Scientia Africana, Vol. 20 (No. 3), December, 2021. Pp 85-94 *https://dx.doi.org/10.4314/sa.v20i3.8*
© Faculty of Science, University of Port Harcourt, Printed in Nigeria *ISSN 1118 – 1931*

ASSESSMENT OF SOME BIOCHEMICAL INDICES IN RED MEAT TENDERED WITH POTASH AND EXTRACT OF *OCIMUM BASILICUM*

[1]Ugbeni, O. C., [2]Ibironke B. A. and [3]Ikhelegbe-Ojo D. I.

University of Benin, Benin City, Nigeria
Corresponding Author: osezele.ugbeni@uniben.edu

Received: 09-10-2021
Accepted: 09-11-2021

ABSTRACT

Some changes in biochemical composition in red meat boiled with potash and extracts of Ocimum basilicum was the crux of this research. Fresh beef were divided evenly into 4 groups labeled A-D. Group A was boiled with distilled water, group B was boiled with potash in distilled water, group C was boiled with extract of Ocimum basilicum while group D was boiled with a mixture of potash and Ocimum basilicum. All samples were boiled for fifteen minutes. Samples were taken for analysis at zero hour and after every 48hrs. The samples were stored in a freezer at -4ºC. The pH, malonedialdehyde concentration, acid value, total protein, percentage metmyoglobin and heme iron were determined. The pH values showed significant increase ($p<0.05$) in red meat boiled with both potash and extracts of Ocimum basilicum from 0 to 144 hours. There was no significant difference ($p>0.05$) observed in the malondialdehyde concentration and acid value for the duration of the experiment. A significant ($p>0.05$) decrease in the total protein content was observed after 96 hours in red meat boiled with potash. The results for percentage metmyoglobin content showed a significant difference ($P<0.05$) and a decrease in red meat boiled with Ocimum basilicum and both potash and Ocimum basilicum after 96 and 144 hours. The results for heme iron concentration showed no significant difference ($p>0.05$) in red meat boiled with Ocimum basilicum from 0 to 144 hours while an increase was observed in red meat boiled with potash after 96 hours.

INTRODUCTION

Tenderization of meat is a process of degrading the structural meat proteins in order to reduce the toughness of the meat. The concept of Tenderization is more applicable to red meat, because it has high toughness and has the lowest correlation with white meat. (Bekhit *et al*., 2014). Some chemical agents are used in this regards. The use of potash and more recently paracetamol is becoming a common phenomenon **in some parts of African countries** including Nigeria. **National Agency for Food and Drug Administration and Control (NAFDAC)** has raised serious concerns about the safety of consumption of such meat (Guardian, 2020). Potash, which is also known as kaun, akanwu, keun or kanwa in different Nigerian languages is a type of lake salt (sodium carbonate) that is dry and hydrated. **The name is derived from pot- ash,** and this refers to plant ashes soaked in water in a pot, which was the primary means of manufacturing the product before the industrial era. It is an alkaline salt that is a mixture of different substances including impurities such as clay, sand, and metals like magnesium, silicon, iron, aluminum, calcium, and titanium (Momoh *et al*.,2019). However, potash has a very low quantity of potassium, compared to sodium and it is the second most popularly used salt in Nigeria (Imafidon *et al*, 2016). It is used for

fertilizer production, fire extinguisher, tooth ache relief, preservatives due to its anti fungal properties, medicinal purposes and especially culinary purposes **where it is used in the preparation** of some local dishes and some certain foods such as meat, beans etc. to tenderize them and shorten the cooking time. Studies have revealed that excessive consumption of potash may lead to its accumulation that could cause severe and irreparable damage to the kidney and disrupt normal body functions which may eventually lead to loss of life. (Bankole *et al*, 2015).

Ocimum basilicum (Sweet Basil) is one of the most important crops with essential oils as well as polyphenols, phenolics, flavonoids and phenolic acids (Silva *et al*, 2016). It is part of a group of medicinal plants widely used in cooking and known for its beneficial health properties, possessing significant antioxidant effects and others (Silva *et al*, 2016). It has been used traditionally for the treatment of anxiety, diabetes, cardiovascular diseases, headaches, nerve pain, as anti-convulsant and anti-inflammatory, and used in a variety of neurodegenerative disorders.(Bora *et al*., 2011).

This study was conducted to evaluate the changes that occur in the biochemical properties of red meat boiled with a fixed concentration of potash and *Ocimum basilicum* in different time intervals of preservation.

MATERIALS AND METHOD

The leaves of *Ocimum basilicum* were obtained from a locality in Edo State South-South Nigeria. It was authenticated in the department of Botany university of Benin, Benin City, Nigeria and a voucher number of UBH-0461 was assigned. The leaves were dried under shade for five days. The dried leaves were ground to powder using the mechanical grinding machine. The plant extract was obtained by soaking 100 g of the ground leaf powder in 1000 ml of distilled water for three days inside a plastic container and was filtered using a muslin bag and whatman 1 filter paper. The filtrate was concentrated at 40 degrees in a water bath. The concentrate was then freeze dried and the powdered form stored in a dry container for use. 100 mg was reconstituted in 1 litre of distilled water for use. Potash solution was prepared by dissolving 3000 mg of potash in 1 litre of water and stored in a plastic container for use. About 2 kg of fresh beef was purchased in an abattoir. The beef were divided into four different portions labeled 1-4. Group 1 was boiled with distilled water, group 2 was boiled with a solution of potash, group 3 was boiled with a solution of *Ocimum basilicum w*hile group 4 was boiled with a mixture of potash and *Ocimum basilicum* in equal proportion, Each set of samples were boiled for 15 mins using a gas cylinder. The appropriate masses were weighed out for biochemical analysis at intervals of 48 hours starting from the zero hour to 144 hrs. The rest of the samples were stored in a freezer at -4^{o}c till required. The following assays were carried out: pH, heme iron content, total protein content, acid value, percentage of Metmyoglobin content, and malondialdehyde content.

*Measurement of pH value***:** pH denoting 'potential of hydrogen' or 'power of hydrogen' is a scale used to specify the acidity or alkalinity of an aqueous solution. Acidic solutions (solutions with higher hydrogen ions) are measured to have lower pH values than basic or alkaline solutions.

Scientia Africana, Vol. 20 (No. 3), December, 2021. Pp 85-94 *https://dx.doi.org/10.4314/sa.v20i3.8*
© Faculty of Science, University of Port Harcourt, Printed in Nigeria *ISSN 1118 – 1931*

pH is measured with a pH meter. Meat was boiled for 15 minutes and allowed to cool. Thereafter an electronic pH meter was placed on the meat to check for the pH of the meat.

*Determination of total protein***:** The assay was carried out using randox kits. The principle is as described by Henry *et al*, (1974). Cupric ions, in an alkaline medium, interact with protein peptide bonds resulting in the formation of a coloured complex. **The absorbance** is taken at 546nm.

Determination of percentage metmyoglobin content: The analysis of metmyoglobin content was performed as described by Krzywicki (1982). Metmyoglobin is the oxidized form of oxygen carrying some protein myoglobin. Metmyoglobin is the cause of the characteristics brown colouration of meat that occurs due to cooking. The chemistry of beef colour is due to the presence of the pigment myoglobin. Meat undergoes oxygenation when exposed to air to form oxymyoglobin. Myoglobin and oxymyoglobin have the capacity to lose an electron which turns the pigment to a brown colour called metmyoglobin.

*Determination of malondialdehyde content of beef***:** Malondialdehyde content of beef was determined using Buege and Aust method (1978). This assay is based on the reaction of malondialdehyde (MDA) with thiobarbituric acid (TBA); forming a MDA-TBA2 adducts that absorbs strongly at 535nm. In the i899presence of heat and acid, MDA reacts with TBA to produce a colored end product. The intensity of the colour at 535nm corresponds to the level of lipid peroxidation in the sample.

*Determination of heme iron contents***: Heme** iron was determined by means of acidified acetone extraction followed by spectrophotometry (Clark *et al*. 1997). The minced beef patties sample (2 g) was transferred into a 50 ml centrifuge tube and 9 ml of acid acetone mixture (90% acetone, 8% deionised water, and 2% HCl) was added. The mixture was macerated using a glass rod and allowed to stand for 1 h at room temperature. The extract was centrifuged at 2200 g for 10 minutes. The supernatant was filtered through Whatman #42 filter paper, and the absorbance was read at 640 nm against the acid acetone blank. The total pigments were calculated as haematin using the following formula (Lee *et al*., 1999): Total pigment (mg/kg) = A640 × 680 and haem iron was calculated as follows (Clark *et al*., 1997): Haem iron (mg/kg) = total pigment (mg/kg) × 8.82/100

Determination of acid value: The analysis of acid value was performed as described by AOCs (1999): The acid value is also a measure of the amount of fatty acids, which have been liberated by hydrolysis from the glycerides. The acid value is determined by directly titrating the fat in an alcoholic medium against a standard potassium hydroxide/sodium hydroxide solution.

Statistical analysis: The statistical significance was evaluated by one-way ANOVA using Graph prism 6 followed by post –hoc LSD and Tukey tests for individual comparisons. In all the results, the P-value of 0.05 was used to check for the level of significance.

RESULTS AND DISCUSSION

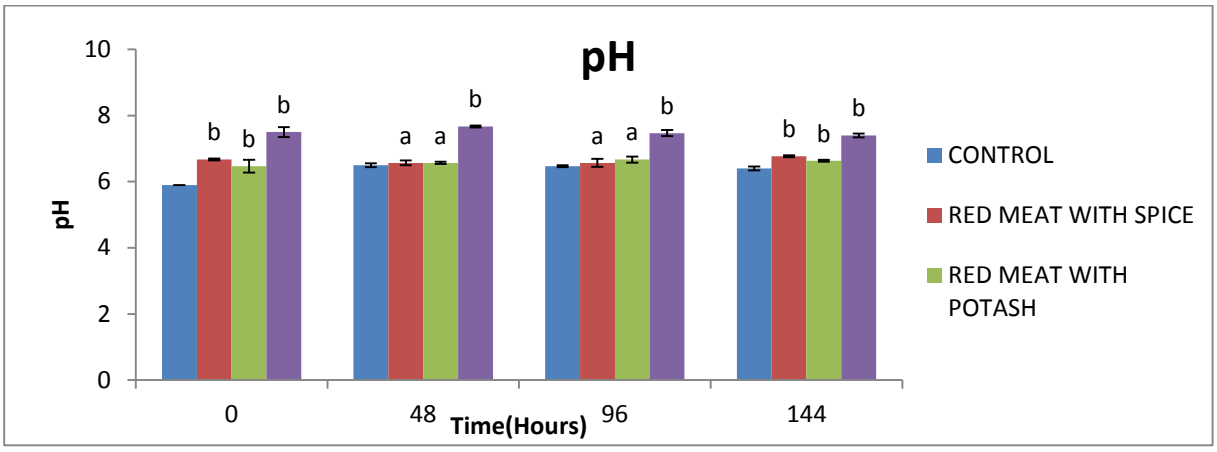

Figure 1.0: Effect of potash and extracts of *Ocimum basilicum* on the pH of red meat.

The results for pH revealed that there was significant difference ($p<0.05$) between the control and red meat boiled with potash, *Ocimum basilicum* and both potash and *Ocimum basilicum* for the first assay carried out. When the storage time increased from 0 to 96 hours, there was no significant difference ($p>0.05$) in the pH values for red meat boiled with potash and *Ocimum basilicum* respectively while that of red meat boiled with both potash and *Ocimum basilicum* increased. At 144 hours, the pH values of all the group of samples increased. Therefore, red meat boiled with both potash and *Ocimum basilicum* showed a significant increase in pH which was constant throughout the different hours of analysis. Weglarz (2010) in his work stated that meat of high quality has ultimate pH at the range of 5.4 – 5.6. Decline in meat quality starts to occur at pH > 5.8 and meat of high pH have been characterized by gummy structure, increase in water holding capacity, decrease in specific taste and reduced shelf life as bacteria grow more rapidly due to the higher pH and moisture.(Pipek *et al*., 2003).

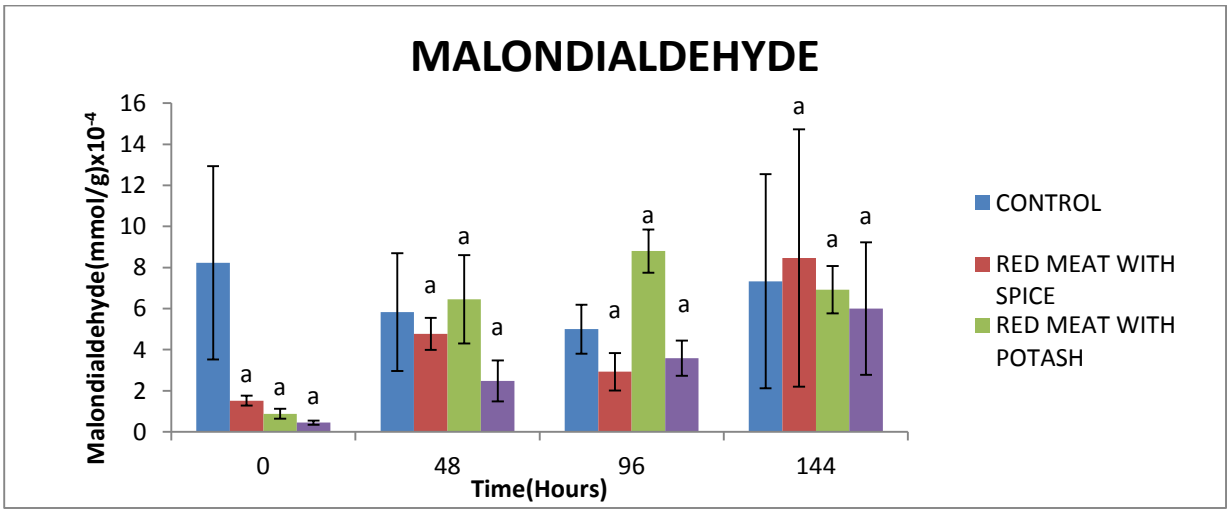

Figure 2.0: Effect of potash and extracts of *Ocimum basilicum* on the malondialdehyde content of red meat.

Scientia Africana, Vol. 20 (No. 3), December, 2021. Pp 85-94 https://dx.doi.org/10.4314/sa.v20i3.8
© Faculty of Science, University of Port Harcourt, Printed in Nigeria *ISSN 1118 – 1931*

Malondialdehyde is widely recognized as a test to monitor spoilage due to lipid peroxidation by microorganisms in bio compounds. The Malondialdehyde values showed no significant difference ($p>0.05$) between the control and red meat boiled with potash, *Ocimum basilicum* and both potash and *Ocimum basilicum* in the first assay carried out and throughout the various storage time from 48 hours to 144 hours. This indicates that the *Ocimum basilicum* extract was effective as an antioxidant against the level of malondialdehyde (lipid peroxidation marker). Similar findings in the use of *Ocimum basilicum* leaves extract in treating deltamethrin induced rat was reported by Sakr and Amoudi (2012). Also potash was effective in reducing the level of malondialdehyde. Similar findings were reported by Soleimanzadeh *et a*l. (2010) when studying the effect of potassium levels on antioxidant enzymes and Malondialdehyde content under drought stress in sunflower.

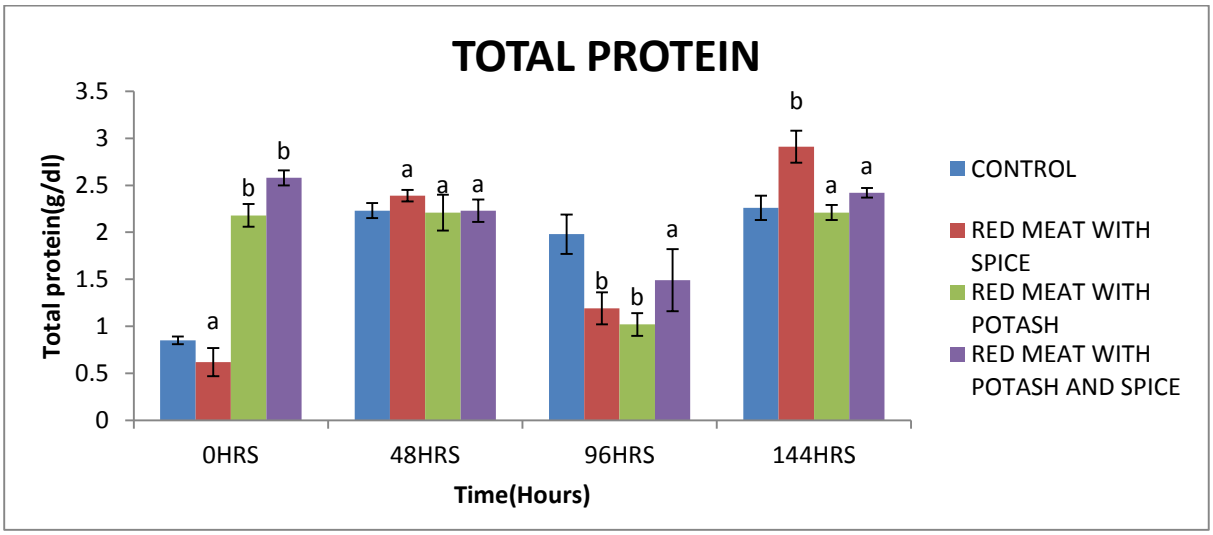

Figure 3.0: Effect of potash and extracts of *Ocimum basilicum* on the total protein content of red meat

The Total protein content results showed that there was significant difference ($p<0.05$) only in red meat boiled with potash and both potash and *Ocimum basilicum* in the first assay carried out. At 48 hours, no significant difference ($p>0.05$) was observed in all the group of samples. At 96 hours, significant difference ($p<0.05$) was observed only in red meat boiled with potash and *Ocimum basilicum* respectively. At 144 hours, significant difference ($p<0.05$) was observed only in red meat boiled with *Ocimum basilicum*. Therefore, the total protein content in red meat boiled with both potash and *Ocimum basilicum* decreased as storage time increased from 0-96 hours. This is in contrast to the work done by Kadhim (2016) where he reported that total protein concentration increased in broilers fed with basal diet containing basil seeds. Fluctuations were observed in the total protein concentration of the other group of samples.

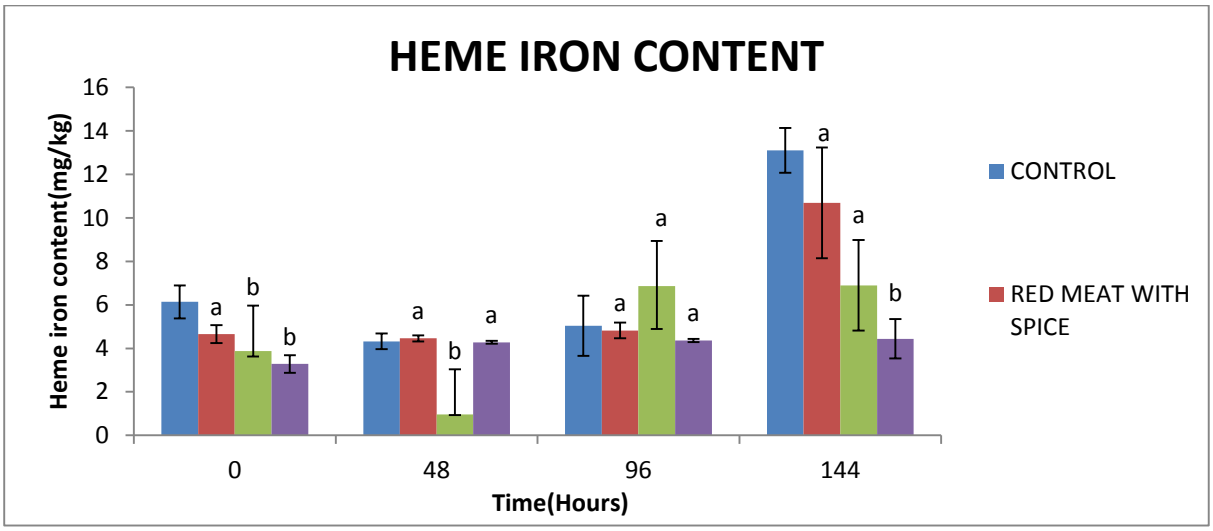

Figure 4.0: Effect of potash and extracts of *Ocimum basilicum* on the heme iron content of red meat.

The result of Heme iron content revealed that there was significant difference ($p<0.05$) in red meat boiled with potash and both potash and *Ocimum basilicum* at 0 hour. At 48 hours, significant difference ($p<0.05$) was observed only in red meat boiled with potash. At 96 hours, no significant difference ($p>0.05$) was observed in all the group of samples while significant difference ($p<0.05$) was observed in only red meat boiled with both potash and *Ocimum basilicum* after 144 hours. No significant difference was observed in red meat boiled with *Ocimum basilicum* from 0 to 144 hours indicating that *Ocimum basilicum* does not have any effect on the heme iron content of the meat. The increase observed in red meat boiled with potash after 96 hours may indicate that the ability of potash to reduce heme iron concentration of meat decreased as storage time increases.

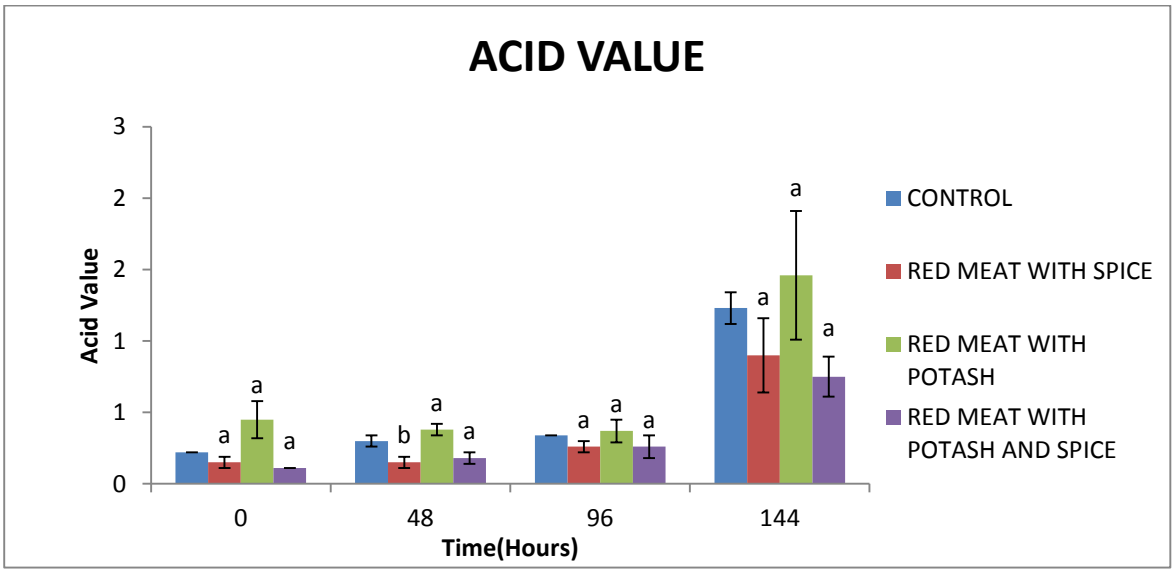

Figure 5.0: Effect of potash and extracts of *Ocimum basilicum* on the acid value of red meat.

Scientia Africana, Vol. 20 (No. 3), December, 2021. Pp 85-94 https://dx.doi.org/10.4314/sa.v20i3.8
© Faculty of Science, University of Port Harcourt, Printed in Nigeria *ISSN 1118 – 1931*

The result for change in acid value showed no significant difference ($p>0.05$) between the control and red meat boiled with potash, *Ocimum basilicum* and both potash and *Ocimum basilicum* in the first assay carried out and throughout the various storage time from 48 hours to 144 hours except for red meat boiled with potash after 48 hours. This indicates that the *Ocimum basilicum* extract does not have any effect on the lipid oxidation of the meat which leads to the development of oxidative rancidity. Similar findings were reported by Chaleshtori *et al*. (2014) in their work done to evaluate the antioxidant and antibacterial activity of (*Ocimum basilicum*) essential oil in beef burger. It is suggestive to state that potash does not have any effect on rancidity of meat.

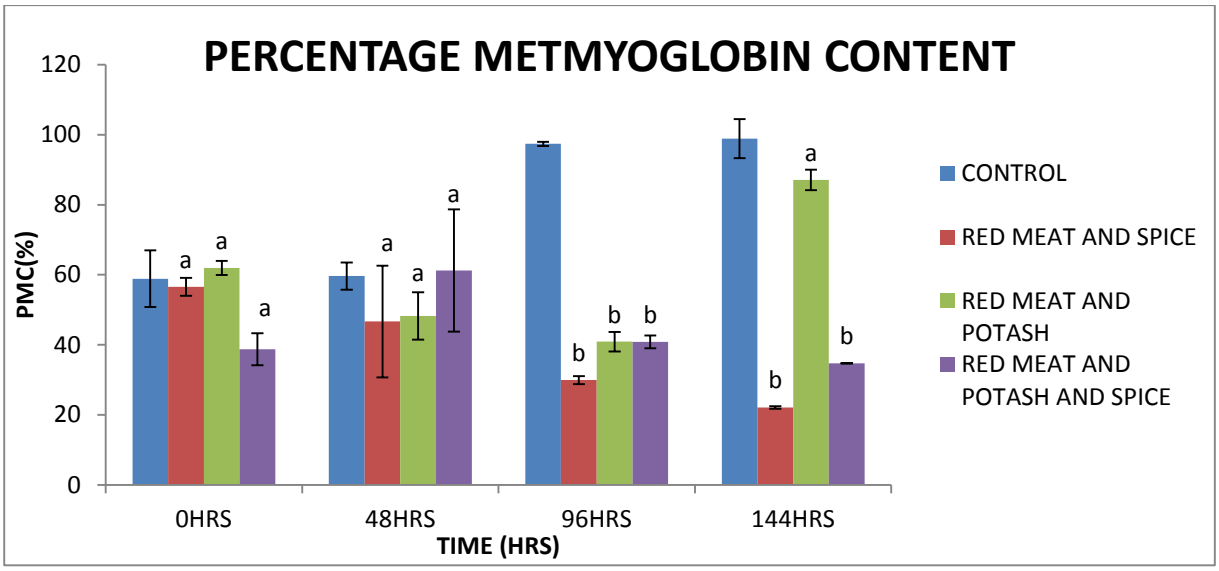

Figure 6.0: Effect of potash and extracts of *Ocimum basilicum* on percentage metmyoglobin content of red meat.

Results for the percentage Metmyoglobin content showed no significant difference ($p>0.05$) between the control and red meat boiled with potash, *Ocimum basilicum* and both potash and *Ocimum basilicum* in the first assay carried out and storage time of 48 hours. At 96 hours, significant difference ($p<0.05$) was noticed in all the group of samples. At 144 hours, significant difference ($p<0.05$) was observed only in red meat boiled with *Ocimum basilicum* and both potash and *Ocimum basilicum*. The typical red colour of fresh meat can change during time to a less desirable brown color. This is caused by the oxidation of myoglobin to metmyoglobin. The decrease observed after 96 and 144 hours in red meat boiled with *Ocimum basilicum* and both potash and *Ocimum basilicum* indicates that *Ocimum basilicum* can prevent the oxidation of myoglobin to metmyoglobin which can help in maintaining the typical red colour of red meat.

CONCLUSION

The present study has demonstrated that potash and *Ocimum basilicum* can affect the quality of meat and reduce its shelf life by increasing the pH value and reducing the total protein content. It also revealed that Ocimum basilicum can prevent the oxidation of myoglobin to metmyoglobin. It also showed that *Ocimum basilicum* and potash have antioxidant properties which

can be justified by the decrease in Malondialdehyde values. Also, potash reduces the heme iron concentration in meat and the ability to do so declines as storage time increases. Potash and *Ocimum basilicum* do not have any effect on the acid value of meat. It was also observed that the presence of *Ocimum basilicum* helped prevent the effect of potash in reducing heme iron content of meat.

REFERENCES

AOACS Cd 3d-63 (1999). AOCS official method, Cd 3d-63: acid value. Champaign, IL(USA): American Chemist's Society

Bankole, J. K., Ngokere, A. A., Ajibade, O. M., Igunbor, C. M. and Eloka, C. C. V. (2015). Degenerating effects of potash (Kaun-K_2CO_3) on the kidney: Unabated continental challenge to human health in Nigeria. *Annals of Biological Research*. **6** (3): 12-18.

Bekhit, A. A., Hopkins, D. L., Geesink, G. and Franks, P. (2014). Exogenous proteases for meat tenderization. *Critical Reviews in Food Science and Nutrition*. **54** (8): 1012-1031.

Bora, K. S., Arora, S. and Shri, J. R. (2011) Role of Ocimum basilicum L. in prevention of ischemia and reperfusion induced cerebral damage, and motor dysfunctions in mice brain. *Journal of Ethnopharmacology*. **137**(3): 1360-1365.

Buege, J.A. and Aust, S.D. (1978). Microsomal lipid peroxidation. *Methods in Enzymology*, **53:** 302-310.

Chaleshtori, R.S., Rokni, N., Rafieian-Kopael, M., Drees, F., Chaleshtori, A. S. and Salehi, E. (2014). Use of Tarragon (Artemisia dracunculus) essential oil as a natural preservative in beef burger. *Italian Journal of Food Science*. **26** :1-5.

Clark, E.M., Mahoney, A.W. & Carpenter, C.E. (1997). Heme and total iron in ready to eat chickens. Journal of Agriculture and Food Chemistry, 45, 124-126

Henry, R.J., Cannon, D.C., Winkelman, J.W. (1974) "Clinical Chemistry, principles and Techniques", Harper & Row, 2nd Ed.

Imafidon, K. E., Egberanmwen, I. D. and Omoregie, I. P. (2016). Toxicological and biochemical investigations in rats administered "kaun" (trona) a natural food additive used in Nigeria. *Journal of Nutrition and Intermediary Metabolism*. **6**: 22-25.

Kadim, S. (2016). Effect of different levels of basil seeds on some blood biochemical traits. *Global Journal of Biotechnology and Biochemistry*. **5**(4): 477-480.

Krzy-Wick, K. (1982). The determination of haem pigments in meat. *Meat Science*, **7:** 29-36.

Lee B.J., Hendricks D.G., Cornforth D.P. (1999): A comparison of carnosine and ascorbic acid on colour and lipid stability in a ground beef patties model system. Meat Science, 51: 245–253

Momoh, T. B., Yaro, C. A., Usman, S. O. and Iyeh, V.A. (2019). Effect of Potash on the Tenderness and Phytochemical Constituernts of Cajanus cajan. *Trends in Applied Sciences Research*. **14**: 278-282.

NAN, (2020). Cooking with paracetamol causes liver, kidney failure, NAFDAC warns. The Guardian, feb., 15.

Pipek, P., Harberl, A. and Jelenikova, J. (2013). Influence of slaughter house handling on the quality of meat

Scientia Africana, Vol. 20 (No. 3), December, 2021. Pp 85-94 *https://dx.doi.org/10.4314/sa.v20i3.8*
© Faculty of Science, University of Port Harcourt, Printed in Nigeria *ISSN 1118 – 1931*

carcasses. *Czech Journal of Animal Science*. **39**: 371-378.

Sakr, S. A. and Amoudi, W.M. (2012). Effect of leave extract of Ocimum basilicum on deltamethrin induced nephrotoxicity and oxidative stress in albino rats. *Journal of Applied Pharmaceutical Science***. 2**(5): 22-27.

Silva, V. A., da Sousa, J. P., de Luna Freire Pessôa, H., Ramos de Freitas, A. F., Melo Coutinho, H. D., Nogueira Alves, L. B. and Lima, E. O. (2016). *Ocimum basilicum*:Antibacterial activity and association study with antibiotics against bacteria of clinical importance. *Pharmaceutical Biology*. **54**(5): 863-7.

Soleimanzadeh, H., Habibi, D., Ardakani, M. R., Paknejad, F. and Rejali, F. (2010). Effect of potassium levels on antioxidant enzymes and Malondialdehyde content under drought stress in sunflower (*Hellanthus annus* L *). American Journal of Agricultural andBiological sciences*. **5**(1): 56-61.

Weglarz, A. (2010). Meat quality defined based on pH and colour depending on cattle category and slaughter season. *Czech Journal of Animal Science*.**55**(12): 548-556.