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Effect of Dietary Garlic Powder (*Allium sativum*) Supplementation on Lipid Stability of Mutton

Alamuoye Oluwatoyin F
Department of Animal Science, Ekiti State University, Ado-Ekiti, Ekiti State, Nigeria

Abstract
The aim of the study was to investigate the effects of dietary garlic powder supplementation on the muscle cholesterol and lipid stability of primal cuts of West African Dwarf (WAD) rams. Forty WAD rams of average initial body weight of ±15kg, were randomly allotted into five treatment groups in which garlic powder was supplemented at 0% (control), 2%, 4%, 6% and 8% in the diets respectively. After 90 days of feeding, three WADS rams were randomly selected from dietary groups, weighed, slaughtered and dissected into primal cuts (loin, rib, round and shoulder) and *Psoas major* and *Semi membranosus muscles* were dissected. Lipid oxidation was assessed for raw and cooked cuts at different storage periods (7, 14, 21 days). The study showed that feeding garlic powder supplemented diets significantly ($P<0.05$) reduced muscle cholesterol. Lipid oxidative stability of meat samples was assessed by determining TBARS. The TBARS values were significantly ($P<0.05$) influenced by dietary treatments in raw and cooked cuts at 7, 14 and 21 days' storage respectively. Lowest TBARS values were observed in raw and cooked cuts derived from rams fed treatment 5(8% garlic powder).

Keywords: garlic powder, muscles, cholesterol, lipid oxidation

Introduction
Lipid oxidation is a main cause of meat deterioration in terms of quality, as a result of autoxidation of unsaturated fatty acids within the tissue which alters its wholesomeness and nutritional value (Pearson *et al.*, 1983) [1]. Lipid oxidation determines shelf life of meats and meat products due to loss of cellular antioxidant defences (Morrisey *et al.*, 1994) [2]. After 4 or 7 days of storage, lipid oxidation increases in meat (Luciano *et al*., 2009) [3] due to oxidation reaction resulted from physiological process such as a conversion of the red muscle myoglobin (pigment) to brown metmyoglobin and facilitates rancid odours and flavours from the degradation of the unsaturated fatty acids in the tissue (Wood and Enser, 1997; Velasco and Williams 2011) [4, 5]. Synthetic antioxidants are commonly used in meat industry to extend meat shelf-life but consumer's concern over their toxicity geared research towards natural sources of antioxidants (Karami *et al*., 2010) [6].
Consumers' demands for healthier and higher quality meat and meat products of lower fat and cholesterol contents (Scollan *et al.*, 2006) [7], which can be achieve through modifications of the lipid composition of muscle cells membranes by the intake of dietary antioxidant to improving oxidative stability (Barroeta and Cortinas, 2002) [8]. More so, meat is prone to oxidative rancidity and discolouration due to its nutritional contents, enzymatic action and the presence of microorganisms (Forrest *et al*., 2001) [9] this can be controlled through dietary supplementation of spice and herbs. Feeding garlic powder could be one of the dietary sources of improving meat quality. Garlic (*Allium sativum*) is widely used as a spice and herb (Lawson, 1996) [10], lowers serum and tissue cholesterol levels (Stanacev *et al*., 2012) [11] and prevents free radical damage (Chung, 2006) [12]. Garlic possessed antioxidant compounds such as flavonoid and sulfur containing compounds (Leonarduzzi *et al*., 2002) [13]. Garlic supplementation has antioxidant effects, which lowers the thiobarbituric acid re- active substance value and protect lipid oxidation (Hanieh *et al*., 2010) [14].
The study is aimed at assessing the muscle cholesterol contents and oxidative stability of lipids at different storage intervals of raw and cooked muscle types of West African Dwarf rams fed garlic powder supplementation.

Materials and methods
Experimental animals and treatments
Forty yearling West African Dwarf rams with average live weight of 15kg, were randomly allocated into five experimental diets in a Completely Randomized Design. Garlic powder was supplemented at 2%, 4%, 6% and 8% in diet 2, 3, 4 and 5 respectively while diet 1 (control diet) had 0% of garlic powder (table 1). Animals were fed basal diets, with inclusion of *Panicum maximum* and cassava peels, feed and water provided ad-libitum for the period of 90days. Feed was withdrawn for 18 hours prior slaughtering. Animals were slaughtered after mobilization, eviscerated, carcass divided into two halves through the mid-plane. The right halved hot carcass was dissected into standard cuts (shoulder, rib, loin, flank and round) following the reference points. *Psoas major* and *Semi membranosus* were dissected from the carcasses. The cuts were chilled at 4°C overnight prior analyses.

Determination of muscle cholesterol
The determination of cholesterol was done spectrometrically by the method of Paradkar and Irudayaraj (2002) [15].
Determination of TBARs: Lipid stability of primal cuts were assessed for their thiobarbituric acid reactive substances (TBARS) at 7, 14 and 21 days' storage according to methods of Buege and Aust (1978) [16] as described by Wattanachant, *et al. (*2008) [17]. The TBARS were calculated from a standard curve of malondialdehyde (MDA) and

expressed as mg MDA/kg sample.

Statistical analysis

All data were collected in triplicate and statistically analyzed using SAS (2002) [18].

Results and Discussion

Table 2 showed the results of the muscle cholesterol content of raw and cooked muscles derived from WAD rams fed garlic powder supplemented diets. The cholesterol content of raw Psoas major and Semi-membranous muscles was significantly different (P<0.05) between dietary treatments. The cholesterol content of raw Psoas major was closer to those obtained for raw Semi-membranous muscles. The cholesterol content increased in the cooked muscles as compared with raw muscles, cholesterol contents of both cooked Psoas major and Semi-membranous muscles reduced significantly (P<0.05) among dietary treatments at the increment in the level of garlic powder.

The cholesterol values obtained in raw muscles (*Psoas major* and *Semi membranosus*) of treatments 4 and 5 were closer to the values of 66 mg/100g reported by Williams (2007) [19] in mutton while the muscles cholesterol values of raw muscles (*Psoas major* and *Semi membranosus*) of rams on treatments 1, 2 and 4 were closer to the value of 76.12 mg/ 100g reported by Mustafa (2013) [20] for raw mutton. The results of cooked muscles (*Psoas major* and *Semi membranosus*) in the present study were lower than the value of 130mg/100g reported by Williams (2007) [19] for cooked mutton. The muscle cholesterol values obtained for raw and cooked muscles in the study were within the range 40 and 90mg/100g reported in several studies (Valsta *et al*., 2005; Honikel, 2009) [21, 22]. The reduction in the cholesterol contents of raw and cooked *Psoas major* and *Semi membranosus* of treatments 4 and 5 fed rams affirmed the assertion of Konjufca *et al*. (1997) [23] that the reduction in meat cholesterol might be attributed to the reduced cholesterol biosynthesis mediated through the changes in the enzyme (HMG-CoA reductase), this is responsible for regulating cholesterol metabolism due to the principle behind the pharmacological action of garlic containing sulphur compounds that oxidize NADPH which is necessary for cholesterol synthesis. The study also, followed the trends observed by Dieumou *et al.* (2012) [24] in birds fed garlic extracts. More so, decreased in the cholesterol contents of the muscles of WAD rams fed garlic supplemented diets aligned with Rice-Evans *et al.* (1997) [25] that phenolic compounds from spices and herbs have an antioxidative potential due to their possibility to act as a radical scavenger.

Table 1: Muscle cholesterol content of meat products of WAD rams fed garlic powder supplemented diets (mg/dl)

Item	Muscle type	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5
		0% garlic	2% garlic	4% garlic	6% garlic	8% garlic
Raw	*Psoas major*	74.33 ± 0.08^{a}	71.00 ± 1.00^{b}	70.33 ± 1.53^{b}	66.68 ± 1.53^{c}	67.67 ± 0.52^{c}
	Semimembranosus	74.67 ± 1.16^{a}	71.86 ± 1.71^{b}	70.26 ± 1.62^{b}	66.70 ± 1.16^{c}	65.39 ± 1.34^{c}
cooked	*Psoas major*	80.00 ± 0.00^{a}	78.00 ± 0.00^{b}	73.33 ± 1.16^{c}	70.67 ± 1.01^{c}	70.67 ± 1.16^{c}
	Semimembranosus	76.93 ± 1.30^{a}	74.89 ± 1.80^{ab}	73.67 ± 0.83^{b}	71.67 ± 0.68^{b}	70.78 ± 0.28^{b}

Mean ± standard deviation; a, b, c means of different superscripts on same row are significantly (P<0.05) different

TBARS values increased significantly (p<0.05) within seven days of storage thereafter decreased significantly (p<0.05) at 14 to 21 days' storage (figures.1-8) in both raw and cooked cuts of treatments 2, 3, 4 and 5 respectively. The TBARS values of raw rib, loin and leg cuts were similar in the treatments 4 and 5 on day 7 and 14 respectively but significantly differ (P<0.05) on day 21. The TBARS significantly (P<0.05) increased during storage but remained stable in raw and cooked cuts obtained from WAD rams fed treatment 5 (8% of garlic supplementation) on 21 days. TBARS values decreased significantly (P<0.05) at 7, 14, and 21-day storage periods in all raw and cooked primal cuts derived from WAD rams fed garlic powder supplemented diets. The study showed that TBARs values of raw and cooked cuts derived from control fed rams were significantly higher (P<0.05) than other test diets (figures 1 – 8), indicating that the rate at which endogenous lipid oxidation occur within muscle content was higher and that the quantity of antioxidants in control diet was not adequate to suppress or delay the formation of free radicals which hastened rancidity of derived products during storage.

The TBARS values obtained in the present study fell within those reported in other studies on storage of lamb's meat with the use of antioxidants (Luciano *et al.*, 2013; Rippol *et al*., 2013; Memmo, 2014) [26, 27, 28]. The results of TBARS in the current study aligned with those obtained by Faustman and Cassens (1989) [29] for frozen loin. Also, the study followed the trend of Memmo (2014) [28] who used antioxidants in storage of lamb's meat. The TBARs values obtained at 21- day storage in raw and cooked primal cuts of treatment5 fed rams were higher than values above about 0.5 which is considered as critical point, this indicates that such products were tending toward a rancid odour and taste which can be detected by consumers (Wood *et al*., 2008) [30]. Studies have shown that meat shelf-life and quality can be improved by incorporating natural antioxidants into animal diets. The positive effects of natural antioxidants on meat characteristics may retard lipid oxidation according to Velasco and Williams (2011) [5]. The rate of lipid oxidation was controlled in raw and cooked cuts as the level of garlic powder supplementation increased in the diets. This is in support of De Winnie and Dirink (1996) [31] and Grau *et al*. (2001) [32] who reported higher meat oxidative stability with the dietary addition of vitamin E. This also concurs to the report of Berges (1999) [33] that rate of lipid oxidation could be effectively controlled with antioxidants.

The TBARS values obtained in the study for raw and cooked cuts at 7, 14 and 21 days' storage were considerably lower than 0.9mg malonaldehyde per kg suggested as critical point of lipid oxidation for meat and meat products by Atay *et al*. (2011) [34]. This collaborate with the findings of Min *et al*. (2008) [35] that TBARS values of raw beef loin significantly increased during 7-d storage due to high free iron content and high lipoxygenase-like activities by ferrylmyoglobin. This also agrees with Rong *et al*. (2009) [36] that the TBARS values increased as storage time increased. This study did not align with the report of Min *et al*. (2008) [35] who observed no change in TBARS values of raw pork, and chicken breast and thigh meats during a 7-d storage period.

The TBARS values obtained in the study for raw and cooked primal cuts at 7, 14 and 21 days' storage were below the MDA concentration between 1.0 and 2.0 MDA mg/kg indicated by Verme and Sahoo (2000) [37] as threshold values for rancidity. The TBARS values obtained in raw and cooked cuts during 21days storage period were lowered in treatment 5 (8% garlic) fed WAD rams than other test diets showing that garlic supplementation could hinder or delay propagation of free radicals and auto-

oxidation due to presence of peroxide-scavenging properties in garlic Mariutti *et al*. (2008) [38]. This is in support of Jamilah *et al*. (2009) [39] that natural antioxidants had great positive effect in delaying the lipid oxidation of the muscles of different animal species and extending the storage life of fresh meat and meat products. Low TBARS values during storage were as a result of low free iron content and high ferric ion reducing capacity in the meat (Min *et al*., 2008) [35].

The TBARS values obtained for all cooked cuts (ribs, loin, leg and shoulder) were higher than the values obtained in raw cuts; this might be due to the effect of thermal processing on the meat composition, which resulted to increase at the rate of lipid oxidation process of the carcasses This is supported by Wu and Sheldon (1988); Ahn *et al*. (1993) [40, 41] and Min *et al*. (2008) [35] that lipid oxidation in meat products are usually accelerated by cooking due to the presence of heat-stable ferric ion reducing capacity. The study revealed that antioxidant activity of garlic powder was on the positive as the TBARS values were significantly decreasing with an increase in the dietary level of garlic powder supplementation in all cooked cuts when compared with cooked control samples. This implies that the rate of lipid peroxidation was rapid in the cooked meat than raw meat, thereby reducing the shelf life of such products but additional effect of antioxidant on the meat through dietary means reduced rancid odour, this agreed with Mariutti *et al*. (2008) [38] that reduction in TBARS using garlic and ginger is related to peroxide-scavenging enzyme activity

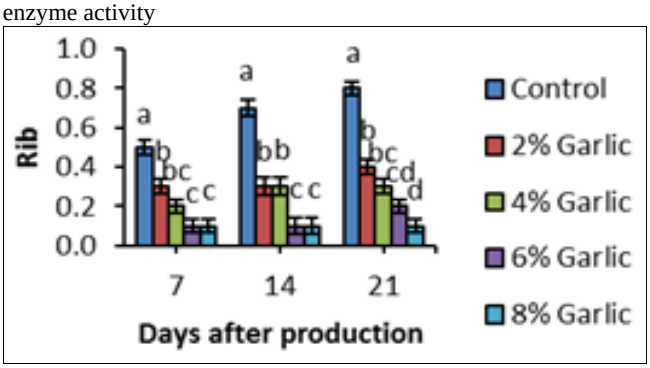

Fig 1: TBARS (mg MDA/kg meat) of raw rib cut of WAD ram at different storage days, a, b, c, d means with different letters on dietary treatments are significantly different (p<0.05); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

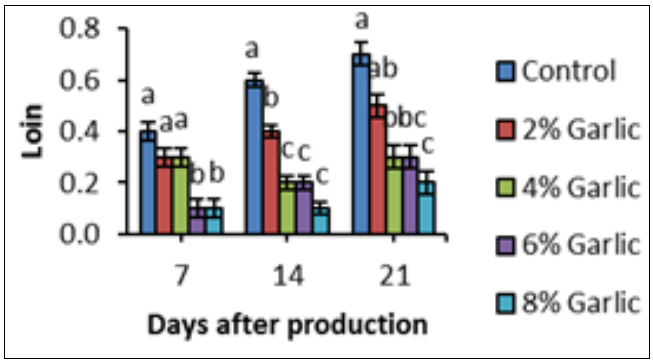

Fig 2: TBARS (mg MDA/kg meat) of raw loin cut at different storage days, means with different letters on dietary treatments are significantly different (p<0.05); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

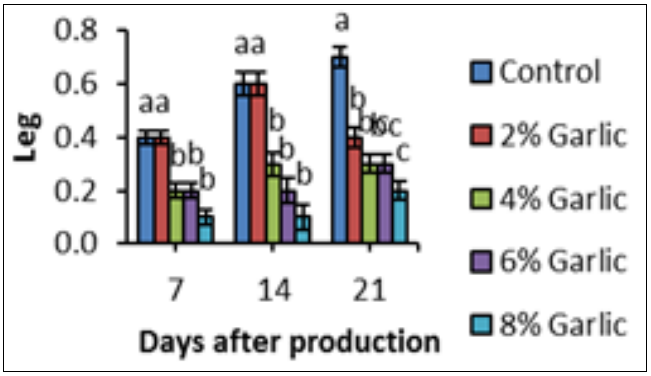

Fig 3: TBARS (mg MDA/kg meat) of raw leg cut at different storage days, a, b, c means with different letters on dietary treatments are significantly different (p<0.05); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

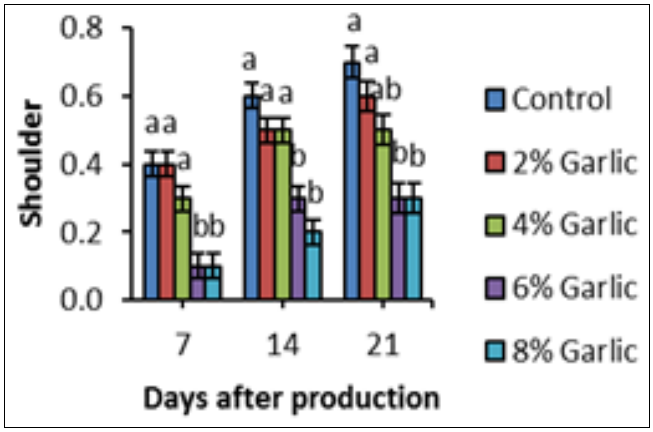

Fig 4: TBARS (mg MDA/Kg meat) of raw shoulder cut at different storage days, a, b means with different letters on dietary treatments are significantly different (p<0.05); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

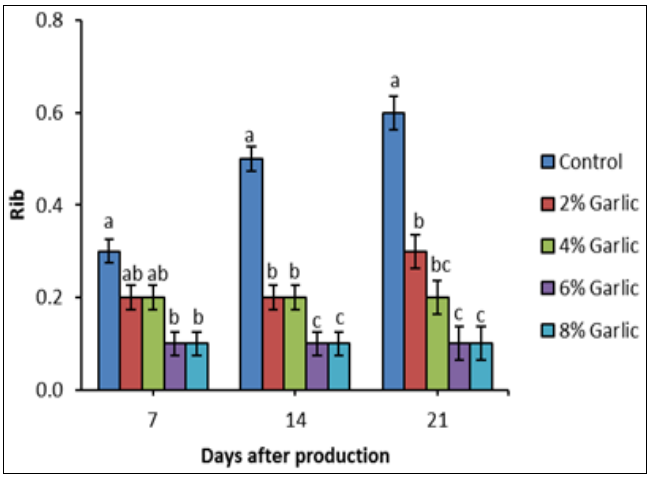

Fig 5: TBARS of cooked rib cut at different storage days, a, b, c, means with different letters on dietary treatments are significantly different (p<0.05); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

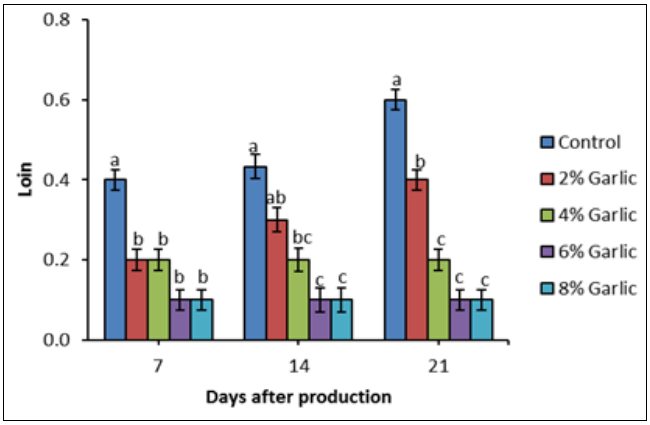

Fig 6: TBARS (mg MDA/Kg meat) of cooked loin cut at different storage days, a, b, c, means with different letters on dietary treatments are significantly different ($p<0.05$); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

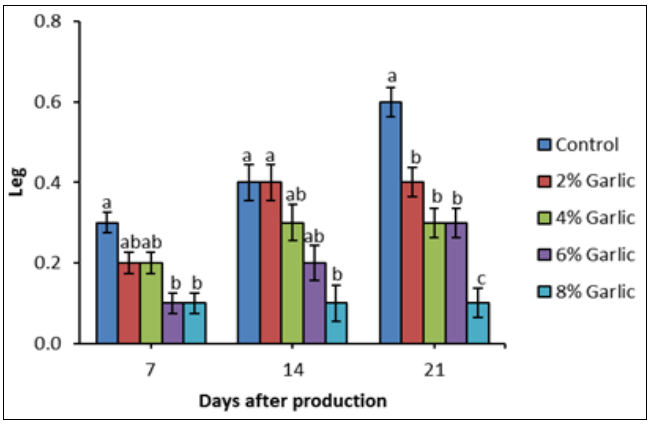

Fig 7: TBARS (mg MDA/Kg meat) of cooked leg cut at different storage days, a, b, c, means with different letters on dietary treatments are significantly different ($p<0.05$); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

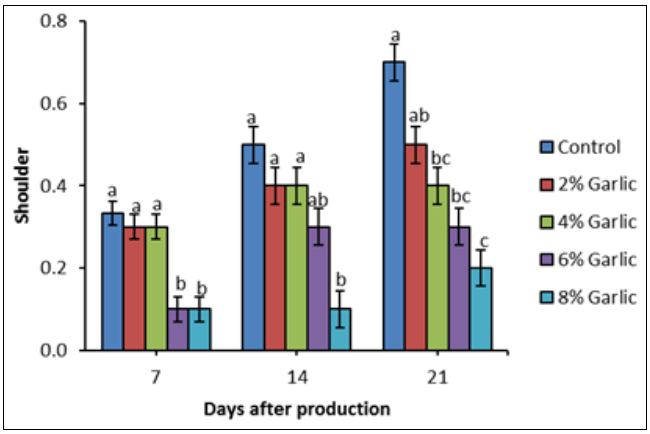

Fig 8: TBARS (mg MDA/Kg meat) of cooked shoulder cut at different storage days, a, b, c, d means with different letters on dietary treatments are significantly different ($p<0.05$); control (treatment 1), 2% garlic (treatment 2), 4% garlic (treatment 3), 6% garlic (treatment 4), 8% garlic (treatment 5)

Conclusion

It was concluded that dietary garlic powder at 8% inclusion reduced cholesterol contents of *Psoas major* and *Semi-membranous* muscles tremendously in both raw and cooked states. Likewise, it enhanced retardation of lipid oxidation in raw and cooked primal cuts at 21-day post-production.

Application

Feeding garlic powder supplementation to sheep could be used as natural source of dietary antioxidants for inhibiting lipid oxidation in red meat, which tends to prolong meat shelf-life. Also, serve as safer source of red meat to the consumers without risk of ill-health attributed to consumption of red meat.

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