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EFFECT OF *HIPPOCRATEA OBTUSIFOLIA* EXTRACTS ON LACTATION INDUCEMENT

[1*]Datti, Y., [2]Idris, M.M., and [2]Mudi, S.Y.
Department of Chemistry, Northwest University, Kano.
[2]Department of Pure and Applied Chemistry, Bayero University, Kano.
*Correspondence author: whydee@yahoo.co.uk

ABSTRACT
The crude extract of the leaves and stalk of Hippocratea obtusifolia (commonly known as gwadayi in Hausa) was sequentially extracted with petroleum ether, chloroform, ethyl acetate and methanol. The extracts were subjected to bioactivity testing against lactation inducement in 24 female guinea pigs. The experimental animals with average weight of 200g were randomly grouped into six; the first five groups A, B, C, D and E were force-fed with feeds mixed with a measured portion of the extract, while the last group was force-fed with food mixed with distilled water only serving as control. The chloroform extract was found to be the only active extract, and this confirms the use of Hippocratea obtusifolia in lactation inducement.
Keywords: Lactation, Hippocratea obtusifolia, force-fed, galactagogue, endogastric tube

INTRODUCTION

One outstanding feature that distinguishes the class mammalia from the rest of the members of the kingdom animalia, is that their young ones are fed from the milk secreted by the mothers mammary glands (Nevelle and Peaker, 1981). The provision of a specialized maternal body fluid for neonatal nutrition allows birth to occur at an early stage of development and provide time of intense maternal infant interaction during early behavioral development. In addition, the nutritional reserves of the mothers can sustain the infant through a period of famine, as it contains all the nutrients required for the development of the body (Collier *et al*., 1977). These nutrients include carbohydrates, proteins, lipids and minerals (Hurley, 1998).

Lactogenesis is the term meaning the initiation of lactation. Lactation is the process of functional differentiation which mammary tissue undergoes when changing from a non-lactating to a lactating stage. This process is normally associated with the end of pregnancy and around the time of parturition (Nagasawa *et al*., 1969). Because lactogenesis is particularly dependent upon specific set of hormones (called lactogenic complex of hormones), mammary tissue from most state of non-lactating mammary gland can be made to undergo some degree of lactogenesis by administration of high amount of such hormones even in non-pregnant animals (Davis *et al*., 1983). The production of breast milk is controlled by interplay of various hormones, with prolactin being the predominant hormone involved. The maturation of breast tissue, resulting in milk production, is controlled by many other factors besides prolactin, including estrogen, progesterone, insulin, growth hormone, cortisol, thyroxine and human placental lactogen (Michael, 2008).

A galactagogue, or galactogogue is a substance that promotes lactation in humans and other animals (Gabay, 2002). It may be synthetic, plant-derived,or endogenous. Synthetic galactagogues such as domperidone and metoclopramide interact with the dopamine system in such a way to increase the production of prolactin (Forinash, *et al.,* 2012). There is some evidence to suggest that mothers who are unable to meet their infants' breastfeeding needs may benefit from galactogogues (McInnes and Chambers J 2008; Osadchy, *et al.,* 2012). Galactagogues may be considered when non-pharmacologic interventions are found to be insufficient (Amir *et al.,* (2011; Forinash, *et al.,* 2012). For example, domperidone may be an option for mothers of preterm babies who at over 14 days from delivery and after full lactation support still have difficulty expressing breast milk in sufficient quantity for their child's needs (Donovan and Buchanan 2012).

Throughout human history, there have been infants who have been nursed by surrogate mothers. Non-maternal lactation may have occurred as a results of maternal death or illness or because the birth mother gave over, or shared the care of her baby with another mother **(**Akers *et al*., 1981). Sometimes, the surrogate mother was already breastfeeding another baby and milk supply simply increased due to additional demand, to meet the growth needs for two or more babies. In the event that no already-nursing mother was available, reports from several continents describe effort by non-lactating woman to induce lactation by putting the baby to breast (Turner *et al*., 1956).

The inducement of lactation without the needs of a proceeding pregnancy could offer several practical and economical advantages (Bauman *et al*., 1977). Lactation has been induced in ruminants using steroids (progestrone and estrogen) as a short-term treatment followed by a wide variety of drugs aimed in increasing prolactin secretion. Those treatment protocols have resulted in lactation with milk production ranging between 25% and 82% of physiological post-portum lactation (Palmer *et al*., 2002).

Inducement of lactation has also been achieved both locally and traditionally, especially among the Hausa communities (Palmer *et al*., 2002). Factors necessitating the need to have the lactation induced include among others, the insufficiency of the milk from the birth mother, and the need to induce lactation on a birth mother, (who could not produce the milk). The method employed is mainly dependent on the necessitating factor: As for the insufficiency of the milk from the birth mother, the use of a local porridge called *kunun kanwa* (literally potash gruel) is employed. By immediately taking this porridge, hot and steaming, it is widely believed and accepted that the milk content will be increased to nearly maximum. Even though, there has not been any scientific proof to this, this tradition alongside the role played by the famous *wankan jego* (the hot water birth) in solving the problem of milk insufficiency, is widely accepted within the Hausa culture and tradition (Sofowora, 1984).

However, when it comes to the situation where there is no milk at all, in many cases use is made of some plants that are believed to be useful for that purpose. In India, lactation inducing herbal remedies are commonly used by nursing mothers to meet the nutritional requirement of their children (Nudrat *et al*., 2007). Notable plants used by the Warlis tribe in India to induce lactation in nursing mothers include *Carica papaya* Linn, *Asparagus racemosa* Wt, *Trigonella foenum graecum* Linn and *Madhuca indica* Gmellin (Nudrat *et al.,* 2007). Herbals and foods used as galactogogues have little or no scientific evidence of efficacy and the identity and purity of herbals are concerns because of inadequate testing requirements (Anderson, 2012; Mortel and Mehta 2013). The herbals most commonly cited as galactagogues are; shatavari (*Asparagus racemosus*) (Mortel and Mehta 2013), fenugreek (*Trigonella foenumgraecum*) (Mortel and Mehta 2013; Caroline, *et al.,* 2004), and torbangun (*Coleus amboinicus*), which has been used by the Bataknese people of Indonesia as a galactogogue for hundreds of years. (Mortel and Mehta 2013; Damanik, *et al.,* 2006). Others include, fennel (*Foeniculum vulgare*), milk thistle (*Silybum marianum*), chasteberry (*Vitex agnus castus*), and goat's rue (*Galega officinalis*)[1](Mortel and Mehta 2013).

Other notable plants believed to be useful for the purpose of inducement of lactation, are *Kigelia aethiopica*, known as *hantsar giwa* in Hausa, as well as *Hippocratea obtusofolia* or *gwadayi* in Hausa.

Hippocratea obtusofolia is a species of flowering plant in the family *hippocrateacea* which occurs in some part of West African countries like Nigeria, Niger, Senegal and Cameroon. *Hippocratea obtusifolia*, a shrub with green leaves and stalk growing up to 1m tall, is an evergreen where rainfall occurs throughout the year, but deciduous where there is long dry season, it produces sticky white fluid if detached (Huxley, 1992).

MATERIALS AND METHODS

Plant Material

The leaves and stalk of *Hippocratea obtusofolia* were collected at Dawaki, in Dawakin Tofa Local Government Area of Kano State, Nigeria, and were identified and authenticated at the Department of Biological Science, Bayero University, Kano.

Extraction of the Plant

Five hundred grams of the dried and ground form of the plant *Hippocratea obtusofolia* was put in 2.5L brown capacity brown bottle, and 1.5L of ethanol added. The set up was left for two weeks with constant shaking, after which the extract was filtered and then concentrated using Gallenkamp rotavapor (Vishnoi, 1979) to get the crude residue labeled HO. Part of HO was then extracted using aqueous methanol, and then extracted with petroleum ether using separating funnel. The petroleum ether extract was concentrated, dried, weighed and labeled HO1, while the aqueous methanol portion was again extracted with chloroform using separatory funnel. The chloroform extract was concentrated, dried, weighed and labeled as HO2, while the aqueous methanol portion was again extracted with ethyl acetate using a separating funnel. The ethyl acetate was concentrated, dried, weight and labeled as HO4. Each of the five extracts HO, HO1, HO2, HO3 and HO4 was tasted for lactation induction activity.

Phytochemical Screening

All the extracts from the plant were screened for the presence of alkaloids, tannins, flavanoids, saponins, sugar, glycosides, proteins and sterols.

Bioactivity Testing

All the four portions of the plant extracts HO, HO1, HO2, HO3 and HO4 were subjected to bio-activity testing. Twenty female guinea pigs of average weight of 200g were randomly divided into six groups of four each. The first five groups A, B, C, D and E were each force-fed with the portions HO, HO1, HO2, HO3 and HO4 of *Hippocratea obtusofolia* respectively for a period of two weeks. Each guinea pig received 2g/kg body weight of the extract dissolved in 2 mls of distilled water and force-feeding was carried out using a 2 ml syringe and endogastric tube. The last group F served as the control and was given distilled water free from the sample extract (Salim *et al*., 2005). After 2 weeks each group was quantitatively examined for the presence or any sign of lactation (Palmer *et al*., 2002). The weight of each guinea pig, with the weight of the force-fed extract (in brackets), is shown in Table 1.

Table1: Weight of guinea pig in g (wt. of fraction force-fed in mg)

	Group A (HO)	Group B (HO1)	Group C (HO2)	Group D (HO3)	Group E (HO4)	Group F (CTRL)
1	201(40.2)	198 (39.6)	190 (38.0)	197 (39.4)	203 (40.6)	195 (0.0)
2	200 (40.0)	201 (40.2)	204 (40.8)	206 (41.2)	202 (40.4)	205 (0.0)
3	196 (39.2)	197 (39.4)	204 (40.8)	203 (40.6)	199 (39.8)	201 (0.0
4	205 (41.0)	203 (40.6)	201 (40.2)	195 (39.0)	195 (39.0)	206 (0.0)

RESULTS

The masses of the dried extracts were obtained alongside their physical appearances, which are presented in Table 2. The phytochemical contents of the various fractions is shown in Table 3. After subjecting all the five extracts to bioactivity testing, the result showed appreciable activity in the chloroform extract HO2, while a mild activity was recorded by the crude. All other extracts and the control, showed no activity at all. The result obtained is tabulated in Table 4.

Table 2: Physical features of the five labeled portions:

S/N	EXTRACT	WEIGHT (g)	COLOUR
1	HO	24.20	Dark green
2	HO1	4.66	Green
3	HO2	3.81	Green
4	HO3	4.00	Green
5	HO4	5.28	Black

Table 3: Phytochemical constituents of the various fractions

SUBSTANCE	HO	HO1	HO2	HO3	HO4
Alkaloids	+	+	+		+
Tannins	+	+	-	+	+
Flavanoids	+	-	+		-
Saponins	+	-	-	+	+
Sugar	+	-	-	+	-
Glycosides	+	-	-	-	+
Proteins	-	-	-	-	-
Sterols	+	-	+		-

Key: + = present, - = absent

Table 4: Result of bioactivity test

Extract	HO	HO1	HO2	HO3	HO4	CONTROL
Activity	+	--	++	--	--	--

Key: + = the extract is active, -- = the extract is inactive.

DISCUSSION

It is not necessary to have been pregnant in order to breast feed. The special closeness fostered by breastfeeding can be profoundly comforting for both mother and child. Many women who have struggled with infertility problems, value the experience of breastfeeding even if the volume of milk they produce is small (Mittal, 2004). Breast milk forms the major source of nutrition for infants. Nursing mothers sometimes find it difficult to produce adequate milk and hence use various plants to enhance their lactation. A few plants that are used as galactogogue have been scientifically proven to be effective while some have no supporting data for their attributed property (Nudrat *et al.,* 2007).

The result obtained from subjecting the extracts of *Hippocratae obtusifolia* to bioactivity against testing (lactation inducement) shows that portion labeled HO2 (the chloroform extract) is the only active extracts, and this can be attributed to the presence of the active ingredient which might have contain the flavanoids of the plant, and according to Nielsen and Hansen (2004), flavonoids have pharmacological activities such as free radical scavenging, cancer protective, anti-inflammatory properties, lactation inducement, as well as antimicrobial and antioxidant activities. The remaining extracts (HO1, HO3 and HO4) were all inactive, and this might be attributed to absence of the active flavanoids in the three extracts. The control was also found to be inactive, and this is attributed to the fact that the control is free from any extract. Lactation was also found to have been induced by the crude extract HO, but this was achieved only to a lesser degree of activity, and this can be attributed to the presence of the lactation inducing agent in the crude extract HO, which is not as much as it is in the chloroform extract HO2. The analysis was conducted qualitatively by squeezing the mammary tract for the presence of milk (Palmer *et al.,* 2002).

The lactation inducement activity as achieved by the chloroform extract from the leaves and stalk of the plant *Hippocratea obtusofolia* is a confirmation of its usage as a traditional means of inducing lactation.

Consequently, this has met with the objectives of this research work in its effort to uncover at least a single plant that could be scientifically said to have a lactation inducing activity.

CONCLUSION

From the result obtained in this research, it can be concluded that only the extract HO2 was found to be effective in inducing lactation. The lactation inducement activity of the chloroform extract from the leaves of *Hippocratea obtusofolia* is an important discovery, as such, further work should be geared towards isolating and characterizing the active compound in HO2. This research work attempted to established the scientific base that reflects the claim for the local use of *Hippocratea obtusofolia.*

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