http://dx.doi.org/10.4314/jpb.v12i2.6
Vol. 12 no. 2, pp. 120-128 (September 2015)
http://ajol.info/index.php/jpb

Journal of
PHARMACY AND BIORESOURCES

In vitro evaluation of the effect of *Corchorus olitorius* (Tiliaceae) on isolated mouse uterus

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Received 2nd July 2015; Accepted 31st July 2015

Abstract
This study investigates the *in vitro* activity of the aqueous leaf extract of *Corchorus olitorius* (Tiliaceae) on the isolated mouse uterus. This plant was examined based on its traditional use in arresting miscarriage in Nigeria and also in order to document its potential usefulness in the therapy of uterine related pathologies. The aqueous leaf extract was tested on the isolated uterus of mice previously primed with diethylstilboestrol (0.2 mg/kg) 24 h prior to the experiment. The effect of the extract (0.003 – 3.0 mg/ml) and salbutamol (0.41 – 411.0 ng/ml) on spontaneous uterine contractility, in the presence and absence of oxytocin (0.004 IU/ml) and in the presence of high KCl (60 mM) were examined. The effect of the extract on $CaCl_2$-induced contraction in Ca^{2+} -free medium was additionally examined. All data obtained were analysed using one way ANOVA with post-hoc test for linear trend. It was observed that the extract significantly inhibited spontaneous and oxytocin-induced uterine contractions ($p < 0.05$) at lower concentrations while higher concentrations augmented contractions. Salbutamol, the reference drug, inhibited spontaneous and oxytocin-induced uterine contractions at all concentrations used. The extract had no significant effect on high KCl-induced contractions, however lower concentrations inhibited $CaCl_2$-induced uterine contractions (in Ca^{2+}-free medium) ($p < 0.05$) while higher concentrations appeared to augment contractions. The leaf extracts of *C. olitorius* inhibits uterine contractions of the isolated mouse uterus at lower concentrations while higher concentrations stimulates and augments uterine contractility.

Keywords: Oxytocin; Calcium; Salbutamol; In vitro uterine activity; *Corchorus olitorius*; Uterus

INTRODUCTION

Corchorus olitorius Linnaeus (Tiliaceae), also known as the jute plant (Zeghichi *et al*., 2003) is an annual herb plant whose leaves and roots are used as herbal medicine and consumed as a vegetable in cuisines of various cultures. The plant grows to an average height of about 2-4 m, possessing a few side branches. The leaves range between 5-15 cm in length and are alternate, simple, and lanceolate, with an acuminate tip and a finely serrated or lobed margin (Benor *et al*., 2012). The leaves are consumed in parts of Asia, the Middle East and Africa (Zeghichi *et al*., 2003). In Nigeria it is consumed especially by the 'Yorubas' to make a dish known as 'ewedu', and also consumed by the Northern part of Nigeria where it is called 'rama' (personal communication). It is used in the North of Nigeria to make a dish called 'taushe' and 'kwado' (personal communication).

Besides being used in cuisine, the plant is reported to have several medicinal

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ISSN 0189-8442

uses. The plant has also been reported to be rich in ascorbic acid, carotenoids, and α-tocopherol (Simopoulos *et al*., 1995; Zeghichi *et al*., 2003). The phenolic content of *Corchorus olitorius* is also reported to be higher than most vegetables and cereals (Velioglu *et al*., 1998). The plant has been reported to exhibit great antioxidant effects and to significantly suppress inflammatory responses (Yan *et al*., 2013). The seeds of *C. olitorius* have been used as a purgative and the leaves as demulcent and as a diuretic in parts of India (Chatterjee and Pakrashi, 1992). The seeds are also used in traditional medicine in India (Gupta *et al*., 2003b) and Nigeria (personal communication) for birth control. A study by Gupta and colleagues reported anti-steroidogenic effects of the plant in mature female mice (Gupta *et al*., 2003a). It has also been reported that the administration of the leaves of *C. olitorius* delayed the onset of sexual maturity and the appearance of the first oestrus (Gupta *et al*., 2003b). Studies have also reported a decrease in the weight of the ovary and the uterus as well as an elevation in cholesterol levels with the administration of *C. olitorius* (Gupta *et al*., 2003b). In Southern Nigeria, the tender leaves of the plant are collected and boiled and a glass of the liquid is consumed daily to prevent miscarriage (Akaneme, 2008), the leaves can also be washed in water at room temperature and consumed daily to prevent or arrest miscarriage (personal communication).

This study is therefore aimed at the pharmacological investigation of the direct effects of the leaf extract of *C. olitorius* on uterine contractility, in a bid to identify potential sources of drugs or prodrugs that will be useful in preventing pre-term birth.

EXPERIMENTAL

Preparation of plant material. Fresh leaves of *C. olitorius* were collected from 'Efihin' (a village market) in Benin City, Nigeria in the month of October, 2014. The plant was authenticated by Prof. M. Idu of the Department of Botany, University of Benin, Nigeria. The leaves were cleaned of debris and extraneous materials and then air-dried for about one week.

Extraction. The dried leaves were pulverized to powder using a milling machine (Brook Crompton Parkinson Motors, England). The powdered leaves were weighed and sufficient quantity of distilled water enough to cover 680 g of the powder was added. The plant material was then macerated in distilled water for 24 h with regular stirring. At the end of the maceration period, the set up was decanted and filtered. The resulting decoction was then concentrated using a water bath set at 80°C. The aqueous extract so obtained gave a yield of 37.9 g (0.06%) and was stored in a refrigerator at 4°C till needed.

Animals. Experiments were performed using female Swiss albino mice (20-25 g). The animals were obtained from Ambrose Ali College of Medicine, Ekpoma, Nigeria and maintained at the animal house Department of Pharmacology and Toxicology, University of Benin, Nigeria. They were kept to acclimatize for one month and fed with standard feed (Pellet from Oshomagbe Nigeria Limited, Adolor, Edo state, Nigeria) and water. The animals were exposed to natural lighting conditions. All experimental protocols were approved by the Institutional Animal Ethics Committee (IAEC) and complied with the Guide for the Care and Use of Laboratory Animals, Eighth Edition (Committee 2011). Only animals in pro-oestrus or estrus states were selected and used for the study.

Isolated tissue experiments. Diethylstilbesterol which was constituted in a 1:1 ethanol/ water solution was administered at a dose of 0.1 mg/kg (Crankshaw 2001) intraperitoneally as pre-treatment to the rats 24 h prior to the isolation of the uterus. This was necessary in order to bring all rats to a state of oestrus prior to the experiments, in order to institute a

state of hormonal homogeneity (Crankshaw, 2001; Bafor *et al*., 2013). On the day of the experiment, the rats were humanely sacrificed by cervical dislocation. The lower abdomen was dissected and the uterine horns were located and rapidly but carefully excised and transferred to a Petri dish containing aerated De-Jalon's solution according to prescribed methods (Bafor *et al*., 2010). The horns were separated, freed of connective tissue and adhering blood vessels and cut into segments of about 2 mm in length. The uterine segments were then threaded and mounted in a warmed (37°C) 10 ml organ bath containing continuously aerated physiological salt solution of the following composition in mM: 154.0 NaCl, 5.63 KCl, 0.648 $CaCl_2.2H_20$, 5.95 $NaHCO_3$ and 2.77 D-glucose. The mounted tissues were equilibrated under resting tension of 1.0 g for 30 min. The force and frequency of uterine contractions in the longitudinal muscle layers were measured using a 7003E-isometric force transducer (UgoBasile, Varise, Italy) connected to a 17400 data capsule digital recorder with an inbuilt bridge amplifier (UgoBasile, Varese, Italy). The channel recorder was previously set at a sensitivity of 1.00 chart speed of 5 mm/min, while the transducer was set to record a tension of 1.00 g so as to establish a relationship between the force applied to the transducer and the gauge deflection.

Studies on effect of extract on spontaneous uterine contractility. The direct effect of successive concentrations of the aqueous extract of *C. olitorious* on uterine smooth muscle contractility was investigated. Cumulative concentration-response relation-ships were obtained using final bath concentrations from 0.003– 3.0 mg/ml of the extract. A contact time of 10 min was allowed following each concentration of extract administered. At the end of the experiment, the tissue was washed and observed for another 10 min. Salbutamol a uterine relaxant was used as a standard uterine relaxant drug in this study at a concentration of 0.41 – 407.0 $\times 10^{-6}$ µg/ml.

Studies effect of extract on oxytocin-induced uterine contractility. The concentration-response relationships for the effect of the extract on oxytocin-induced uterine contractions were obtained. A submaximal concentration of oxytocin (0.004 IU/ml) was administered to the bath and left in contact for 3 min and then washed. Subsequently a single addition of the extract was added to the bath, left in contact for 2 min and without washout, the concentration of oxytocin was repeated and left for 3 min. After this period, the tissue was washed and allowed a rest time of 5 min before subsequent additions. This procedure was repeated for the different extract concentrations (0.003– 3.0 mg/ml).

Studies on effect of extract on high KCl-induced uterine contraction. In order to assess possibility of the interaction of the extract with calcium channels, KCl solution (60 mM) was administered to pre-contract the isolated uterine segments according to a modified method (Bafor and Okunrobo, 2010). Briefly, KCl was added, at 60 mM predetermined from preliminary experiments, to the bath and left in contact for 5 min and washed. This was then repeated and to the KCl-induced uterine contraction, the extract at 0.003 – 3.0 mg/ml were added.

Studies on effect of extract on uterine contractility in Ca^{2+}-free medium. In order to investigate possible calcium antagonist activity, the following protocols were performed as previously described (Gilani *et al*., 2005; Bafor and Okunrobo, 2010). The uterine tissue was equilibrated in normal De-Jalon's physiological salt solution (PSS) and after 30 min, PSS was replaced with Ca^{2+}-free solution containing 0.1 mM (Ethylene diaminetetraacetic acid (EDTA) and left to equilibrate for another 30 min. After 30 min, the PSS containing EDTA was again replaced

with a K^+-rich, Ca^{2+}-free PSS of the following composition (in mM): NaCl 154, KCl 50, $NaHCO_3$ 5.95, D-glucose 2.77, and EDTA 0.1. The new PSS was then left for a further equilibration time of 30 min after which calcium (6.8 mg/ml) was added in the absence and presence of extract (0.03 - 3.0 mg/ml).

Data analysis. Data were presented as mean ± standard error of the mean (S.E.M.). Concentration-response curves were obtained for each experimental model. Statistical analysis was carried out using computer statistical software GraphPad-Prism v.6 (GraphPad Software Inc, CA, USA). In datasets with sufficient data points, mean log concentration-response curves were analyzed by fitting data to a four-parameter logistic equation, using non-linear regression with GraphPad Prism 6.0 (GraphPad Software, San Diego, CA, USA) to determine the pEC_{50} values

Y = Bottom + (Top-Bottom)/(1+10^((LogEC50-X)*HillSlope)).

Where Y = response which starts at the Bottom and goes to the Top in sigmoid shape; X = logarithm of concentration and EC_{50} is the concentration that produces half the maximal responses.

Mean percentage responses were determined and compared where necessary. One-way analysis of variance (ANOVA) with post-hoc test for linear trend and Student's t-test were adopted for comparison. $p < 0.05$ was considered statistically significant in all cases.

RESULTS

Effect of extract and salbutamol on spontaneous uterine contraction. The aqueous extract of *C. olitorious* (ACO) was observed to concentration-dependently decrease the frequency of uterine contractions but increase the amplitude (Fig. 1 and 2 respectively). Salbutamol, the standard, was found to significantly inhibit both the amplitude and frequency of uterine contractions (Fig. 3).

Effect of Extract and Salbutamol on Oxytocin-induced Uterine Contraction. It was observed that at lower doses the extract inhibited the amplitude of OT-induced uterine contractions while higher concentrations appeared to augment the effect of oxytocin. (Fig. 4). Salbutamol on the other hand inhibited the effect of oxytocin at all concentrations (Fig. 5).

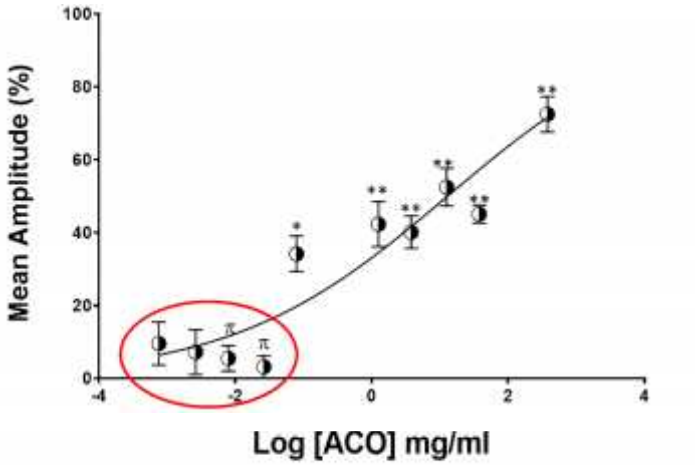

Figure 1: Concentration-response curves showing the effect of different concentrations of the aqueous leaf extract of *C. olitorius* (ACO) on the amplitude of uterine contractions.

The circle highlights concentrations at which inhibition occurred. Increasing cumulative concentrations of ACO was observed to increase the amplitude of uterine contractility. n= 4 animals. * $p < 0.05$; ** $p < 0.01$ of stimulatory concentrations compared to control; π $p < 0.05$ of inhibitory concentrations compared to control.

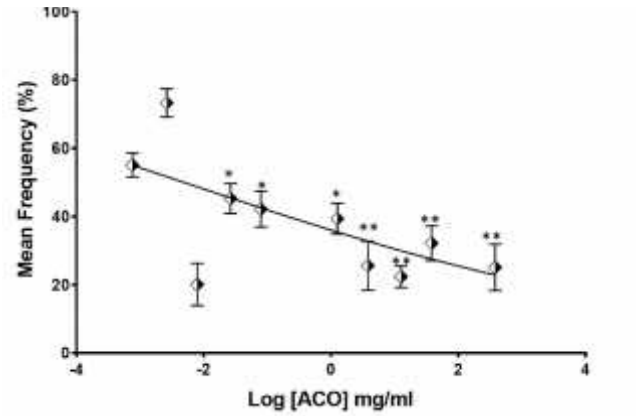

Figure 2: Concentration-response curves showing the effect of different concentrations of the aqueous leaf extract of *C. olitorius* (ACO) on the mean frequency of uterine contractions.
Increasing cumulative concentrations of ACO was observed to decrease the frequency of uterine contractility. n= 4 animals. * $p < 0.05$; ** $p < 0.01$ compared to control.

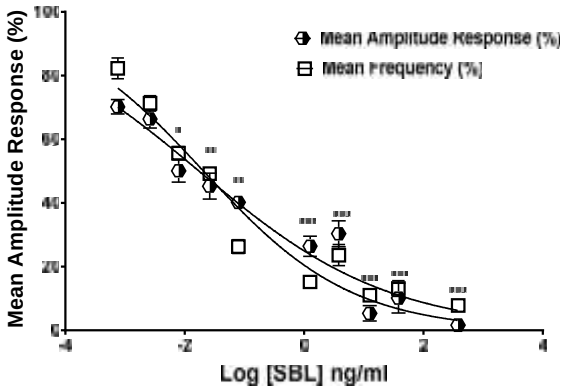

Figure 3: Concentration-response curves showing the effect of different concentrations of salbutamol (SBL) on the mean amplitude and frequency of uterine contractions.
Increasing cumulative concentrations of SBL was observed to decrease both the amplitude and frequency of uterine contractility. n= 4 animals. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$ compared to control.

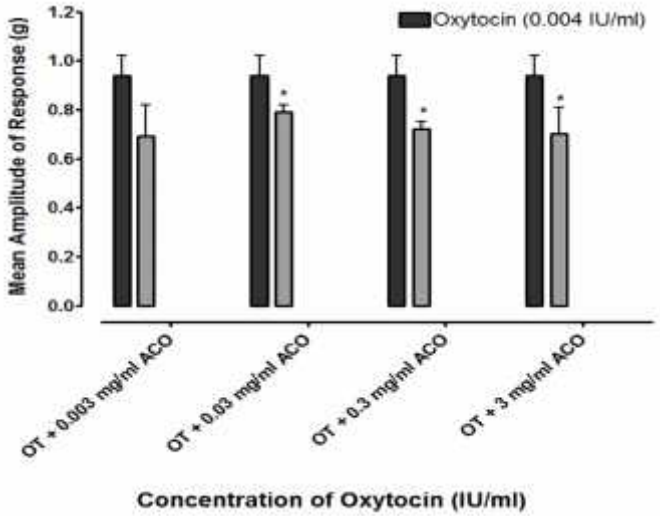

Figure 4 A bar graph showing the effect of the aqueous leaf extract of *C. olitorius* (ACO) on oxytocin (OT)-induced uterine contractions. Non-cumulative increases in ACO, concentration-dependently decreased the effect of OT (0.004 IU/ml) on uterine contractility. n = 4 animals. * $P < 0.05$. Black-coloured bars represent OT alone; Grey-coloured bars represent different concentrations of ACO.

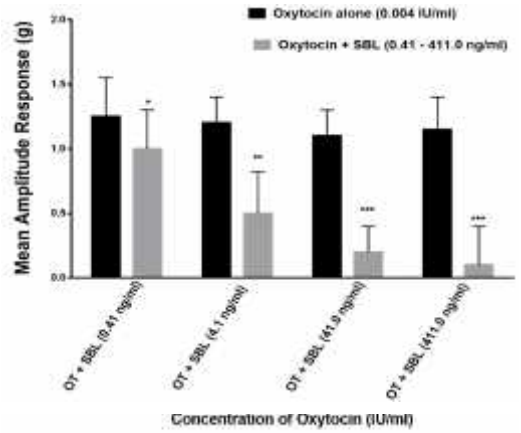

Figure 5: A bar graph showing the effect of salbutamol (SBL) on oxytocin (OT)-induced uterine contractions.
Non-cumulative increases in SBL, concentration-dependently decreased the effect of OT (0.004 IU/ml) on uterine contractility. n = 4 animals. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$ compared to control. Black-coloured bars represent OT alone; Grey-coloured bars represent different concentrations of SBL.

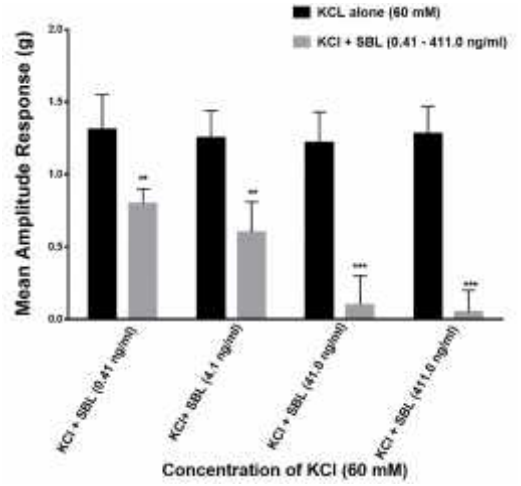

Figure 6: Concentration-response curve of showing the effect of SBL on high KCl (60 mM) – induced uterine contraction.
SBL concentration-dependently inhibited KCl-induced contractions. n = 4 animals; $^{**}p < 0.01$; $^{**}p < 0.001$.

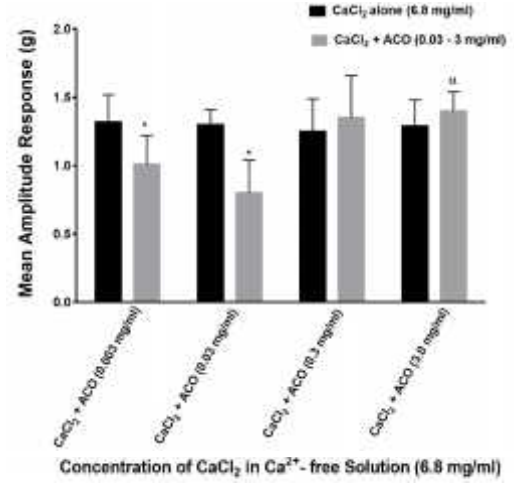

Figure 7: A bar graph of $CaCl_2$ alone in Ca^{2+}-free medium and in the presence of the aqueous leaf extract of *C. olitorius* (ACO) (0.03 – 3.0 mg/ml).
The extract inhibited the effect of $CaCl_2$ at lower concentrations but higher concentrations appeared to increase contractions. n = 4 animals. *$p <$ 0.05 of inhibitory effect compared to control; $^{\alpha}p < 0.05$ of stimulatory effect compared to control.

Effect on high KCl-induced Uterine Contractility. The extract at all concentrations used (0.03 – 3.0 mg/ml) had no significant effect on high KCl-induced contraction (data not shown) while salbutamol exhibited significant (P <0.05, P < 0.001) inhibition of high KCl-induced uterine contractility (Fig. 6).

Effect of Extract on uterine contractility in Ca^{2+}-free medium. At lower concentrations of the extract, $CaCl_2$-induced contractions were also inhibited by the extract in a concentration-dependent manner in Ca^{2+}-free medium (Fig. 7).

DISCUSSION

This study reports for the first time the direct effects of *C. olitorius* on uterine contractions. The study showed the plant to exhibit varying effects on uterine contractility. It was demonstrated in this study that the aqueous extracts of *C. olitorious* decreased the force of uterine contractility (amplitude) at lower concentrations while higher concentrations appeared to increase the amplitude. However it was observed to decrease the frequency of uterine contractions at all concentrations used. A similar effect was observed in the presence of the agonist oxytocin. On the other hand, the reference drug used in this study inhibited both spontaneous and oxytocin-induced uterine contractions at all concentrations used. That the plant exhibited dual effects suggests the presence of phytochemical constituents with differing activity on uterine contraction.

This intriguing ability of the extract to affect one parameter of contraction in a manner different from another parameter suggests that the extract is able to interact with multiple components and signaling pathways of uterine contractility. A probable reason why a drug can affect the frequency of uterine contractions independent of the amplitude might be due to a lack of effect or an opposing effect on the endogenous pacemaker cells, which was noted by Mackler and colleagues, (1999) to reside in uterine tissues (Mackler *et al*., 1999). The pacemaker cells in the uterus directly affect the gap junction assembly and will either increase or decrease cellular communication, and consequently uterine contractions. The presence of endogenous oscillator within the uterus regulates the amplitude of contractions (Mackler *et al.* 1999). This uterine oscillator differs from the neuroplexus pacemaker which coordinates smooth muscle activity in the gastrointestinal tract. Endogenous pacemaker cells exist in the uterine smooth muscle and are similar to the interstitial cells of Cajal (in the gastrointestinal and urethral smooth muscle cells) and they function similarly to the pacemaker cells in the cardiac muscle (Berridge, 2008). These pacemaker cells possess an oscillator present in the cytosol. This oscillator is able to generate repetitive Ca^{2+} transients that are able to activate the inward currents and cause them to spread through structures known as gap junctions and thus provides the signal that results in contraction (Berridge, 2008).

Though this study did not set out to exhaustively investigate the mechanisms by which *C. olitorious* affects uterine contractility certain hypotheses can be put forward. For instance the extract's ability to increase amplitude of spontaneous and agonist-induced uterine contraction suggests that the extracts may augment the opening of L-type voltage operated calcium-channels which allows entry of Ca^{2+} into the cell to trigger contraction (Berridge, 2008). However this was somewhat contradicted when it was observed that the extract had no significant effect on high KCl-induced contractions. In the presence of high KCl membrane depolarization is stimulated resulting in the opening of voltage-operated Ca^{2+} channels (VOCCs) thus promoting Ca^{2+} influx (Blaustein 1975). The Ca^{2+} current obtained in

such a manner is characterised by a slow onset of action and long duration and has been reported to be mediated by VOCCs (Blaustein, 1975). It may be therefore, that the opposing effects of the constituents may have interfered with the activity of the extract on high KCl-induced contraction. As such, fractionation and isolation of the active compounds may present a clearer picture. Possible interaction with prostaglandins is also suggested since prostaglandins are directly related with the regulation of spontaneous uterine contraction as well as that of oxytocin (Dublin *et al*., 1979). Since oxytocin is also known to cause the production of inositol-triphosphate and diacylglycerol on binding to its receptors (Schrey *et al.* 1988). It also therefore hypothesized that the extract may also interfere with the production of these second messengers related to oxytocin's effect. It was also observed in this study that the extract was found to inhibit $CaCl_2$-induced uterine contractions in Ca^{2+}- free medium. Calcium activated chloride channels have been reported to play a role in the excitability of the myometrium and also promote Ca^{2+} entry through VOCCs (Karaki *et al.* 1982, Carl *et al.* 1996, Wray *et al.* 2003, Jones *et al.* 2004, Hartzell *et al.* 2005)

Conclusion. Taken together, our results indicate that the aqueous leaf extract of *C. olitorius* inhibits spontaneous uterine activity but has dual effects of inhibition and stimulation in the presence of the agonist oxytocin which is dependent on the concentration. Interaction with intracellular calcium might play a role in the effect of the extract however this remains to be exhaustively verified.

Acknowledgements. The authors would like to thank Mrs. Osemelomen Omoruyi, Miss Isibhakhomhen V. Okosun and Mr. Philip A. Obarisiagbon for their technical assistance.

REFERENCES

Akaneme, Florence, I., 2008. Identification and preliminary phytochemical analysis of herbs that can arrest threatened miscarriage in Orba and Nsukka towns of Enugu State. Afr. J. Biotech. 7 (1), 006–011.

Bafor, E.E., Lim, C. V., Rowan, E.G., and Edrada-Ebel, R., 2013. The leaves of Ficus exasperata Vahl (Moraceae) generates uterine active chemical constituents. J. Ethnopharmacol. 145 (3), 803–812.

Bafor, E.E. and Okunrobo, L.O., 2010. In vitro myometrial inhibition by the partitioned aqueous fraction of Anthocleista djalonensis leaves. Can.J. Physiol Pharmacol. 88, 880–887.

Bafor, E.E., Omogbai, E.K.I., and Ozolua, R.I., 2010. In vitro determination of the uterine stimulatory effect of the aqueous leaf extract of Ficus exasperata. J. Ethnopharmacol. 127 (2), 502–507.

Benor, S., Demissew, S., Hammer, K., and Blattner, F.R., 2012. Genetic diversity and relationships in Corchorus olitorius (Malvaceae s.l.) inferred from molecular and morphological data. Gen. Res. Cr. Evol. 59, 1125–1146.

Berridge, M.J., 2008. Smooth muscle cell calcium activation mechanisms. J. Physiol. 586, 5047–5061.

Blaustein, M.P., 1975. Effects of potassium, veratridine, and scorpion venom on calcium accumulation and transmitter release by nerve terminals in vitro. J. Physiol. 247, 617–655.

Carl, A., Lee, H.K., and Sanders, K.M., 1996. Regulation of ion channels in smooth muscles by calcium. Am. J. Physiol. 271, C9–C34.

Chatterjee, A. and Pakrashi, C., 1992. *The Treatise of Indian Medicinal Plants*. New Delhi: Publication and Information Directorate, CSIR.

Committee, 2011. Guide for the Care and Use of Laboratory Animals: Eighth Edition. *In*: *Guide for the Care and Use of Laboratory Animals*. 118.

Crankshaw, D.J., 2001. Pharmacological techniques for the in vitro study of the uterus. J. Pharmacol. Exp. Toxicol. 45, 123–140.

Dublin, N.H., Ghodgaonkar, R.B., and King, T.M., 1979. Role of Prostaglandin Production in Spontaneous and Oxytocin-Induced Uterine Contractile Activity in in Vitro Pregnant Rat Uteri. Endocrinol. 105 (1), 47 – 51.

Gilani, A.H., Malik, H.M., Janbaz, K.H., Khan, A., and Saeed, S.A., 2005. Ethnopharmacological studies

on antispasmodic and antiplatelet activities of Ficus carica. J. Ethnopharmacol. 119 (1), 1–5.

Gupta, M., Mazumder, U.K., Pal, D.K., and Bhattacharya, S., 2003a. Onset of puberty and ovarian steroidogenesis following adminstration of methanolic extract of Cuscuta reflexa Roxb. stem and Corchorus olitorius Linn. seed in mice. J. Ethnopharmacol. 89, 55–59.

Gupta, M., Mazumder, U.K., Pal, D.K., and Bhattacharya, S., 2003b. Anti-steroidogenic activity of methanolic extract of Cuscuta reflexa roxb. stem and Corchorus olitorius Linn. seed in mouse ovary.Ind. J. Exp. Biol. 41 (6), 641–644.

Hartzell, C., Putzier, I., and Arreola, J., 2005. Calcium-activated chloride channels. Ann. Rev. Physiol. 67, 719–758.

Jones, K., Jones, K., Shmygol, a, Shmygol, a, Kupittayanant, S., Kupittayanant, S., Wray, S., and Wray, S., 2004. Electrophysiological characterization and functional importance of calcium-activated chloride channel in rat uterine myocytes. *Pflügers Archiv :* Eur. J. Physiol. 448, 36–43.

Karaki, H., Murakami, K., Nakagawa, H., Ozaki, H., and Urakawa, N., 1982. Effects of calmodulin antagonists on tension and cellular calcium content in depolarized vascular and intestinal smooth muscles. Br. J. Physiol. 77, 661–666.

Mackler, A.M., Ducsay, C.A., Veldhuis, J.D., and Yellon, S.M., 1999. Maturation of spontaneous and agonist-induced uterine contractions in the peripartum mouse uterus. Biol. Reprod. 61, 873–878.

Simopoulos, A.P., Norman, H.A., and Gillapsy, J.E., 1995. Purslane in human nutrition and its potential for world agriculture. Plts. Hum. Nutr. 77, 47–74.

Velioglu, Y.S., Mazza, G., Gao, L., and Oomah, B.D., 1998. Antioxidant Activity and Total Phenolics in Selected Fruits, Vegetables, and Grain Products. J. Agric. F. Chem. 46, 4113–4117.

Wray, S., Jones, K., Kupittayanant, S., Li, Y., Matthew, A., Monir-Bishty, E., Noble, K., Pierce, S.J., Quenby, S., and Shmygol, A. V, 2003. Calcium signaling and uterine contractility. J. Soc. Gynecol. Investig. 10, 252–264.

Yan, Y.Y., Wang, Y.W., Chen, S.L., Zhuang, S.R., and Wang, C.K., 2013. Anti-inflammatory effects of phenolic crude extracts from five fractions of Corchorus Olitorius L. Food Chem. 138, 1008–1014.

Zeghichi, S., Stamatina, K., and Artemis, P.S., 2003. Nutritional composition of molokhia (Corchorus olitorius) and stamnagathi (Cichorium spinosum). Plt. Human Health Nutrition Policy 91, 1–21.