

Effects of Storage on Some Chemical Indices of Beef Quality

Mark Ehijele Ukhun & Ubu Izi

Chemistry Department, University of Benin, Benin City, Nigeria

(Received 10 October 1989; revised version received and accepted 6 June 1990)

ABSTRACT

The effects on quality of storing beef under conditions that simulate those prevailing in Nigerian traditional markets and, most probably, in traditional markets in most other developing countries, were examined.

Fhe fresh beef samples obtained from the thighs of mature cows were stored for 11 h at $25 \pm 3^{\circ}C$ *, and chemical indices of quality monitored. Chemical indices monitored included total volatile nitrogen (TVN), extract release volume (ER V), free fatty acid (FFA), thiobarbituric acid number (TBA), protein, total fat, insoluble protein, moisture, fatty acid profiles and spectral characteristics. Changes in these indices, generally, indicated loss of quality. Variations in the TBA values of the beef during a 2-week storage period of water activities (a_w) of* 0.11 *,* 0.33 *and* 0.75 *, respectively, suggest that a_w may influence beef lipid quality via oxidation by affecting either the induction period or the overall extent of oxidation.*

INTRODUCTION

Beef constitutes a major source of animal protein for many people of the world, barring constraints on its consumption imposed by cultural or nutritional considerations, such as the need to reduce dietary intakes of saturated fatty acids. In the less developed countries, the major constraints appear to be not just the total supply of beef but also, its cost. The problem is further compounded by the anti-mortem and post-mortem mishandling of the limited quantities of beef.

For example, in the Nigerian traditional open markets, most foodstuffs are usually marketed exposed to the vagaries of weather, without proper control of temperature, bacterial, chemical and enzymic activities. Additionally, variations can occur in the equilibrium relative humidities, and therefore of a_w (Laing & Steinberg, 1981) of foods left exposed this way to the atmosphere. The net effect could be marked changes in the quality and nutritive values of such foods (Martinex & Labuza, 1968; Roubal, 1970; Troller, 1971).

The present studies examine some of the changes in the chemical quality indices of beef stored under conditions that simulate those of Nigerian traditional markets. The results might prove to be relevant to Nigeria and to other less developed countries.

MATERIALS AND METHODS

Fresh beef samples were obtained from the rounds of mature (4-5 years) cows at the New Benin Market-a traditional open market in Benin City, Nigeria, 1 h *post mortem* (storage on open wooden slabs at ambient conditions, temperature about 20° C). The beef cuts included the following muscles: *biceps femoris, semitendinosus, quadriceps, femoris, gastrocnemius* and *trocharter tertius.*

Subsequently, 2 kg of the beef samples were stored for 11 h in aluminium trays sitting on a concrete surface outside the laboratory, exposed to the atmosphere, to simulate the storage conditions and time used in Nigerian traditional open markets. Ambient temperature was 25 ± 3 °C.

For the a_w studies, duplicate 1 kg beef samples were placed inside three separate desiccators where a_w values of 0.11, 0.33 and 0.75, respectively, were established (Rockland, 1960). The samples were stored for 2weeks at $26 \pm 5^{\circ}$ C on a laboratory bench. The three water activities of 0.11, 0.33 and 0.75 are those which approximate that in the three (northern, middle and southern) climatic zones of Nigeria.

In the 11-h storage studies, the TVN, ERV, TBA, FFA, fat, crude protein and moisture contents of the fresh beef and of beef samples exposed for 11 h at 25 ± 3 °C were determined by methods described by Pearson (1976). The water-insoluble protein contents of the beef samples were determined by differences: total protein content minus protein content of 50-ml filtered water extract of the beef. The fatty acid profiles of both the fresh and stored beef lipids were analysed by gas-liquid chromatography (GLC) following extraction of the lipid from the beef samples by the Bligh & Dyer (1959) method. The resulting lipid extracts were methylated (Metcalfe *et al.,* 1966). The methyl esters were subsequently subjected to GLC analyses using a Pye

Unicam series 104 gas chromatograph with the following operating conditions:

Injection, oven and flame ionisation detector temp. $= 180^{\circ}$ C Nitrogen carrier gas flow rate $= 50$ ml/min

Chart speed $= 2$ cm/min

Glass column $(1.7 \text{ m} \times 4 \text{ mm})$ packed with 8% 1,4-butanediol succinate adsorbed on 60/80 mesh Chromosorb W. The accruing peaks were identified by comparison of retention times with those of authentic fatty acid methyl esters. Quantification was by triangulation.

The spectrophotometric studies of the fresh and 11-h stored beef samples were carried out by macerating, in a mortar, a 5 g sample of each in 50 ml of 95% raethanol and benzene, respectively. The holding time in each of the solvents was 30min. Filtration, through a Whatman No. 4 filter paper, followed. The absorbances of the resulting filtrates were measured with an SP 500 spectrophotometer equipped with a recorder. The UV-VIS range of 200-800 nm (Willard *et al.,* 1974) was scanned.

Finally, the TBA values of the beef samples stored at the different a_w values of 0.11, 0.33 and 0-75, respectively, were estimated, every 2 days, for the total 2-week storage period, by the method already indicated.

Parameters	Fresh sample	Stored sample $(11h, 25 + 3^{\circ}C)$	
TVN (mg nitrogen/100 g flesh)	$14.0 + 0.8$	$22.4 + 0.9$	
ERV (ml)	$29.0 + 0.5$	$19.0 + 0.7$	
FFA $\%$ (as oleic acid)	0.02 ± 0.01	$0.05 + 0.02$	
TBA (mg malonaldehyde/			
kg sample)	$0.23 + 0.02$	$0.50 + 0.01$	
Protein content $(\%)$	$20.0 + 0.2$	$21.30 + 0.14$	
Insoluble protein			
content $(\%)$	$17.3 + 0.5$	$18.6 + 0.7$	
Total fat content $(\%)$	$3.07 + 0.05$	$3.01 + 0.06$	
Initial moisture			
content $(\%)$	$75.0 + 1.2$	(not determined)	
Moisture content			
$(a_{\rm w}, 0.11)$ $(\frac{9}{6})$	$75.0 + 1.2$	$13.1 + 0.9$	
Moisture content			
$(a_{\rm w}, 0.33)$ (%)	$75.0 + 1.2$	$63.3 + 1.1$	
Moisture content			
$(a_{\rm w}, 0.75)$ (%)	$75.0 + 1.2$	$65.5 + 0.8$	

TABLE 1 Storage Changes in the Chemical Quality Parameters of Beef

Values are expressed as means \pm SEM.

No. of days	TBA values (mg malonaldehyde/kg sample)			
in storage	$a \sim 0.11$	$a_w 0.33$	$a_w 0.75$	
0	$0.23 + 0.03$	$0.23 + 0.03$	$0.23 + 0.03$	
2	$0.43 + 0.01$	$0.13 + 0.03$	$0.12 + 0.06$	
4	0.31 ± 0.06	$0.33 + 0.05$	0.22 ± 0.03	
6	$0.22 + 0.03$	$0.40 + 0.02$	$0.24 + 0.04$	
8	0.12 ± 0.04	$0.55 + 0.05$	$0.45 + 0.03$	
10	$0.03 + 0.02$	$0.74 + 0.07$	$0.34 + 0.02$	
12	(not detected)	$0.55 + 0.02$	$0.13 + 0.07$	
14				
\triangle TBA $=$ (TBA _{max}) $-TBA$ _{initial})	0.20	0.51	0.22	

TABLE 2 Effect of Water Activity on the TBA Values of Beef during a 2-Week Storage at an Ambient Temperature of $26 \pm 5^{\circ}$ C

Values are expressed as means \pm SEM.

The results presented in Tables 1 and 2 are the means of, at least, six readings from duplicate samples while those in Tables 3 and 4 are the means of two readings from duplicate samples.

RESULTS AND DISCUSSION

In examining Table 1, it can be observed that after an 11-h storage period at ambient conditions, the TVN content of the beef increased from 14 to 22 mg

Sample	Solvent	No. of peaks	λ_{max} (nm)
Fresh beef	95% ethanol	2	400, 315
Fresh beef	Benzene	4	540, 520, 285, 265
Stored beef $(11 h, 25 \pm 3^{\circ}C)$ 95% ethanol Stored beef		$\overline{2}$	415, 275
$(11 h, 25 + 3$ ^o C)	Benzene	3	525, 415, 275

TABLE 4 Changes in the Spectral Characteristics of Fresh Beef Extracts

nitrogen per 100g flesh. This is indicative of protein breakdown by cathepsins (Forrest *et al.,* 1975) and proteolytic enzymes secreted by contaminating microbes.

The ERV of the beef also decreased from 29 to 19 ml; this amounts to about 34% decrease and to a lowering of this quality parameter of beef. The decrease could be due to decreased water-holding capacity (WHC) which is usually a manifestation of post-mortem meat protein denaturation (Hamm, 1975).

The beef had been subjected to some lipolytic action, as is evident by the increase in its FFA content, as shown in Table 1. The lipases, obviously secreted by contaminating microbes, indicate a need for improved sanitary conditions in the marketing of beef in Nigerian traditional markets. However, the practice in Nigeria, of boiling beef for a long time before consumption, means that FFA values indicative of microbial contamination, may not constitute a health hazard, if toxins have not accumulated.

The marked increase in the TBA number of the stored beef observed in Table 1 indicates extensive beef lipid oxidation. Undesirable organoleptic quality changes associated with lipid oxidation in food systems are documented (Drerup *et al.,* 1981; Du Plessiss *et al.,* 1981).

The increase in protein content of the beef, as presented in Table 1, can be ascribed to extraneous sources, such as the contaminating microbes, since net protein synthesis, *post mortem,* would not occur in the stored beef. Postmortem protein denaturation explains the increased protein insolubility shown in Table 1.

No marked changes in the total fat content of the beef samples are discernible in Table 1. This is probably because of the lean nature of the beef cuts and probably because volatile lipid components arising from oxidation and hydrolysis were not in sufficient quantities to affect the total fat content during the ll-h storage period.

By comparing the initial moisture content of the beef with the a_w values 0" 11, 0-33 and 0-75, respectively, a desorption phenomenon in the stored beef is inferred, with the degree of desorption decreasing with increases in $a_{\rm w}$.

Water activity affects the nutritive and organoleptic attributes of foods (Martin, 1958; Moreya & Pelleg, 1981). Thus, in Table 2, it is observed that changes in a_w led to differences in the oxidation status of the beef lipid as assessed by TBA values (Greene & Cumuze, 1982). The results in Table 2 can be viewed from two perspectives: (1) that of the relationship of $\triangle TBA$ values to changes in a_w and (2) from the point of view of the time lag in the attainment of the respective highest TBA values. With respect to (1), it appears that the a_w of 0.33 was the least effective in mitigating lipid oxidation in the stored beef, contradicting previous findings, especially, in dehydrated food systems (Labuza *et al.*, 1972) where very low and high a_w values have been reported to have pro-oxidant effects. However, with respect to the latter perspective, the a_w of 0.33 appears to be exerting an antioxidant effect (that is, the maximum TBA value was attained in 10 days' storage at an a_w 0.33, at 2 days at an a_w 0.11 and at 8 days at an a_w 0.75). The implication is that while the intermediate a_w of 0.33 increased the induction period of lipid oxidation, it was nevertheless not a guarantee against the attainment of high oxidation status. A complex pattern of lipid oxidation may be expected in beef where components such as the proteins, vitamins and carbohydrates may interact to complicate the oxidation (Ukhun, 1987). The absence of any measurable malonaldehyde after 14 days of storage, in all the beef samples maintained at the various a_w values, can be attributed to the lability of the malonaldehyde and in its transformation to oxidation end products, such as short chain fatty acids (Kaitaranta, 1981).

The fatty acid profiles of the fresh and 11-h stored beef depicted in Table 3 are suggestive of lipid oxidation phenomenon in the stored beef and are, therefore, in agreement with the results in Tables 1 and 2, where overall increases in TBA values are observed.

The decrease in the relative amount of the total unsaturated fatty acids *vis-à-vis* that of the saturated ones, after the 11-h storage (Table 3) may be due to the greater rate of oxidation of the unsaturated fatty acids (Ukhun, 1984). The lowered unsaturation of the beef lipids may be nutritionally undesirable since saturated fatty acids are believed to be linked with cardiovascular disorders (Scheig, 1968).

Table 4 depicts the UV-visible spectral characteristics of the solvent extracts of the fresh and stored beef. Because of the nature of the solvents used, chemical components such as lipids, myoglobin, vitamins, muscle glycogen (if any), proteins, peptides and amino acids, would have been extracted, thereby, contributing to the values indicated in the table. For example, between 300 and 500 nm, absorption by unsaturated vitamins and

conjugated compounds, such as the oxidising lipids, could have occurred (Mehlenbacher, 1960), while at 540 nm, a value close to 555 nm, reported to be the λ_{max} (Price & Schweigert, 1971) for myoglobin, the beef myoglobin may have shown considerable absorption. The strong absorption at 520 and 525 nm reported in Table 4 could have been due to metmyoglobin. The λ_{max} for metmyoglobin is about 505nm (Price & Schweigert, 1971). Since metmyoglobin is the oxidation product of myoglobin, it is interesting that while the benzene extract of the stored beef did not show any absorption at 540 nm (myoglobin), it did at 525 nm (metmyoglobin). This appears to be another indication of an oxidation process in the stored beef. Therefore it might be possible to demonstrate some correlation between spectral changes of beef, especially as they relate to myoglobin/metmyoglobin ratio, and changes in TBA, peroxide values and other conventional measurement of lipid oxidation. The data presented in Table 4 suggest that the type of solvent used to extract beef affected the spectral picture of the resulting extracts. This is a reflection of the differing affinities of the beef components for the different types of solvents.

CONCLUSIONS

Storage of beef under conditions that usually prevail in Nigerian traditional markets leads to losses in some chemical indices of beef quality. Control of water activity, exposure time and temperature could help to mitigate these losses.

ACKNOWLEDGEMENT

The authors acknowledge the kindness of Prof. F. I. Opute in allowing the use of his GLC equipment.

REFERENCES

- Bligh, E. G. & Dyer, W. J. (1959). A rapid method of total lipid extraction and purification. *Canadian J. Biochem. Physiol.,* 37, 911-17.
- Drerup, D. L., Judge, M. D. & Aberle, E. D. (1981). Sensory properties and lipid oxidation in pre-rigor processed pork sausage. *J. Food Sci.*, **46**, 1659–61.
- Du Plessiss, L. M., Twisk, P. V., Niekerk, P. J. & Steyn, M. (1981). Evaluation of peanut and cottonseed oils for deep frying. J. *Amer. Oil Chem. Soc.,* 58, 575-8.
- Forrest, J. C., Aberle, E. D., Hedrick, H. B., Judge, M. D. & Merkel, R. A. (1975). *Principles of Meat Science.* W. H. Freeman and Co., San Francisco, pp. 154.
- Greene, B. E. & Cumuze, T. H. (1982). Relationship between TBA numbers and inexperienced panelists' assessment of oxidized flavor in cooked beef. J. *Food Sci.,* 47, 52-8.
- Hamm, R. (1975). Water holding capacity of meat. In *Meat,* eds D. J. A. Cole & R. A. Lawrie. The Butterworth Group, UK, p. 321.
- Kaitaranta, J. K. (1981). TLC-FID assessment of lipid oxidation as applied to fish lipids rich in triglycerides. J. *Amer. Oil Chem. Soc.,* 58, 710-13.
- Labuza, T. P., McNally, L., Gallagher, D., Hawkes, J. & Hurtado, F. (1972). Stability of intermediate moisture foods. I. Lipid oxidation. J. *Food Sci.,* 37, 154-9.
- Lang, K. W. & Steinberg, M. P. (1981). Predicting water activity from 0.3 to 0.95 of a multicomponent food formulation. J. *Food Sci.,* 46, 670-80.
- Martin, H. F. (1958). Factors in the development of oxidative rancidity in ready-toeat crisp oatflakes. J. *Sci. Food Agric.,* 9, 817-24.
- Martinex, F. & Labuza, T. P. (1968). Rate of deterioration of freeze-dried salmon as a function of relative humidity. J. *Food Sci.,* 33, 241-7.
- Mehlenbacher, V. C. (1960). *The Analysis of Fats and Oils~* The Gerrard Press Publ., Illinois, p. 33.
- Metcalfe, L. D., Schmitz, A. A. & Petka, J. R. (1966). Rapid preparation of fatty acid esters for gas chromatographic analysis. *Anal Chem.,* 38, 514-18.
- Moreya, R. & Pelleg, M. (1981). Effect of equilibrium water activity on the bulk properties of selected food powders. *J. Food Sci.,* 46, 1918-22.
- Pearson, D. (1976). *The Chemical Analysis of Foods,* 7th edn. Churchill Livingstone, London, p. 365.
- Price, J. F. & Schweigert, B. S. (1971). *The Science of Meat and Meat Products*, 2nd edn. Freeman, San Francisco, p. 103.
- Rockland, L. B. (1960). Saturated salt solutions for static control of relative humidity between 5 and 40°C. *Anal. Chem.*, 32, 1375–6.
- Roubal, W. T. (1970). Trapped radicals in dry lipid-protein systems undergoing oxidation J. *Amer. Oil Chem. Soc.,* 47, 141-4.
- Scheig, R. (1968). Absorption of dietary fat: Uses of medium chain triglycerides in malabsorption. Amer. J. Clin. Nutr., 21, 300-4.
- Troller, J. A. (1971). Effect of water activity on enterotoxin B production and growth of *Staphylococcus aureus. Appl. Microbiol.,* 21, 435-9.
- Ukhun, M. E. (1984). Fatty acid composition and oxidation of cowpea *(Vigna unguiculata)* flour lipid. *Food Chem.,* 14, 35-43.
- Ukhun, M. E. (1987). Palm oil: Fatty acid profile and effects of selected antioxidants in freeze-dried model systems. *Human Nutrition: Food Sciences and Nutrition,* 41F, 113-19.
- Willard, H. H., Merritt, L. L. & Dean, J. A. (1974). *Instrumental Methods of Analysis,* 5th edn. Van Nostrand Co., NY, p. 42.