

EVALUATION OF ANTIFERTILITY AND TERATOGENIC EFFECTS OF CRUDE EXTRACTS OF *Portulaca oleracea* IN MALE AND FEMALE ALBINO RATSOYEDEJI K.O.¹, BOLARINWA A.F.²,¹Department of Physiology, College of Medicine and Health Sciences, Afe Babalola University, Ado-Ekiti, Nigeria; ²Department of Physiology, College of Medicine, University of Ibadan, Ibadan, Nigeria

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ABSTRACT

Aqueous and methanolic extract *Portulaca oleracea* are designated as AEPO and MEPO respectively. The antifertility and teratogenic effects of AEPO and MEPO (75 mg/kg BW) were investigated in male and female albino rats.

Cohabitation of 50 days AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) treated male rats and untreated female rats for four weeks produced no positive mating, while the cohabitation of the control group male rat with untreated female rats led to positive mating.

Treatment of rats from day 1 to 5 of gestation (early pregnancy) with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused non-significant ($p > 0.05$) changes in the number of implantation sites relative to the control. Treatment of rats from day 6 to 15 of gestation (mid-pregnancy) with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused non-significant ($p > 0.05$) changes in fetal size relative to the control as well as absence of gross malformations and resorption sites in all the treated and control rats. Treatment of rats from day 16 to 20 of gestation (late pregnancy) with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused no significant ($p > 0.05$) changes in litter size and litter weights relative to their respective controls as well as absence of gross malformations and resorption sites in all the treated and control rats.

These findings probably indicate that the extracts (AEPO and MEPO) have antifertility effects in male albino rats but have no deleterious effects on the fertility of female albino rats.

Keywords: *Portulaca oleracea*, Implantation sites, Litter size, Resorption sites, Albino rats.

INTRODUCTION

Portulaca oleracea belongs to the family of *Portulacaceae*. It is commonly called Purslane in English language, 'Babbajibi' in Hausa language and 'Esan omode' or 'Papasani' in Yoruba language. It is a fleshy annual herb, much-branched and attaining 30 cm long (Burkill, 1997).

It is used medicinally in Ghana for heart-palpitations (Johnson, 1997). The plant is used as a diuretic in Nigeria (Ainslie, 1937). A tisane of the plant is drunk in Trinidad as a vermifuge (Wong, 1976).

At some areas near Benin City (Nigeria), the plant, along with other ingredients is taken as an aid to the development of the foetus (Vermmer, 1976).

It has been reported that aqueous and methanolic extracts of *Portulaca oleracea* have contractile effects on isolated intestinal smooth muscle in in-vitro preparations (Oyededeji *et al*, 2007).

It has also been reported that aqueous and methanolic extracts of *Portulaca oleracea* have some toxic and beneficial potentials on the blood chemistry of albino rats (Oyededeji and Bolarinwa, 2012).

The extracts of *Portulaca oleracea* have been reported to have protective effects on hypoxic nerve tissue (Wang *et al*, 2007), anti-inflammatory effects (Xiang *et al*, 2005) and wound-healing activity (Rashed *et al*, 2003). Parry *et al* (1987) also reported the skeletal muscle relaxant effect of the plant.

This study aims at investigating the antifertility activities of aqueous and methanolic extracts of *Portulaca oleracea* in male and female albino rats.

MATERIALS AND METHODS**Experimental Animals**

Adult male and female albino rats weighing between 160 g and 180 g bred in the Animal House of Physiology Department, LAUTECH, Ogbomoso were used. They were housed under standard laboratory conditions with a 12 hours daylight cycle and had free access to feed and water; they were acclimatized to laboratory conditions for

two weeks before the commencement of the experiments. All experiments were carried out in compliance with the

recommendations of Helsinki's declaration on guiding principles on care and use of animals.

Plant Material

Fresh specimens of *Portulaca oleracea* were collected from the Botanical Garden of the Forestry Research Institute of Nigeria, Jericho, Ibadan, and was authenticated in the above named institute where a voucher specimen (No FHI 108334) was deposited.

Preparation of the Extracts

Large quantity of the fresh specimens of *Portulaca oleracea* were washed free of soil and debris, and the roots were separated from the leaves and stems. The leaves and stems were air-dried for six weeks, and the dry specimens were pulverished using laboratory mortar and pestle, and then divided into two samples A and B.

Aqueous Extract of *Portulaca oleracea* (AEPO)

Weighted Portions (431.33g) of sample A were macerated and extracted with distilled water (1:2 wt/vol) for 72 hours at room temperature (26 – 28°C). The resulting solution was then filtered using a wire-gauze and a sieve with tiny pores. The distilled water was later evaporated using steam bath to give a percentage yield of 11.8% of the starting material. The dried material was reconstituted in distilled water to make up test solutions of known concentrations.

Methanolic Extract of *Portulaca oleracea* (MEPO)

Weighted portions (420.52g) of sample B were macerated and extracted with 70% methanol (1:2 wt/vol) for 72 hours at room temperature (26 – 28°C). The resulting solution was then filtered using a wire-gauze and a sieve with tiny pores. The 70% methanol was later evaporated using steam bath to give a percentage yield of 10.2% of the starting material. The dried material was reconstituted in distilled water to make up test solutions of known concentrations.

Ten gramme of AEPO and MEPO were dissolved in 100ml of distilled water to give a concentration of 0.1g/ml.

The dosages of AEPO and MEPO administered in these studies were in accordance with those reported by Miladi-Gorgi *et al.* (2004).

EXPERIMENTAL DESIGN

Fertility study in male rats (mating experiment)

Three male rats (160-180 g) were used. The rats were divided into three groups with each group consisting one male rat. The first two groups were orally given AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) for 50 days respectively, while the third group rat was orally administered with 0.5 mL of distilled water for the same number of days (control group). On day 51 of the experiment, three untreated female rats of proven fertility (160-180 g) were cohabitated with each of the male rats in the three groups; the cohabitation lasted for four weeks. Vaginal lavages were carried out on daily basis to observe the presence of spermatozoa which normally indicates positive mating.

Fertility and teratogenic studies in female albino rats

Vaginal lavages from adult female rats (160-180 g) were monitored on daily basis and only animals exhibiting three 4-5 day estrous cycles were used for the studies. Rats found in the proestrous phase of the cycle were caged with males of proven fertility overnight in the ratio of 2:1 and were examined the following morning for evidence of copulation. Vaginal lavages were carried out and rats with motile spermatozoa in their vaginal secretions were separated and that day was designated as day one of pregnancy. The pregnant rats were divided into different groups (with each group consisting of five pregnant rats) for the study of the effects of the extracts (AEPO and MEPO) on different phases of pregnancy.

Implantation or early pregnancy study (1st-5th Day)

Group I to III rats were used for the implantation study.

Group I rats received 75 mg/kg BW of AEPO from day 1 to 5 of gestation.

Group II rats received 75 mg/kg BW of MEPO from day 1 to 5 of gestation.

Group III rats received 0.5 mL of distilled water for the same number of days and served as the control group.

On the 6th day of gestation, the pregnant rats (groups I to III) were sacrificed by cervical dislocation to determine the number of implantation sites in both horns of the uterus using a dissecting microscope.

Mid-pregnancy or period of organogenesis study (6th -15th day)

Group IV to VI rats were used for the mid-pregnancy study.

Group IV rats received 75 mg/kg BW of AEPO from day 6 to 15 of gestation.

Group V rats received 75 mg/kg BW of MEPO from day 6 to 15 of gestation.

Group VI rats received 0.5 mL of distilled water for the same number of days and served as the control group.

On the 16th day of gestation, the pregnant rats (group IV to VI) were sacrificed by cervical dislocation, fetuses were removed from the rats by ventral laparotomy and examined. The number of fetuses and resorption sites were counted, the fetuses were also examined for gross malformations.

Late pregnancy study (16th -21st Day)

Group VII to XI rats were used for the late pregnancy study.

Group VII rats received 75 mg/kg BW of AEPO from day 16 to 20 of gestation.

Group VIII rats received 75 mg/kg BW of MEPO from day 16 to 20 of gestation.

Group IX rats received 0.5 mL of distilled water for the same number of days and served as the control group.

On the 21st day of gestation, the dams (pregnant rats) (groups VII to IX) were allowed to deliver their litters naturally. At birth, the number of pups (litters) were counted, weighed and examined for gross malformations; the number of resorption sites were also counted.

RESULTS

Effects of extracts (AEPO and MEPO) on fertility of male albino rats

Cohabitation of 50 days AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) treated male rats and untreated female rats for four weeks produced no positive mating (sterile mating or no pregnancy), while the cohabitation of the control group male rats with untreated female rats led to positive mating (pregnancy) with 6.24 ± 0.48 litter size and 5.26 ± 0.24 g litter weight.

Effects of extracts (AEPO and MEPO) on early pregnancy

The effects of AEPO and MEPO on the number of implantation sites during early pregnancy is shown in Figure 1.

Treatment of rats from day 1 to 5 of gestation with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused non-significant ($p > 0.05$) changes in the number of implantation sites when compared to the control.

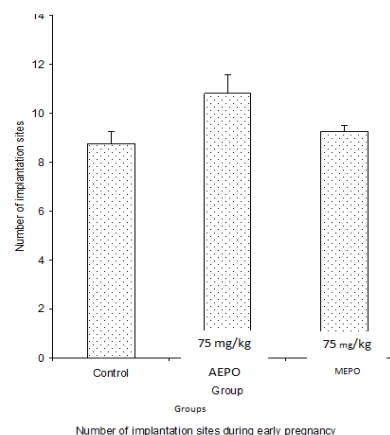
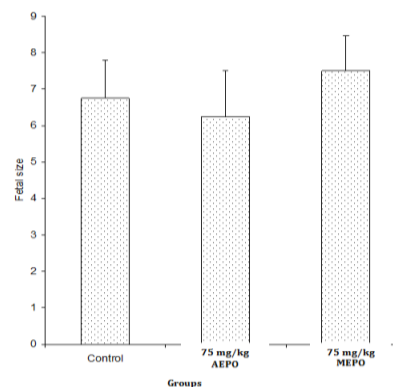


Figure 1: Effects of AEPO and MEPO on number of implantation sites during early pregnancy (1st - 5th day) (n=5)

Effects of extracts (AEPO and MEPO) on mid-pregnancy

The effects of AEPO and MEPO on fetal size during mid-pregnancy is shown in Figure 2.

Treatment of rats from day 6 to 15 of gestation with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused non-significant ($p > 0.05$) changes in fetal size relative to the control. Also, there were no resorption sites and no gross malformations (morphological anomalies) in all the treated and control rats.



Fetal Size during early Pregnancy

Figure 2: Effects of AEPO and MEPO on fetal size during mid-pregnancy (6th - 15th day, period of organogenesis) (n=5)

Effects of extracts (AEPO and MEPO) on late pregnancy

The effects of AEPO and MEPO on litter size and litter weights at birth are shown respectively in Figures 3 and 4.

Treatment of rats from day 16 to 20 of gestation with AEPO (75 mg/kg BW) and MEPO (75 mg/kg BW) caused no significant ($p > 0.05$) changes in litter size and litter weights relative to their respective controls. Also, there were no resorption sites and no gross malformations (morphological anomalies) in all the treated and control rats.

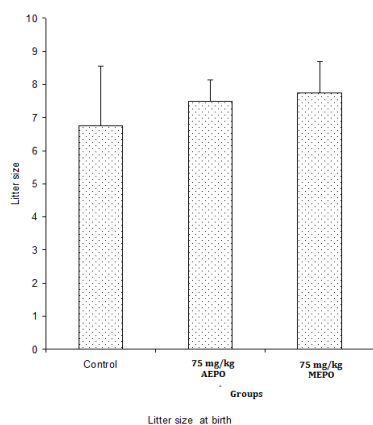


Figure 3: Effects of AEPO and MEPO on litter size at birth (21st day) (n=5)

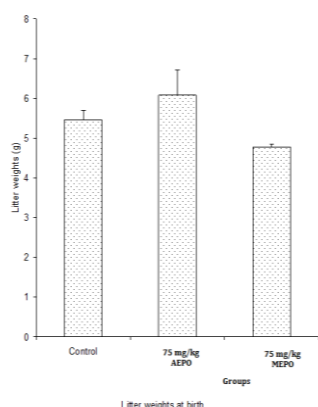


Figure 4: Effects of AEPO and MEPO on litter weights at birth (21st day) (n=5)

DISCUSSION

Cohabitation of the crude extracts (AEPO and MEPO) treated male rats and untreated female rats for four weeks produced no positive (sterile mating). Similar results were reported in rats treated with *Barleria prionitis* extracts (Gupta et al., 2000) and *Carica papaya* extract (Lohiya et al., 1994). This observation could be due to reductions in sperm motility, sperm counts, sperm viability and increase in the percentage of abnormal sperm cells induced by the crude extracts as reported by Oyedeji and Bolarinwa (2013).

Treatment of pregnant rats with the crude extracts (AEPO and MEPO) during early pregnancy caused non-significant changes in the number of implantation sites relative to the control. Contrary result was reported by Vasudeva and Sharma (2007) in *Hibiscus rosa-sinensis* extract treated rats. This could indicate that the extracts did not cause the disturbance of endocrine-endometrial synchrony which is dependent on estrogen and progesterone balance, since it has been reported that for implantation to take, exact equilibrium of

estrogen and progesterone is essential and any disturbance in the level of these hormones may cause infertility (Psychoyos, 1966).

Treatment of pregnant rats with the crude extracts (AEPO and MEPO) during mid-pregnancy caused no significant changes in fetal size relative to the control; also there were no resorption sites and no gross malformations (morphological anomalies) of fetuses in all the treated and control rats which probably indicates that the extracts have no teratogenic and deleterious effects on the fertility of female albino rats at this phase of pregnancy.

During late pregnancy, the crude extracts (AEPO and MEPO) treated pregnant rats were delivered normally with no evidence of prematurity or abortion or death, suggesting that the crude extracts were not abortifacients or having prostaglandin-like activities on the uterus. Also, treatment of pregnant rats with the crude extracts during late pregnancy caused no significant changes in litter size and litter weights relative to the control, as well as absence of gross malformations which probably indicates that the crude extracts have no teratogenic and deleterious effects on the fertility of female albino rats at this phase of pregnancy.

In conclusion, this study has shown that the crude extracts of *Portulaca oleracea* have antifertility effects in male albino rats. However, these extracts have no teratogenic and deleterious effects on the fertility of female albino rats, this could be the reason why the plant (*Portulaca oleracea*) along with other ingredients is taken as an aid to the development of fetus in some areas in Edo State, Nigeria (Vermeer, 1976) and to prevent miscarriage in Tanganyika, Tanzania (Bally, 1937).

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