Esxon Publishers

International Journal of Applied Research and Technology
ISSN 2277-0585

Publication details, including instructions for authors and subscription information:
http://www.esxpublishers.com

Effects of crude oil contaminated forage on the oestrous cycle of female rabbits

Nodu, M. B. and Ohimain, E. I.

Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria

Available online: April 30, 2014

To cite this article:

Nodu, M. B. and Ohimain, E. I. (2014). Effects of crude oil contaminated forage on the oestrous cycle of female rabbits. *International Journal of Applied Research and Technology.* 3(4): 111 – 117.

International Journal of Applied Research and Technology
Vol. 3, No. 4, April 2014. 111 – 117.

Esxon Publishers

Effects of crude oil contaminated forage on the oestrous cycle of female rabbits

Nodu, M. B. and Ohimain, E. I.

Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria

(Received: 21 March 2014 / Accepted: 03 April 2014 / Published: 30 April 2014)

Abstract

Oil spill incidence is increasing in Nigeria due to the activities of oil producing companies and ethnic militias. This study was designed to assess the impacts of oil spill on the oestrous cycle of female rabbits. Forty semi-adult female chinchilla rabbits aged 16 – 17 weeks, weighing 1.30 - 1.42kg were used for this study. The rabbits were fed with graded oil contaminated forage; O% 0.05%, 0.1%, 0.15% and 0.20% for treatments A (control), B, C, D and E respectively in a completely Randomized Design (CRD) for 24 weeks. Detection of oestrous was based on the observation of some vital oestrous signs of rabbits. Results showed that frequency of oestrous decreased, duration of oestrous decreased, length of oestrous cycle increase and heat intensity decreased as the concentration of crude oil contamination increased. The study showed that crude oil could be detrimental to the reproduction of rabbit both in the wild and in domestication. Highlights from this study could be useful for conservation of species, breeding, ecological food chain and pollution control.

Keywords: Oil Spills, Oestrous Cycle, Rabbits

For corresponding author:
E-mail: *info@esxpublishers.com*
Subject: *0314-0226*

Introduction

Nigeria is a major crude oil producing nation, producing 2.0 – 2.7 million barrels of oil daily (Ohimain, 2012; 2013). Nigeria is ranked as the 12th largest producer and 7th largest exporter of crude oil in the world (Ohimain, 2013). Despite the impressive crude oil production records, the country depends on the importation of refined petroleum products including gasoline, diesel and kerosene to meet her domestic demand (Ohimain, 2010; 2011, 2012). The country now spends over ₦ 4 trillion ($ 1 = ₦ 160) annually for the importation of refined petroleum products for domestic consumption. Oil spills occur at different stages during the entire value chain such as exploration, drilling, production, transportation, refining and distribution of refined products. In the past, oil spill is mostly caused by corrosion of pipelines and equipment failures. But now oil spill is mainly caused by sabotage, illegal bunkering and artisanal refining of petroleum (Ogbeibu and Iyobosa, 2013; Ogodo and Esemuede, 2013). Statistics hare shown that oil spill is increasing in the country owing to the activities of ethnic militias. Both the volume and number of oil spills are increasing. NNPC (2011) reported pipeline oil spil incidence in Port-Harcourt, Warri, Mosimi, Kaduna and Gombe terminals. In 2011, a total of 2,787 line breaks incidence occurred with 2,768 from vandalism and 19 by rupture leading to loss of 157.81 trillion MT of product. The Department of Petroleum Resources (DPR) recorded 4647 incidents oil spill incidence between 1976 and 1996 releasing 2,369,470 barrels of oil into the environment (Kadafa, 2012; Nwilo and Badejo, 2005) and 2,097 oil spill incidents between 1997 and 2011.

Nigeria oil resources is mostly located in the southern part of the country particularly in the Niger Delta region. The Niger Delta is a fragile ecosystem comprising of 70,000 sq km of wetlands consisting of mangroves, freshwater, tidal flats, beach ridges and riparian forests. Oil spills have variously occurred on land, swamps and offshore locations both in the continental shelf and deep offshores. When spills occurs, the impacts is quite devastating. UNEP (2011) reported that oil spills have impacted Ogoni land affecting soil and vegetation including mangroves, surface and ground water. The activities of oil producing companies and ethnic militias involved in artisanal refining was the major cause of oil spill in Ogoni.

The impacts of oil spills on land particularly soil and vegetation have been widely reported in literature (Ogodo and Esemuede, 2013). Also, widely reported is the impacts on fisheries and marine mammals particularly sea turtles, whales, dolphins, seals and sea otters (Ogbeibu and Iyobosa, 2013). But literatures on the impacts of oil spill on land mammals such as rabbits are few (Berepubo et al., 1994; Johnson, 1994). Mammals particularly herbivores and omnivores such as rabbits feed on vegetation and other food substances. The impacts of crude oil contaminated forage on mammals are scarce. Literature on the effects of oil spill on the oestrous cycle of rabbit is also scarce. Hence, this study is aimed at assessing the impacts of contaminated forage on the oestrous cycle of female rabbits. Such information could be useful for conservation of species, breeding, ecological food chain and pollution control.

Materials and Methods

Forty semi-adult female chinchilla rabbits aged 16 – 17 weeks were used for this study. The rabbits initially weighed between 1.30 - 1.42kg. The rabbits were procured from Rivers State Agricultural Development Programme (ADP) farm (livestock unit), Rumuodomaya in ObioAkpor Local government area. The pre-experimental period lasted two weeks during this period all the animals used in this study were subjected to the same management conditions including housing, feeding and medications (antibiotics, coccidiostat and vitamins). During the pre-conditioning period, all the rabbits were properly housed in hutches. These single tier hutches had three compartments per hutch. The rabbits were kept two per compartment. These single tier conventional hutches were made of wire mesh and wooden frame. The compartments were large enough to accommodate about four rabbits each. All the hutches were housed in a single room (shed) under the same environmental condition. This made it convenient for the experiment. All the hutches and animals were properly identified on individual basis using identification tags.

All the rabbits during the two weeks of pre-conditioning were given the same feed and water. Feeders and drinkers in the hutches which were made of heavy clay to prevent over turning and spillage of feed and water by the rabbits. The animals were fed *ad libitum* with feed made of green forage prepared from mixture of commonly available grasses and legumes. The forage was supplemented with concentrate feed to boost their nutrition and enhance their sustenance (Aduku and Olukosi, 1990; Fielding, 1991). The green forage fed to these rabbits composed of *Panicum maximum* (Guinea grass), *Brachiara mutica* (Para grass), *Centrocemapubescens* (Centro), *Puereria phaseolides* (Pecero), *Calapogonium muconoides* (Calapo) and *Aspiria africana* (Bush marigold). The concentrate feed supplement was the grower's mash as recommended by Ibeawuchi and Fajuyitan (1986). Clean and cool water was provided *ad libitum* on daily basis. The feeding and drinking troughs were cleaned and washed on daily basis and also disinfected as often as necessary.

During pre-conditioning, all the animals were administered prophylactic coccidiostat (25% embazin) as well as broad spectrum antibiotics like Terramycin soluble power. Some vitamin drugs like biovite were also administered to the rabbits. These drugs are to protect the animals from some common rabbit diseases that would have attacked the animals and interfered with the study during the experimental period. The crude oil used for the study was Bonny light, which was obtained from Nigeria Agip Oil Company (NAOC) at Obrikom flow station in Ogba-Egbema-Ndoni local government area of Rivers state. Prior to actual commencement of the study and contamination of the forages, the crude oil was exposed for 24hours in shallow pans to allow evaporation of the light fraction in order to ensure a stable product (White, 1975). This was to stimulate naturally occurring conditions following oil spillages in the oil bearing communities. The completely Randomized Design (CRD) experimental method was used for this study. The use of the CRD experimental design was based on the fact that the subjects (animals) in this study were homogenous. The forty female rabbits used for this study were randomly divided into five groups (Table 1) of eight animals per group representing one control group and four treatment groups (A, B, C, D and E). Group A served as the control group. The forty rabbits were therefore randomly allocated to the

five dietary groups including the control (A). Each treatment group comprised of four replicates with two animals per replicate giving a total of eight animals per treatment group. The duration of the experiment was 24 weeks.

The use of these levels of crude oil contamination in this study was informed by the results of a study reported by Berepubo, *et al.*, (1994) providing these dose of crude oil not to be too lethal compared to the pilot study of Johnson (1994) that reported a high mortality of 25 out of 32 rabbits (78.13%) used in the experiment was recorded within one week of administration of the crude oil at the different level of 10g (1%), 20g (2%), 30g (3%) contamination. It therefore suggest that the above levels of crude oil contamination/ingestion by animals are highly lethal. Hence, lower concentration were used in this study (Table 1). On commencement of the experiment, all the animals from the various treatment groups were then fed forage contaminated with the graded levels of crude petroleum. The contamination of the forage was done by incorporation of the measured amounts of crude oil in the forage by simple mixing and homogenization for at least two hours to ensure imbibition. Rabbits in treatment, A which serve as the control group, were not however fed the crude oil contaminated forage. They were fed crude oil free forage. In addition to forage, all the rabbits in the various group (A – E) were fed of commercial growers mash (Ibeawuchi and Fajuyitan, 1986). The supplementation of concentrate feed was to provide basic required nutrients in order to avoid malnutrition and its attendant effects. The concentrate was not however given to the rabbits until a substantial proportion of the test feed have been eaten by rabbits in the treatment groups (B – E). Detection of oestrous was based on the observation of some vital oestrous signs of rabbits (Berepubo, *et al.*, 1993). Such signs include aggressive restlessness (which include mounting and riding each other), increased vascularization of the vulva (moist, purplish, slightly swollen vulva), and exposition of the hind quarters. Other signs, which are often considered secondary signs are rubbing of the chin on feeding and drinking troughs and on the hutch (wire mesh), stretching of ears and frequent micturition (Hatfez, 1960; Okorie, 1983; Fielding, 1991). In this study, the oestrous parameters considered include frequency of oestrous, duration of oestrous, length of oestrous cycles and heat intensity.

Data on the intensity of oestrous in the rabbits was based on the detection of oestrous as exhibited by the rabbits, which was categorized as "intense heat", "less intense heat", "mild heat: and "silent heat". These are based on the manifestation of heat in rabbits (Hafez, 1960). The manifestation of the heat signs as categorized above attracted arbitrary scores of 5, 3, 1 and 0 respectively. This implies that where the animal manifested a combination of the first three aforementioned vital signs and any other secondary signs stated above, "intense heat" was recorded and score 5 points. Manifestation of only two of the vital signs with/without other sign was regarded as "less intense heat" and was scored 3 points. While manifestation of only one vita sign was interpreted mild heat and scored 1 point. The manifestation of only one or none of the secondary signs by the animal was interpreted as "silent heat" and zero was scored against such animal. These scores were later computed on treatment basis as intensity of heat for the animals/treatment group.

Frequency of oestrous was determined by keen observation of the aforementioned visual signs of oestrous in the rabbits. The number of times (n) these signs are observed in a rabbit within a given period of time was determined and noted as frequency of oestrous. In the same vein, the length of time (in hours or minutes) these observed heat signs lasted in the rabbit on heat per heat exhibition was determined and recorded as heat or duration of oestrous. Data for this parameter was generated by noting the times difference (in days) between one oestrous cycle in a particular rabbit and the onset of the next heat or oestrous period in the same rabbit. This was also computed on treatment group basis to serve for length of oestrous cycles in the rabbits per treatment. Data obtained from this study were subjected to analysis of variance (ANOVA) according to the procedures of Steel and Torrie (1981), where significant differences existed, the treatment means were compared using Duncan Multiple Range Test (Duncan, 1955). SSPS version (SPSS Inc. Chicago) was used for statistical analysis.

Results and Discussion

Oestrous signs are less obvious in rabbits but they exhibit certain behavioral characteristics during their oestrous period. The oestrous behaviors (frequency of oestrous, length of oestrous, duration of oestrous and the intensity of heat) of the experimental animals are presented in Table 2. The results indicated highly significant differences ($P<0.001$) in oestrous behavior among the experimental animals. The results obtained on oestrous behavior of the does used in this study revealed a highly significant difference ($P<0.01$) between the control (A) and the dose placed on crude oil contaminated diets (B – E). Results for the frequency of oestrous as exhibited by the does during the experimental period of the study (Table 1), indicate that the treatment was a source of variation ($P<0.001$) of the frequency of oestrous among the animals. The highest frequency of oestrous of 4.63 ±0.18 was observed in treatment A (control), while the treatment E had the least mean score of 1.50±0.29. The other groups had means scores of 3.50±0.22, 2.60±0.40 and 1.80 ± 0.20 for treatment B, C and D respectively. Frequency of oestrous results indicated higher frequency of oestrous among the control as against less frequent cycles among the other treatment groups B – E. The results revealed that animals in treatment B group have high frequency, but was still significantly less ($P<0.05$) when compared to the control group. The data showed that the frequency of oestrous decline as crude oil contamination of the forage increased. It is plausible to suspect that the treatment factor (crude oil) suppressed oestrous cycle in the rabbits, reducing oestrous frequency among them, whereas the control group had normal oestrous cycle with the cycles occurring as frequent as normal within the experimental period.

Frequency of oestrous is of practical importance as it will guide and enhance the farmers breeding programme. Therefore, a higher frequency of oestrous is desired, as this would enhance chances of breeding in case the previous oestrous cycle was missed. This would mean a greater number of rabbits could be produced within a planned breeding period. Of significance in this study is the finding that crude oil suppresses and depresses oestrous behavior in animals especially rabbits resulting in reproductive deficiency which spells doom for the livestock industry particularly animals in the Niger Delta region where oil spillage occur frequently. The finding of this study agrees with report by previous researchers who reported that oil (both crude and refined) and other heavy metals are toxic stressors to the system including the reproductive system

of both man and animals (Anderson et al., 1993; Plyaschenko and Sidorov, 1987; Ekweozor and Snowden, 1987). It is to this effect that Mattison (1983) noted that occupational exposure to lead compounds decreased fertility of male workers and increased the rate of spontaneous abortions in their wives and that blood lead concentration ≥53mg lead/100ml blood were associated with reduced fertility. It suffice to state here that the hydrocarbons present in the crude oil ingested into the blood stream of the tested rabbits played an antagonistic role in the efficient functioning of the reproductive cycle of the rabbits.

A similar gradual significant decrease ($P<0.05$) was observed for heat intensity with increasing dosage of contaminant among the experimental animals (Table 1). Among the rabbits fed the crude oil contaminated forage, treatment B had the highest score of 2.20±0.21 while treatment E recorded the least intensity, exhibiting silent heat with a mean score of 0.50±0.00. Scores for the other treatments were intermediate between the two groups. However, the control animals exhibited the highest heat intensity score of 4.04±0.14 among all the groups and is categorized "intense heat". The treatment group C and D exhibited silent heat a mean scores of 1.63±0.19 and 1.60±0.24 respectively. The importance of heat signs manifestations in farm animals can never be over emphasized. This is because it would ensure a more and accurate recognition of oestrous in the animals as well as guiding the farmer on accurate time of breeding to ensure conception and efficient reproduction. Although oestrous signs are less obvious in rabbits (Paufler, 1985; Fielding, 1991; Kamwanja and Hauser, 1983), some heat manifestations have been identified which Hafez (1960) categorized into three signs as; increased vascularization and smelling of the vulva; exposition of the rear quarters and arching of the back and frequent micturition. Other supportive signs also identified include stretching of the ears, rubbing of the chin on feed trough or waterer, and aggressive restlessness. Exhibition of one or two of the supportive signs implies silent heats, which were the signs, exhibited by those placed on treatments B to E. Treatment group B (0.50% contamination) showed better oestrous signs compared to other treatment groups (C, D and E). Though, these signs were not pronounced enough and were therefore considered as exhibiting silent heat. It is plausible to assert that the depression in exhibition of oestrous signs in these experimental rabbits is due to the suppressive effects of the ingestion crude oil on the reproductive system. Buttressing this assertion is the fact that rabbits in the control group displayed all the oestrous signs as categorized by Hafez (1960); otherwise referred to as "intense heat".

In recognition of the importance of oestrous in the reproductive efficiency of farm animals, Zemjanis (1980) had reported that failure of oestrous is considered one of the most common causes of infertility. This condition seems however relatively rare in rabbits as they are induced ovulators with less visible or obvious oestrous signs (Paufler, 1985; Fielding, 1991; Berepubo, *et al.,* 1993). Mean scores for the duration of oestrous exhibited by the rabbit (Table 2), revealed that animals in the control group (A) had the highest oestrous duration of 6.71±0.11 hours. The shortest oestrous duration was observed in animals in treatment E with 5.25±0.14 hours. However, animals in treatment B were not significantly different from those of the control group in the duration of oestrous. The oestrous duration among animals in the other treatment groups are 6.65±0.20 hours for treatment B; 6.40±0.19 hours for C and 6.00±0.22 hours for treatment D. Observations on duration of oestrous revealed a reduction in time between the treatment groups and the control group. The duration of oestrous in treatments C, D and E were significantly different ($P<0.05$) from those of groups A and B. short oestrous is not a favorable condition in rabbit production as well as in other farm animals species. The implication is that a farmer may miss the oestrous period in an animal with short duration of estrous which will consequently affect breeding (mating programme). This means that the farmer has to wait until when the animal returns to heat again and this may eventually delay breeding and subsequently alter the breeding programme of a livestock farm. The longer oestrous duration observed in the control animals is in consonance with the investigation of Berepubo, *et al.,* (1993) and Okere (1997) in which does were subjected to some treatments with the control does exhibiting longer oestrous duration than does in the different treatment groups. Oestrous cycle has been presumed to be 5 – 7 days (Paufler, 1985; Berepubo, *et al.,* 1993). Observations from the present study indicate that the oestrous cycles for the does fall within the average of 6.59±0.12 and 7.38±0.31 days. This observation agrees with the presumed 5 – 7 days cycle reported previously by other researchers (Paufler, 1985; Berepubo, *et al.,* 1993).

The mean length of oestrous cycle observed during the experimental period (Table 2) ranged from 6.59±0.12 to 7.38±0.31 days. The group A (control group) showed the shortest cycle of 6.59±0.12 days and the longest oestrous cycle of 7.38±0.31 days in treatment E (0.20% crude oil contamination). The other treatment groups showed oestrous lengths of 7.35±0.11, 7.28±0.17 and 7.20±0.20 days for animals in B, C and D groups respectively.

Conclusion and Recommendations

This result indicated significant differences ($P<0.01$) in the length of oestrous between the control group and other treatment groups. There was however, no significant differences ($P>0.05$) in the length of oestrous among animals fed the crude oil contaminated forage. The study revealed a significant difference in the oestrous cycles between the control and other treatment groups. Treatment E (which is the treatment group with the highest dosage of crude oil contamination) showed the longest oestrous cycle. This is an indication that high crude oil exposure to female rabbit could prolong their oestrous cycle.

Acknowledgement

The authors wish to thank Sylvester Izah for the editorial work.

References

Aduku, A.O. and Olukosi, J.O. (1990). Rabbit management in the tropics. Living books series. GU publications, Abuja, Nigeria.

Anderson, M.B., Lepak, K., Farinas, V. and George, W.J. (1993). Protective action of zinc against cobalt-induced testicular damage in the mouse. Reproductive Toxicology, 7: 49 – 54.

Berepubo, N.A., Johnson, N.C., Sese, B.T. (1994). Growth potential and organ weights of weaner rabbits exposed to crude oil contaminated forage. Int. J. Anim. Sci. 9: 73 – 76.

Berepubo, N.A., Nodu, M.B., Monsi, A. and Amadi E.N. (1993). Reproductive response of prepubertal female rabbits to phtoperiod and/ or male presence. World Rabbit Science. 1(2): 83 – 87.

Duncan, D.B. (1955). Multiple range and multiple F-tests. Biometrics 11: 1 – 42.

Ekweozor, I.K.E. and Snowden, R.J. (1987). The studies of the impact of a minor oil spillage in the upper Bonny Estuary. In: Proceedings: Seminar on the Petroleum Industry and the Nigeria Environment. NNPC/FMOW&H, 11 – 14 Nov., Kaduna Nigeria. pp. 2320 – 242.

Fielding, D. (1991). Rabbits. The tropical agriculturalist, CTA/Macmillian Education Ltd. 21 – 25.

Hafez, E.S.E. (1960). Sex drive in rabbits. Southwest Vet. 14: 46 – 49.

Ibeawuchi, J.A. and Fajuyitan, A.O (1986). The rabbit industry: A source of meat for human consumption. Nigeria Livestock Farmers 6:26 – 27.

Johnson, N.C. (1994). Growth performance and organ weights of weaner rabbits exposed to crude oil contaminated forage. M.Phil. Thesis, Department of Animal Science, Rivers State University of Science and Technology, Port Harcourt.

Kadafa, A.A. (2012). Oil Exploration and Spillage in the Niger Delta of Nigeria. Civil and Environmental Research, 2(3): 38 – 51.

Kamwanja, L.A. and Hauser, E.R. (1983). The influence of phtoperiod on the onset of puberty in female rabbit. J. Anim. Sci. 56: 1371 – 1375.

Mattison, D.R. (1983). Reproductive toxicology. American Journal of Industrial Medicine 4: 1 – 2.

Nigeria National Petroleum Corporation (NNPC), (2011). 2011 Draft Annual StatisticalBulletin. Corporate Planning & Strategy Division (CP&S). 1st Edition.

Nwilo, P.C. and Badejo, O.T. (2005). Impacts and Management of Oil Spill Pollution along the Nigerian Coastal Areas. www.fig.net/pub.figpub36/chapters/chapter_8.pdf

Ogbeibu, A.E. and Iyobosa S.E. (2013). Illegal crude oil bunker in Nigeria: proximate and ultimate factors. Journal of Nigeria Environmental Society, 7(4): 1 – 18.

Ogodo, A.D. and Esemuede, I.H. (2013). Crude oil theft and its environmental consequences: The way forward. Journal of Nigeria Environmental Society, 7(4): 66 – 37.

Ohimain, E.I. (2013). The challenges of liquid transportation fuels in Nigeria and the emergence of the Nigerian automotive biofuel programme. Research Journal of Applied Sciences, Engineering and Technology 5(16): 4058 – 4065.

Ohimain, E. I. (2012). The benefits and potential impacts of household cooking fuel substitution with bio-ethanol produced from cassava feedstock in Nigeria. Energy for Sustainable Development. 16: 352 – 362

Ohimain, E. I. (2011). Environmental impacts of the proposed 1MWe wood gasification power plant in Nigeria. Nigeria Journal of Agriculture, Food and Environment, 7(4): 12 – 18.

Ohimain, E. I. (2010). Emerging bio-ethanol projects in Nigeria: Their opportunities and challenges. Energy Policy. 38: 7161-7168.

Okere, H.C. (1997). Oestrous behavior and reproductive performance of female rabbits fed raw soyabean supplemented diets. M.Sc. Thesis, Department of Animal Science, Rivers State University of Science and Technology, Port Harcourt.

Okorie, J.U. (1983). A guide to livestock production in Nigeria. Macmillian publication, London.

Paufler, S. (1985). A compendium of rabbit production appropriate for conditions in developing countries Eschborn, Germany. Pp. 115 – 130.

Plyaschenko, S.I. and Sidorov, V.T. (1987). Stresses in form animals. Aropromizdat, Moscow. Pp. 122.

Steel, R.G.D. and Torrie, J.H. (1981). Principles and procedures of statistics. 2nd edition. McGraw Hill, Singapore.

United Nations Environment Programme (UNEP) (2011). Environmental assessment of Ogoniland.

White, I.C. (1975). Toxicity testing of oils and oil dispersants. Aquatic pollution in relation to the protection of living resources. Bioassays and Toxicity Testing. Bull FAO/UN, 3(4):168

Zemjanis, R. (1980). Repeat breeding or conception failure in cattle; In: Current Therapy in Theriogenology: diagnosis, treatment and prevention of reproduction diseases in animals, D.A. Morrow (ed). W.B. Saunders Co., Philadelphia, London and Toronto. Pp. 205 – 213.

Tables

Table 1: Graded oil composition in the forage

Treatment No	% oil composition of forage	Mass composition
Treatment A	Plain feed with contamination (0.0% crude oil)	-
Treatment B	0.05% crude oil contamination	0.5g crude oil/kg of forage
Treatment C	0.10% crude oil contamination	1.0g crude oil/kg of forage
Treatment D	0.15% crude oil contamination	1.5g crude oil/kg of forage
Treatment E	0.20% crude oil contamination	2.0g crude oil/kg of forage

Table 2: Oestrous behavior of female rabbits fed crude oil contaminated forage

Oestrous Parameter	Treatment				
	A	B	C	D	E
Frequency of Oestrous (No	4.63 ± 0.18^{a}	3.50 ± 0.22^{b}	2.60 ± 0.40^{c}	1.80 ± 0.20^{d}	1.50 ± 0.29^{d}
Length of oestrous (Days)	6.59 ± 0.12^{b}	7.35 ± 0.11^{a}	7.28 ± 0.17^{a}	7.20 ± 0.20^{a}	7.38 ± 0.31^{a}
Duration of oestrous (Hrs)	6.71 ± 0.11^{a}	6.65 ± 0.20^{a}	6.40 ± 0.19^{ab}	6.00 ± 0.22^{b}	5.25 ± 0.14^{b}
Heat intensity	4.04 ± 0.14^{a}	2.20 ± 0.21^{b}	1.63 ± 0.19^{b}	1.60 ± 0.24^{b}	0.50 ± 0.00^{c}

Within row mean ±SEM with different superscripts are significantly different ($P<0.05$)