Effects of Oral Administration of a Decoction on Serum Levels of Leutinizing Hormone, Follicle Stimulating Hormone, Progesterone and Estradiol in Female Dutch-White Rabbits

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ABSTRACT
This study investigated the effects of oral administration of a decoction of twenty medicinal plants used by South-Western local healers with a view to establish the effects of various doses on serum levels of leutinizing hormone, follicle stimulating hormone, progesterone and estradiol using animal models. Three different concentrations of the extract were prepared according to the body weights of rabbits: 20 mg kg\(^{-1}\) mL\(^{-1}\) (low dose), 40 mg kg\(^{-1}\) mL\(^{-1}\) (medium dose) and 80 mg kg\(^{-1}\) mL\(^{-1}\) (high dose). Twenty female Dutch-white rabbits were divided into four groups of five rabbits each on the basis of uniform average weight. Groups 1, 2 and 3 were dispensed with low dose, medium dose and high dose of the extract respectively while group 4 was served none being the untreated control. After twenty one days of administration of extract, the animals were fasted overnight, anaesthetized with diethyl ether and blood sample collected for hormonal assay. The level of progesterone was reduced in low dose and medium dose groups but significantly increased in high dose group relative to control. The level of estradiol was significantly reduced in all the three groups relative to control. There was a decrease in luteinizing hormone level in medium dose and high dose groups but no effect on low dose group relative to control. Follicle stimulating hormone level was reduced in all the three groups relative to the control. The result showed the effect of the decoction to be dose-dependent and it has the potential to cause hormonal imbalance.

Key words: Decoction, progesterone, estradiol, dutch-white rabbits

INTRODUCTION
A medicinal plant is any plant which contains substances that can be used for therapeutic purposes in one or more of its organs. Information on the use of medicinal plants has been obtained from herbalists, herb sellers and indigenous people of Africa over many years (Sofowora, 2002). However, the herbal portions prescribed by local healers are usually not standardized, documented, specified and dosage are not precise. Although, the World Health Organization supports the appropriate use of herbal medicine and encourages the use of safe and effective remedies, it has also stated that most herbal medicines need to be studied scientifically (Ukueze and Abariku, 1998). However, scientific studies supporting the use of plants in traditional medicine remain very poor. Hence, this study investigated the safety of a decoction which is used in Western part of Nigeria in the treatment of various ailments. The active substances in medicinal plants are not evenly contained in all parts of a plant. Sometimes, they are contained mainly in flowers, leaves, roots, seeds, fruits or in the bark. One of the ways the active substances can be extracted for use is by decoction. The decoction process involves the simmering of the thicker and less permeable part
of the plant such as the roots, bark, fruit and seed, for easy extraction of their medicinal constituents. The plant material is cut into smaller pieces and placed in the simmering pot which is covered so, as to conserve the volatile components of the decoction. The joint use of multiple medicinal plants could be due to synergistic or additive effects of constituents (Igoli et al., 2005). This study investigated the effects of oral administration of a decoction of twenty medicinal plants used by South-Western local healers with a view to establish the effects of various doses on serum levels of leutinizing hormone, follicle stimulating hormone, progesterone and estradiol using animal models.

MATERIALS AND METHODS

Preparation of plant materials: The twenty plant materials chosen for this study include: Alstonia congensis, Tetrapleura tetraptera, Aristolochia repens, Citrullus colocynthis, Lonchocarpus cyanescens, Treculia africana, Anthocleista djalonensis, Uvaria afzelii, Plumbago zeylanica, Croton lobatus, Khaya ivorensis, Mondia whitei, Nauclea latifolia, Rauwolfia vomitoria, Securidaca longepedunculata, Urena lobata, Uvaria chamae, Olax subcorpioidea, Adenopus breviflorus and Petiveria alliacea.

The information about their local names, part(s) of plants used, therapeutic effects, methods of preparation and administration, as well as duration of treatment were obtained from South Western local healers. The plant materials were purchased from Oyingbo market in Lagos state. The plant materials were washed and boiled in 14 L of distilled water for 2 h. The decoction was allowed to cool, decanted and kept in air-tight container. Ten milliliter of the decoction was concentrated and the stock concentration of the extract was used to prepare different doses of the decoction in low dose (20 mg kg\(^{-1}\) mL\(^{-1}\)), medium dose (40 mg kg\(^{-1}\) mL\(^{-1}\)) and high dose (80 mg kg\(^{-1}\) mL\(^{-1}\)). To prepare the low dose, 2.6 g of the concentrated extract was dissolved in water and made up to 100 mL. The medium dose was prepared by dissolving 4.8 g of the concentrated extract in water and made up to 100 mL. The high dose was prepared by dissolving 11.2 g of the concentrated extract in water and made up to 100 mL.

Phytochemical screening: Phytochemical screenings for alkaloids, tannins, saponins, flavonoids, cardiac glycosides, phyllobatannin and anthraquinone were carried out on the concentrated extract, according to the methods of Trease and Evans (2002) and Wall et al. (1952).

Administration of extracts: Twenty mature female Dutch-white rabbits weighing 1.2-1.4 kg were divided into four groups of five rabbits each. Groups 1-3 received 1 mL of the appropriate dosage of the extract every morning before meal for 21 consecutive days while group 4 served as the control which received no dosage of the extract. Administration of extract was done orally by means of a catheter.

Collection of blood samples: After administration of extract, prior to sacrifice, the animals were fasted overnight, anaesthetized with diethyl ether and blood sample collected by cardiac puncture into plain bottle. Serum was obtained by allowing the blood in the plain bottle to clot at room temperature, after retraction of the clot, the sample was centrifuged at 1000 rpm for 10 min and the supernatant decanted.

Hormonal assay: The Radio immunoassay procedure described by Yalow and Berson (1960) was used to determine the LH, FSH, progesterone and estradiol levels.
Statistical analysis: The tests in this study were carried out in triplicate and subjected to statistical analysis. The student’s t-test was used in analyzing the data collected and p-values of <0.05 were considered statistically significant.

RESULTS AND DISCUSSION

Phytochemical screening of the plant extract revealed the presence of alkaloids, cardiac glycosides and anthraquinones while tannins, flavonoids, saponins and phylobatannins were absent (Table 1).

The effects of the aqueous extract of the decoction at low dose, medium dose and high dose on progesterone, estradiol, leutenizing hormone and follicle stimulating hormone levels in the female rabbits are shown in Fig. 1a-d, respectively. The level of progesterone was reduced in low dose and medium dose groups but significantly increased in high dose group relative to control (Fig. 1a). The level of estradiol was significantly reduced in all the three groups relative to control (Fig. 1b).

Table 1: Phytochemical screening of the decoction of twenty medicinal plants

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Inference</th>
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<tbody>
<tr>
<td>Alkaloids</td>
<td>+</td>
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<tr>
<td>Tannins</td>
<td>-</td>
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<tr>
<td>Flavonoids</td>
<td>-</td>
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<tr>
<td>Cardiac glycosides</td>
<td>+</td>
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<td>Anthraquinones</td>
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<td>Saponins</td>
<td>-</td>
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<tr>
<td>Phylobatannins</td>
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+: Presence of phytochemical and -: Absence of phytochemical.

Fig. 1(a-d): Effects of aqueous extract of a decoction on (a) Progesterone levels, (b) Estradiol levels, (c) Leutinizing hormone levels and (d) Dollicle stimulating hormone level.
There was a decline in LH concentration at medium and high doses while there was no change at low dose relative to control (Fig. 1c). The concentration of FSH declined at all doses of the decoction relative to control (Fig. 1d).

Hormonal imbalances may be attributed to some chemical agents contained in plant extracts. Phytochemical screening has revealed the presence of some bioactive and toxic agents of plant extracts that can affect the regulation of oestrus cycle, conception and reproduction (Benie et al., 2003). Alkaloids have been shown to reduce plasma concentrations of LH, FSH and estradiol (Bianco et al., 2006). In this study, the presence of alkaloids may be responsible for the alterations in the hormone levels. In females, ovulation of mature follicles in the ovary is induced by a large surge of LH secretion during the pre-ovulatory periods. The reduction in the LH levels at medium dose and high dose may be explained by an inhibitory effect of the extract on the release of LH which may trigger disruption of ovulation. This may result in impairment of oestrous cycle and consequently affect conception and normal reproduction in females (Al-Qarawi et al., 2000).

Progesterone is known to inhibit the secretion of LH and FSH (Crowley et al., 1985). Hence, the rise in progesterone level at high dose may be responsible for the significant fall in LH and FSH levels at high dose. Estradiol is directly responsible for the growth and development of reproductive organs. In synergy with FSH, estradiol stimulates granulosa cell proliferation during follicular development (Levin and Hammes, 2011). Follicle stimulating hormone is the central hormone of mammalian reproduction, essential for gonadal development and maturation at puberty as well as gamete production during the fertile phase of life (Simoni and Nieschlag, 1995). It stimulates the growth and maturation of ovarian follicles by acting directly on the receptors located on the granulosa cells. Administration of the extract at all doses was found to significantly decrease the serum FSH level and this might be responsible for the corresponding decrease in the serum estradiol level at all doses. This agrees with the fact that FSH is known to stimulate ovarian follicle cell proliferation and therefore, to stimulate estradiol synthesis. The reduction in the levels of FSH by the extract may hamper folliculogenesis and delay maturation of the follicle in the pre-ovulatory phase (Kumar et al., 1997). The extract may have exerted its effect on the anterior pituitary or the hypothalamus since FSH secretion is regulated by the gonadotropic releasing hormone secreted by the hypothalamus. The reduction in the levels of the hormone may have remarkable effect on conception in females. No previous scientific study has been carried out on this decoction. Thus, this study presents a preliminary report on the effect of this decoction used by South Western Nigeria, on hormone levels in Dutch-white rabbits.

CONCLUSION

The effect of the aqueous extract of the decoction was found to be dose-dependent and it has the potential to cause imbalance in hormone levels in female Dutch-white rabbits.

REFERENCES
