

Antioxidants And Oxidant Statuses Of Growing Pigs Fed Graded Levels Of Crude Oil-Contaminated Diets

Johnson, N. C., Joy A. Okejim., J. A., Amakiri, A. O.

Rivers State University, Department of Animal Science,
Nigeria, 234 909 462 4825
ntinya@alumni.uoquelpa.ca

Federal College of Education (Technical) Omoku, Department of Vocational Studies,
Nigeria, 234 805 856 5900
okejinjoy@gmail.com

Rivers State University, Department of Animal Science,
Nigeria, 234 803 552 3285
aoa4u@yahoo.com

Abstract: This study investigated the impacts of ingested graded levels of crude oil-contaminated diets on the antioxidant and oxidant statuses in the growing pig. 24 growing pigs weighing on average 8 ± 1.1 (mean \pm SD) kg of body weight (BW) were used in the investigation. Pigs were randomly assigned to six dietary treatment groups of 4 pigs per treatment: 0g, 2g, 4g, 6g, 8g and 10g of crude oil/kg of diet, respectively. The experiment lasted for four weeks. Blood samples were collected from all treatment groups into EDTA treated tubes and immediately snap frozen for later anti-oxidant and oxidant analyses. Antioxidants: glutathione (GSH), superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GSH-P-x) and the oxidant malondialdehyde (MDA), respectively were analyzed for. There were no significant ($P > 0.05$) differences in all the antioxidants measured. Similarly, there were no significant ($P > 0.05$) differences in MDA contents for all dietary treatment groups. It was therefore concluded that crude oil contaminations up to 10g/kg of diet does not induce oxidative stress in growing pigs.

Key words: Antioxidants, Crude Oil-Contaminated Diets, Oxidants and Pig.

1 Introduction

Fast growing animal species, such as the growing pig are often susceptible to environmental stressors, including their diet types leading to sudden deaths usually referred to as 'sudden death syndrome' in swine production [7]. This is mainly due to the oxidative stress the animal undergoes during their growth processes. To this end therefore, the animal has developed its defensive systems or mechanisms to defend its self against oxidative stress [4]. One of the major known defensive systems of the body is GSH. GSH is the major antioxidant that aids to preserve all other body antioxidants and therefore has been termed 'mother of all antioxidants', master detoxifier working in concert with the immune system to maintain animal health [4]. Some of the cohorts of the GSH defense system are SOD, CAT and GSH-P-x. SOD catalyzes the dismutation or partition of superoxide radicals into oxygen and hydrogen peroxide. CAT converts hydrogen peroxide to carbon dioxide while GSH-Px also protects the animal against oxidative stress due to reactive oxygen species [9]. It also aids in re-generating GSH from its oxidized form [10]. Antioxidants therefore have a special role as they are the major players in health-related conditions of the animal [12]. Malondialdehyde (MDA) is the major byproduct of oxidative stress. Crude oil being a toxicant may therefore reduce antioxidants levels while increasing MDA levels due to oxidative stress induced by the ingested crude. Therefore, the objectives of this study are to evaluate the effects of ingestion of graded levels of crude oil-contaminated diets on the statuses of GSH, SOD, CAT, GSH-Px and MDA.

2 Materials and methods

Animals and management

24 growing pigs of average BW of 8 ± 1.1 (mean \pm SD) kg were obtained from Cape Farms, Irete, Imo State, Nigeria and humanely transported to the venue of the study. The animals on arrival at the Animal Wing, Rivers State University Teaching and Research Farm were randomly assigned to their individual experimental pens. Before their arrival the animals' pens were thoroughly washed with hypochlorite /disinfected and allowed to dry. The animals were given 14-days to acclimatize to their new environment. At this time the animals were given ivermectin injection sub-cutaneous and amoxyciline antibiotic injection intramuscularly to ensure good health status before commencement of study and fed similar grower diet. At the end of the adaptation period, the animals were offered their crude oil-contaminated diets at 5% of their BW and fed twice daily at 09:00h (half of the daily meal) and 16:00h, respectively. As expected, water was provided ad libitum via low pressure nipples and pens were constantly kept cleaned throughout the experimental duration.

Crude oil treatment and experimental diets

The crude oil used in this study is specifically Bonny Light obtained from the Nigerian Agip Oil Company limited. Before the commencement of study, the crude oil was exposed to sunlight for 24 h in a shallow pan to enable the escape of the light volatile fractions via evaporation thereby ensuring a stable product that simulates crude oil natural form during oil spillage and pollution [8]. Six corn-soybean

meal-based diets that were isocaloric and isonitrogenous to meet or exceed the [7] recommended nutrient requirements of growing pigs of 10 – 20 kg BW were used in the study. Although the diets had similar nutrient levels they differed in their dietary crude oil contents as: diet1, the control diet (0g crude oil), diet 2, (2g crude oil), diet 3, (4g crude oil), diet 4, (6g crude oil), diet 5, (8g crude oil) and diet 6, (10g crude oil)/kg of diet, respectively. Animals received their respective experimental diets for 4 weeks (28d).

Data collections/analyses and experimental design

At the end of the experiment blood samples were collected from all animals in each dietary treatment group into EDTA treated tubes and immediately snaps frozen for antioxidants and oxidant later analyses. Antioxidants analysed for were GSH, CAT, SOD, and GSH-Px whereas oxidant analysed for was MDA, respectively. GSH, GSH-Px and MDA were analysed for according to the methods of [2]. CAT was analysed for according to the method of [1] and SOD was analysed for by the method of [6]. The experimental data were analyzed as a CRD. Data were subjected to analysis of variance (ANOVA) using PROC GLM of SAS (SAS Inst. Inc., Cary, NC) according to the experimental model: $Y_{ij} = \mu + D_i + E_{ij}$; where Y_{ij} is the observation, μ = overall mean common to all treatments, D_i = the effect of the i^{th} diet and E_{ij} = the error term. Means were compared using Tukey's test and α -level of 0.05 was used for all statistical comparisons to represent significance.

3 Results and Discussion

Animals readily consumed their respective diets without any signs of rejection throughout the experimental duration. This was also an indication that the animals ingested their dietary crude oil contents of their diets, respectively. Animals were also seen to be healthy during the study period. The results of the antioxidants and oxidant are shown in Table 1.

Table 1. Antioxidants (GSH, SOD, CAT and GSH-Px) and oxidant (MDA) Statuses of Pigs Fed Graded Levels of Crude Oil-Contaminated Diets.

Item	DIETS				SEM	P-value
	Diet 1 n = 4	Diet 2 n = 4	Diet 3 n = 4	Diet 4 n = 4		
GSH (mg/dl)	4.75	4.50	4.50	4.75	5.00	0.220
SOD (U/mg)	2.10	1.98	2.00	2.00	5.00	0.607
CAT (U/mg)	13.97	14.00	13.87	13.95	1.99	0.958
GSH-Px (nmol/dl)	1.50	1.50	1.27	1.25	14.04	0.880
MDA (nmol/ml)	0.73	0.70	0.71	0.73	1.25	0.976

As shown in the Table 1 above, there were no significant ($P > 0.05$) differences among all the antioxidants (GSH, SOD, CAT and GSH-Px) as well as in the oxidant (MDA) assessed for all dietary treatment groups. Antioxidants protect the animals during oxidative stress and MDA is the major byproduct of oxidative stress [3], [5]. Therefore, for the fact that the concentrations of the antioxidants were similar across all dietary treatment groups is an indication that the animals did not experience any oxidative stress. Antioxidants protect cells, tissues and eventually animals' organs by reacting with free radicals [7]. In this study, the concentrations of the antioxidants were similar for all treatment groups. Here, it is very imperative to note that ingested diets are mostly responsible in stimulating the antioxidants that can be synthesized endogenously by the body as the animal reacts to the diet's consumption leading

to antioxidant productions that react with free radicals, leading to their reduced values in the process of detoxifying [5], [7]. More importantly when so produced work synergistically together in the animals' body to achieve their defense functions [12]. To this extent therefore these antioxidants work in concert since they complement each other function. SOD in most cases is the first level of antioxidant defense as superoxide radical is the major radical produced in physiological conditions in the cell [11]. SOD is mainly responsible for converting superoxide radicals into hydrogen peroxide and oxygen. GSH-Px and CAT are mainly responsible for the conversion of the hydrogen peroxide to water. GSH is mostly required as a coenzyme by a variety of enzymes, such as GSH-Px and also mostly responsible for some crucial life processes, including detoxification of foreign bodies and removal of hydroperoxides and other free radicals [11]. The findings that antioxidant levels were similar for all dietary treatments and as such no oxidative stress occurred are further supported by the observed similar concentrations of MDA in all animals' dietary treatment groups. This is true because MDA levels signifies oxidative stress and thus it is a major yardstick in measuring the degree of oxidative stress an animal has been exposed to [3], [5]. In this case all animals in all treatment groups had similar low concentrations of MDA indicating absence of oxidative stress for all dietary groups.

4 Conclusion

Growing pigs can ingest crude oil in their diets up to 10g/kg of diet without experiencing any oxidative stress as measured by oxidative stress biomarkers namely antioxidant statuses and MDA concentrations.

5 References

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