline and post treatment serum prolactin levels of the mothers were determined using a standard protocol. Organoleptic scores of Garlic galactagogue mix was superior than Fenugreek galactagogue mix attributing to its bitter taste. The mean measured serum prolactin levels of the mothers who received galactagogue mixes were higher than the control group ($p < 0.05$). Galactagogue mixes using natural ingredients especially fenugreek seeds were found to be highly acceptable and supplementation of these galactagogue mixes seems to be useful for enhancing breast milk production.

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Introduction

Breastfeeding is considered the optimal source of nutrition for infants from birth to 1 year and is supported by the American Academy of Paediatrics (AAP) and the World Health Organization^{1, 2}. Breast milk is the natural food for the infant and it is "species specific".

The American Academy of Paediatrics recommends exclusive breastfeeding for a minimum of 4 months but preferably for 6 months. Breastfeeding should be continued, with the addition of complementary, at least through the first 12 months of age and thereafter as long as mutually desired by mother and infant³. The recommendation for feeding full term and preterm infants with human milk arises because of its acknowledged benefits to infant nutrition, gastrointestinal function and host defence, and the potential beneficial influence on neuro development and chronic diseases of childhood².

Infants who are exclusively breastfed for six months experience less morbidity from gastrointestinal infection than those who are missed breastfed as of three or four months, and no deficits have been demonstrated in growth among infants. Moreover, the mothers of such infants have more prolonged lactational amenorrhea and that the available evidence demonstrates no apparent risks in recommending, exclusive breastfeeding for the first six months of life⁴. However, for normal infants too, exclusive breastfeeding has not been possible in certain households as mother is employed or for cosmetic and social reasons. Similarly, some mothers lack in milk production for various reasons that include poor nutrition, hormonal problems, parity, gravid, age and other associated problems. Yet, there has been evidence that some food products may enhance lactogenesis. Such foods are known as galactagogues

Galactagogues are substances or medications believed to stimulate initiation, maintenance, and augmentation of maternal milk production⁵⁻⁸. Galactagogues act by increasing the prolactin secretion which in turn the milk production. They also work psychologically and have a marginal effect on milk production. Suckling is the best lactagogue exercise. Garlic is commonly used galactagogue food. Fenugreek (*Trigonella foenum-graceum*), an herb from the pea family, is the most commonly used herbal galactagogue foods in the world^{5, 9}.

Although the exact mechanism of action is still unknown, the fenugreek herb may increase milk flow by its phytoestrogens and diosgenin contents. In terms of milk production, fenugreek is suggested to stimulate sweat production, which would enhance milk secretion as the breast is a kind of sweat gland. Fenugreek is listed as GRAS (generally regarded as safe) by the U.S. Food and Drug Administration⁵⁻⁹.

Objectives

1. To formulate and standardize fenugreek and garlic based galactagogues mixes.
2. To evaluate the organoleptic characteristics of the two galactagogues mixes.
3. To assess the Knowledge, Attitude and Practices regarding galactagogues usage and milk production among the selected nursing mothers.
4. Assess the prolactin levels of nursing mothers before and after intervention with the selected galactagogue mixes in both experimental and control groups

Materials and methods

Population of the study

The population included the mothers who had delivered babies in the maternity wards of Kovai Medical Centre and

Hospitals, Coimbatore, India. Postnatal mothers selected were capable of breastfeeding but the PROLACTIN secreting level was poor. Ethical approval was obtained from human ethics committee of Kovai Medical Centre & Hospitals Human Ethics Committee.

Sample Size And Sampling Technique

The sample size for study was 30 mother-infant pairs. 10 pairs were included in each group. The groups were categorized as Experimental – fenugreek, Experimental – garlic and the third one as Control group who were fed with any galactagogue mixes. The samples were chosen according to the inclusion and exclusion criteria through purposive sampling.

Criteria for selection of samples

The following criteria were used for selecting the samples:

Inclusion criteria:

For infants

- ▲ Full term and healthy neonates

For mothers

- ▲ Having been willing to exclusively breastfeed their infants
- ▲ Having not enough secretion of breast milk without any abnormalities of nipples - pain, engorgement, mastitis, cracks etc.,
- ▲ Having consented to follow-up visit
- ▲ Having been consented to check the serum prolactin levels before and after the seventh day following the nutritional intervention

Exclusion criteria:

For infants

- ▲ Infants with low birth weight, low APGAR scores and intrauterine growth retardation
- ▲ Infants with any illnesses or congenital abnormalities

For mothers

- ▲ Mothers who had chronic illness such as diabetes, hypertension, bronchial asthma and any allergies
- ▲ Mothers with any breast problems such as inverted nipples, mastitis, engorgement, cracks etc.,
- ▲ Mothers who had a history of smoking, alcohol, or any drug use meant for improving breast milk production

Phase I

Formulation, Standardization and Organoleptic evaluation of selected Galactagogue mixes

Two galactagogue mixes made out of Sprouted Ragi, Oat meal, Coconut powder, Jaggery, Turmeric powder with Fenugreek or Garlic were selected, formulated and standardized for the study.

Standardization of the galactagogue mixes

Fenugreek and Garlic galactagogue mixes were standardized with different proportions and finally the best to the knowledge of the nursing mothers and the researchers, a pilot evaluation was carried out with ten nursing mothers who were selected purposively. The physical characteristics were also observed after preparing the mix as porridge. Alas, the following proportions of the ingredients were considered for experimentation.

Two variations of galactagogue mix were prepared with the following ingredients:

Fenugreek- galactagogue mix:

Sprouted, dried and powdered Ragi flour(60gm), Oat meal (20gm), Desiccated coconut powder(10gm) , Jaggery(20gm), Fenugreek (20gm), Turmeric (200gm), Water(200ml)

Garlic- Galactagogue Mix

Sprouted, dried and powdered Ragi flour(60gm), Oat meal (20gm), Desiccated coconut powder(10gm) , Garlic(30gm),

Turmeric(2gm), Water(200ml), Curds/Buttermilk(100ml), Salt(to taste), Jeera powder(a little), Coriander leaves(a little)

Organoleptic evaluation

A ten member panel (10 females) comprising of dieticians and nurses from the evaluated the samples using the 5 points hedonic scale method: 5 (excellent) to 1 (very poor). Evaluation of the galactagogues immediately after preparing. Sensory testing was done on both the galactagogues. Each panelist was presented with 6 coded randomized samples. Each sample was coded with three random digit numbers and the positions of the samples were randomized. Panelists were seated in individual customized sensory booths. Each sample was replicated twice. The average scores were analysed by ANOVA.

Questionnaire

Part A

The demographic characteristic of the babies consists of the sex of the baby, gestational age and birth weight of the baby. The mothers' demographic data consists of age, type of delivery, parity of mother, educational status and economical status.

Part-B

The observational checklist for selected bio physiological and anthropometric parameters of breastfeeding mothers are height, weight, BMI and serum prolactin levels and their babies are birth weight, length, chest circumference, head circumference, mid arm circumference.

(These data obtained out of Part I and Part II have been analysed and the results have been published separately)

Part-C

A **KAP** questionnaire consisting questions to gather data regarding the knowledge, attitudes and practices of breastfeeding mothers towards the benefits of breast milk, breastfeeding and nutrition.

Part -D

The observational checklist for dietary pattern of breastfeeding mothers included food frequency and food preferences.

Phase II

Analysis Of Prolactin Levels

Prolactin levels were determined individually before the commencement of intervention with galactagogue mix. Similarly the same was estimated after the study period.

The serum prolactin was estimated by the **Electrochemiluminescence Immunoassay "ECLIA" - Roche Elecsys and MODULAR ANALYTICS E170 (Elecsys module) immunoassay analyzers.**

Results And Discussion

Phase I

Standardization of formulated Galactagogue mix Recipes

Fenugreek- Galactagogue Mix

Ingredients	Quantity
Sprouted,dried and powdered Ragi flour	60 gm
Oat meal	20 gm
Desiccated coconut powder	10 gm
Jaggery	20 gm
Fenugreek	20 gm
Turmeric	2 gm
Water	200 ml

Method:

Mix sprouted,dried and powdered ragi flour, oat meal, desiccated coconut powder, jaggery, fenugreek and turmeric powder in a heavy bottomed pan and add 200ml water and mix thoroughly. Now this mixture should be boiled for 2 minutes with constant stirring. Serve hot with chopped nuts.

Garlic- Galactagogue Mix

Ingredients	Quantity
Sprouted,dried and powdered Ragi flour	60 gm
Oat meal	20 gm
Desiccated coconut powder	10 gm
Garlic	30 gm
Turmeric	2 gm
Water	200 ml
Curds/Buttermilk	100 ml
Salt	to taste
Jeera powder	a little
Coriander leaves	a little

Mix sprouted,dried and powdered ragi flour, oat meal, desiccated coconut powder, ground garlic, turmeric powder and salt in a heavy bottomed pan and add 200ml water and mix thoroughly. Now this mixture should be boiled for 2 minutes with constant stirring. Remove from fire and allow the mixture to cool. Add curds or buttermilk and mix thoroughly. Serve cold, garnished with jeera powder and chopped coriander leaves.

Table 1 Formulation, Standardization And Organoleptic Evaluation Of Selected Galactagogue Mixes

Two galactagogue mixes made out of Sprouted Ragi, Oat meal, Coconut powder, Jaggery, Turmeric with Fenugreek or Garlic were selected, formulated and standardized for the study.

Table 1. List Of Galactagogue Mixes

S.NO	FOOD ITEMS
1	Fenugreek -Galactagogue mix
2	Garlic -Galactagogue mix

Table 2. Organoleptic Qualities Of The Selected Galactagogue Mixes

Name of the Recipe	Mean Scores (10 Judges)				
	Colour and Appearance (5)	Texture (5)	Flavour (5)	Taste (5)	Overall Acceptability (20)
Fenugreek - Galactagogue mix	5	5	4	3	17
Garlic - Galactagogue mix	5	5	5	5	20

Acceptability of the selected food Galactagogue mix recipes (Test foods)

Table 1(b) gives the mean scores obtained by the selected galactagogue mix preparations. The criteria studied were colour and appearance, texture, flavour, taste and overall acceptability. The score of the overall acceptability criteria were maximum, 17 for Fenugreek -Galactagogue mix and 20 for Garlic-Galactagogue mix. The overall acceptability of the Garlic-Galactagogue mix registered excellent score (20 out of 20) showing the highest Organoleptic quality, but the score for Fenugreek-Galactagogue mix was only 17 out of 20 attributing to its slight bitter like taste and flavour. Though the colour of the sprouted ragi used for the mixes were appearing brownish black in colour, still it was highly considered to be appealing. These results indicate that all the two items were highly acceptable.

Table 3. Pearson Correlation of Knowledge, Attitude and Practice in breast feeding mother among the groups

	Knowledge	Attitude	Practice	S/NS
Knowledge N	1 30	0.247	0.083	NS
Attitude N	0.247 30	1	0.001	S*
Practice N	0.083 30	0.001	1	S*

The 2-tailed significant values between correlations of knowledge, attitude and practices using the Pearson Correlation

among all the three groups, were 0.247 for Knowledge to attitude and 0.664 for knowledge to practice and 0.001 to attitude for practice. However, the Pearson correlation test showed there was a significant difference among all the three groups. Thus, it is conversely understood that there was significant difference at $P < 0.001$ level, between the attitude and practice of the different groups of breast feeding mothers.

Phase II**Table 4. One way ANOVA of mean baseline Prolactin levels among the groups**

SL NO	Groups	N	Mean $\pm$ SD	S/NS (P<0.05)
1	Fenugreek	10	116.64 $\pm$ 40.71	0.993 NS
2	Garlic	10	117.91 $\pm$ 89.86	
3	Control	10	120.04 $\pm$ 59.15	
	Total	30	118.20 $\pm$ 64.10	

The above table no. 5 depicts the mean $\pm$ Standard deviation of baseline mean Prolactin levels of all the three groups namely fenugreek, Garlic and Control groups. However, the one way ANOVA test showed that there was no significant difference among all the three groups. Thus, it is conversely understood that there was no much difference between the initial mean Prolactin levels of the lactating mothers. Even, Duncan's Mean Range Post Hoc Tests of the mean Prolactin levels revealed that there was no significant difference between the baseline initial mean Prolactin levels for groups in homogenous subsets.

Hence, it is understood that all the three groups were not having a significant difference in their initial mean Prolactin levels.

Table 5. One way ANOVA of final mean Prolactin levels among the groups

SL NO	Groups	N	Mean $\pm$ S D	S/NS (P<0.05)
1	Fenugreek	10	236.98 ^a $\pm$ 59.36	0.013* S
2	Garlic	10	226.74 ^a $\pm$ 100.98	
3	Control	10	142.49 ^b $\pm$ 48.69	

* Significant at $P < 0.05$

Study was conducted among 30 breast feeding mothers and infant pairs,

- 10 mothers-infant pairs were included in control group
- 10 mother-infant pairs for experimental-fenugreek group
- 10 mother-infant pairs for experimental-garlic group

Demographic characteristics of breastfeeding mothers included in the study belong to the age group of 25-30 years which constituted about 40% in control group, 70% in experimental-fenugreek group and 40% in experimental-garlic group.

The pre- interventional mean prolactin levels of postnatal mothers of the control group was 120.04 ng/ml and the initial mean PRL of experimental fenugreek and garlic group were 116.64 ng/ml and 117.91 ng/ml respectively.

The post-test mean of the prolactin level of postnatal mothers of the control group was 141.49 ng/ml and the final mean PRL of experimental fenugreek and garlic group were 226.74 ng/ml and 236.98 ng/ml respectively.

The above table no. 5 depicts the mean $\pm$ Standard deviation of final mean Prolactin levels of all the three groups namely fenugreek, Garlic and Control groups. The mean prolactin level is comparatively higher in the experimental groups and lower in control group.

However, the one way ANOVA test showed that there was a significant difference among all the three groups. Thus, it is conversely understood that there was significant difference between the final Prolactin levels of the early postnatal lactating

mothers at the level of $p < 0.05$. Thus, further looking at the mean prolactin values of experimental groups which are quite higher than the Control groups and with the inferential statistics computed proved that there was a significant difference that makes to understand that such an increment could be due to the supplementation of galactagogue mixes. This result also correlates with a study done by Turkylmaz et al., 2011, in which they have proven that by giving herbal tea containing fenugreek as galactagogue agent improved the breast milk production in the early postnatal period.

Discussion

More than two decades of research have established that breast milk is perfectly suited to nourish infants and protect them from illness. Breast-fed infants have lower rates of hospital admissions, ear infections, diarrhoea, rashes, allergies, and other medical problems than bottle-fed babies. The primary benefit of breast milk is nutritional. Human milk contains just the right amount of fatty acids, lactose, water, and amino acids for human digestion, brain development, and growth (www.breastfeedingindia.com)

Natural herbal medications, such as fenugreek (*Trigonella foenum-graecum*), milk thistle (*Silymarin*; *Silybum marianum*), *A. racemosus* (shatavari), alfalfa (*M. sativa*), blessed thistle (*O. benedictus*), goat's rue (*G. officinalis*), fennel (*F. vulgare*), and brewer's yeast, are often recommended to breastfeeding mothers to increase milk production. However, data on herbal products are limited and often based on anecdotal evidence. Fenugreek, milk thistle, and asparagus are frequently recommended and are the only herbal galactagogues that have some, although minimal, published clinical data on use in humans.^{6,10}

In this study, the researcher made an attempt to compare the effectiveness of nutritional interventions on breast feeding mothers with the selected biophysiological parameters of both mother and babies were evaluated on the day of birth and at 7th day of life.

With reference to the Organoleptic evaluation of the galactagogue mixes, scores (overall acceptability) were 17 out of 20 for fenugreek-galactagogue mix and maximum (20/20) for garlic-galactagogue mix, as they were satisfying all the criteria viz. Colour and Appearance, Texture, Flavour, Taste and Overall acceptability, though for the fenugreek-galactagogue mix, the flavour and Taste was scoring less.

With reference to the baseline prolactin levels between the experimental and control group revealed that there was no significant difference. However, after intervention with the galactagogue mixes for one week, the mothers prolactin levels were significantly high when compared with control group.

Several herbal galactagogues have been reported to be useful for nursing mothers. Mortel and Mehta (2013) in their systematic review of the efficacy of herbal galactagogues revealed that the outcomes included effect on serum prolactin and oxytocin levels, breast milk volume, infant weight, weight loss as a percent of birth weight, time to regain birth weight and the chemical composition of breast milk¹¹. In one of the studies, Turkylmaz (2011) has documented that commercially prepared herbal tea containing fenugreek increased milk production within 24-72 hrs after consumption¹², however, the herbal tea also included fennel, raspberry leaf and goats rue, substances that are traditionally used to augment lactation¹³. Hence, it was not possible to assess which herb or combination of herbs used in this study and that of Ushiroyama (2007) was responsible for the reported positive galactagogue activity¹⁴. Nevertheless, in our study only fenugreek was used as a galactagogue for an experimental group. Fenugreek seeds belong to pea family and

its effect on milk production has been reported anecdotally. Although there is contradictory evidence of its impact, yet it is attributed to stimulation of sweat production after inducing sweat gland, and since the breast is a modified sweat gland, thus the effect of fenugreek on milk production is possible¹⁴. Looking at the bioactive components in fenugreek, phytoestrogens and diosgenin (a steroid sapogenin) have been expected to exert estrogenic activity. In this lineage, Sreeja et al (2010) in their invitro assays found that fenugreek seeds contain estrogen like compounds, and they induce pS2 expression in MCF-7 cell lines. This pS2 is frequently used as a bio-marker for assessing the estrogenicity of a compound, and this steroidal sapogenin of fenugreek appear to account for the increase in milk flow observed from its use¹⁵. However, the exact mode of action is not yet clear. Hence, further studies are required to explore the exact mechanism of action at molecular level.

Human milk is complex mixture of substances that best meets the nutritional requirement of infants. It has been documented that when mother's milk added with garlic, the infants breast feed longer and suck more overall than they do when garlic is absent, at least under circumstances in which mothers have been consuming bland diet (without garlic) for several days. Desor et al (1997), claimed that infants may be attracted to or stimulated by garlic volatiles which in turn would increase the suckling process. This phenomenon implies prolonged suckling process would in turn induce production of more oxytocin and as a consequence more milk is that will be ejected. However these researchers point out that there is no evidence that will flavour of garlic in milk inherently be attractive to human infants in the same way that sweets and sugars do¹⁶. Similarly, Mennella et al (1993) found out in her research that the infants of mothers who have repeatedly consume garlic during the experimental periods breast feed for more periods of time when compared with the mothers who had ingested the placebo¹⁷. Similarly, when aroused with the volatile sulphur containing garlic like compounds, the mammalian infants would suck more¹⁸ and exhibit variety of other oral behaviour^{19, 20}.

Thus with the above documentation it is imperative that garlic consumption in nursing mothers would enhance the suckling ability of the infants and thereby sustaining long period of feeding. Nonetheless there is no clear evidence that garlic can increase the milk production. However, in our study comparatively garlic-galactagogue consumed nursing mothers showed a significant improvement in prolactin levels. Yet, as the population size for each group was less, a large population based interventional study would further clarify our hypothesis.

Conclusion

Consumption of maternal galactagogue mix supplementation seems to be useful for promoting and increasing the breast milk production in early postnatal days. However, use of any galactagogue should never replace lactation evaluation and counselling. Close follow-up of both mother and baby is essential even in instances of galactagogue support.

Hence, it is understood that all the three groups were having a significant difference in their final mean Prolactin levels. Indeed this result implies that such galactagogue supplementation could increase the production of Prolactin levels which in turn can therefore raise the breast milk production and thereby can also help to accomplish catch-up weight of the infants in the first week of the postnatal period.

Consumption of maternal galactagogue mix supplementation seems to be useful for promoting and increasing the breast milk production in early postnatal days.

However, use of any galactagogue should never replace lactation evaluation and counselling. Close follow-up of both mother and baby is essential even in instances of galactagogue support.

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