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# COMPARATIVE STUDY OF NITROSAMINE IN ROASTED FOOD BASE ON THE ROASTING METHODS

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**ABSTRACT.** The present study examined the interrelationships of roasting methods and nitrosamine compounds (NA) content in eleven food sample investigated. The total level of NA in examined food ranged from  $1.1 \times 10^{-3}$  µg per g in oven roasted white maize to  $5.0 \times 10^{-3}$  µg per g in wood roasted pig meat that is lower than the legal level in WHO (0.001 µg per kg). The significant increase of NA concentration in all food samples on roasting was observed. Methods of roasting have significant effects on the level of nitrosamine in each of the material used.

#### 1. INTRODUCTION

Nitrosamines are a class of chemical compounds with the generic chemical structure  $R_2NN=0$ . They are produced under certain conditions (acidic pH, high temperature, presence of certain reducing agents) in a variety of media (products, biological systems, air, etc) when nitrites react with the so called nitrosatable substances, mainly secondary amines. They are found in tanneries and plants manufacturing pesticides, rubber products and tires as well as fish processing plants, foundries, dye-making plants and research labs[1].

Nitrosamine occur commonly because their chemical precursors-amine and nitrosating agents occur commonly and the chemical reaction for nitrosamine formation is quite facile [2]. The carcinogenic properties of N-nitrosodimethylamine were established and consequently it is important to ensure that the concentration present in food is below a level which is hazardous to man. Nitrosamines are believed to be formed due to the interaction of various nitrosating agents (e.g. nitrite, nitrogen oxide) and amines in the foods [3]. Nitrosamine is found in many food stuffs especially beer [4], fish by products and in meat and cheese products preserved with nitrite picking salts. Their existence has been confirmed in food products, cosmetic product, tobacco smoke [5] soil, ground water [6]. Most N- nitrosamines like N-methylaniline and dimethylaniline have been shown to be carcinogenic in laboratory animals [7]. The N-nitroso compounds are known to be acutely toxic, mutagenic, teratogenic and carcinogenic. No animal species is known to be resistant to the carcinogenicity effect of nitrosamines and other N-nitroso compounds [8]. Naturally N-nitrosamines could be found as a result of biological, chemical or photochemical processes [9]. This study is therefore aimed at comparing the effects of roasting methods on the level of nitrosamines in some roasted food materials.

# 2. MATERIALS AND METHODS

Samples food materials fish(*Tilapia*), cashew nut(*Anacardium occidentale*), white yam(*Discorea rotunda*), plantain(*Musa sapientum*), groundnut(*Arachis hypogaea*), cocoyam(*Colocasia esculenta*), bush meat, beef meat, pig meat, coconut(*Cocos nucifera*), white maize(*Zea may*), yellow maize(*Zea may*) were purchased from Ado-Ekiti in Nigeria.

# **Methods of Roasting**

Coal Method: Each sample was roasted by using coal smoking method. Direct smoking method was used at 160°C, and the moisture level was controlled. Each roasted sample materials were

homogenized well with blender and stored in a refrigerator at 4<sup>o</sup>C. The analysis was done within a limited time.

**Oven Method:** Each sample was roasted by using oven roasting method. Samples were placed in the oven at 160°C, and the moisture level was controlled. Each roasted sample materials were homogenized well with blender and stored in a refrigerator at 4°C. The analysis was done within a limited time.

**Wood Smoking Method:** Each sample was roasted by using wood smoking method. Samples were placed directly on wire gauze on a burning wood at  $160^{\circ}$ C, and the moisture level was controlled. Each roasted sample materials were homogenized well with blender and stored in a refrigerator at  $4^{\circ}$ C. The analysis was done within a limited time.

**Standard calibration curve:** For the standard calibration graph 1, 2, 3, 4 and 5cm<sup>3</sup> sodium nitrate working solution (corresponding to 0.2, 0.4, 0.6 and 1.0µg NaNO<sub>3</sub>) were added to 50cm<sup>3</sup> volumetric flasks. To each flask was added 1cm<sup>3</sup> N-1 naphthyl reagents. Each flask was made up to 50cm<sup>3</sup> with distilled water. After standing for 25 min, absorbance at 540nm was measured. Absorbance at 540nm was plotted against concentration in µgcm<sup>3</sup> to obtain the calibration graph.

**Determination of nitrosamine:** Ammonium sulphamate was added to 10g of the roasted food samples to stabilizer any N-nitrosamine and also as a free nitrite scavenger. An aqueous sodium chloride solution was then added to liberate the nitrosamine from the nitrosamine-water emulsion. The aqueous mixture was quantitatively transferred to a separating funnel where it was extracted with pentane to remove any non polar components. The aqueous phase was extracted with ethyl acetate and the organic phase was washed with water and then dried with (Na<sub>2</sub>SO<sub>4</sub>). The solvent was concentrated in vacuo using a rotary evaporator. The residue was dissolved in glacial acetic acid and an aliquot of denitrosation reagent (3% v/v) HBr in glacial acetic acid) was added. Sulphanilamide was mixed with the test aliquot and the N – naphythyl reagent was added. The absorbance of the test sample was measured at 540nm using spectrophotometer 20 [10]

#### **Data Evaluation**

The total N-nitroso content express as

$$= \underbrace{\mu g.cm^{-3} \text{ NaNO}_3 \text{ from standard graph}}_{\text{Mass of sample}} \times \underbrace{1}_{0.5} \times \underbrace{\frac{30}{69}}_{\text{69}} \times \underbrace{\frac{100}{22.4}}_{\text{100}} \mu g/g$$

Where the portion of the equation between the vertical lines is the conversion factor from NaNO<sub>3</sub> to nitrosamine.

#### 3. STATISTICAL ANALYSIS

The analytical data are reported as mean  $\pm$  standard deviation of triplicate independent measurements and were subjected to ANOVA, the significance of mean differences was determined by Duncans posthoc test and t-test using SPSS version 14.0.

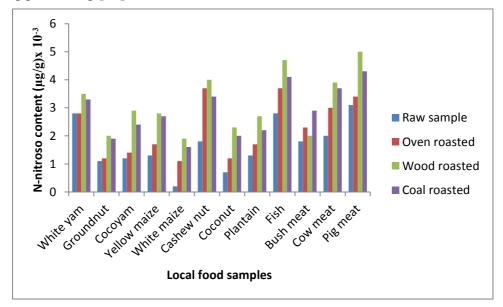
# 4. RESULTS AND DISCUSSION

**Table 1.** Total N-nitroso content(μg/g) in raw, oven roasted, wood roasted and coal roasted food material.

N-nitroso content $(\mu g/g) \times 10^{-3}$						
Sample	Raw sample	Oven roasted	Wood roasted	Coal roasted		
White yam	2.8±0.03	2.8±0.01	3.5±0.02	3.3±0.01		
Groundnut	1.1±0.01	1.2±0.01	2.0±0.01	1.9±0.03		
Cocoyam	1.2±0.03	1.4±0.01	2.9±0.01	2.4±0.01		
Yellow maize	1.3±0.01	1.7±0.02	2.8±0.02	2.7±0.01		
White maize	0.2±0.01	1.1±0.01	1.9±0.01	1.6±0.02		
Cashew nut	1.8±0.01	3.7±0.03	4.0±0.02	3.4±0.01		

Coconut	$0.7\pm0.03$	$1.2\pm0.01$	$2.3\pm0.02$	$2.0\pm0.01$
Plantain	1.3±0.01	1.7±0.01	2.7±0.01	2.2±0.02
Fish	2.8±0.01	3.7±0.03	4.7±0.03	4.1±0.01
Bush meat	1.8±0.03	2.3±0.01	2.0±0.01	2.9±0.01
Cow meat	2.0±0.01	3.0±0.01	3.9±0.02	3.7±0.02
Pig meat	3.1±0.02	3.4±0.02	5.0±0.01	4.3±0.01

Table 1 shows the nitrosamine levels in the raw and roasted food materials. The nitrosamine levels varied from  $1.1 \times 10^{-3} \ (\mu g/g)$  in white maize to  $3.7 \times 10^{-3} \ (\mu g/g)$  in fish in oven roasted food sample which was higher than the nitrosamine level in raw food samples. Nitrosamine levels in wood roasted food samples varied from  $1.9 \times 10^{-3} \ (\mu g/g)$  in white maize to  $5.0 \times 10^{-3} \ (\mu g/g)$  in pig meat, while nitrosamine levels in coal roasted food sample varied from  $1.6 \times 10^{-3} \ (\mu g/g)$  in white maize to  $4.3 \times 10^{-3} \ (\mu g/g)$  in pig meat. These values were here than the level in raw food samples. These were at the American Food and Drug Administration (FDA) action level. Higher levels may result from microbial reduction of nitrate stored under in appropriate conditions [11]. While roasting process increased the nitrosamine levels in all the samples as shown in table I. Similar changes in levels of nitrosamine in food after cooking were observed for vegetables by Ezeagu and Fafunso [12]. The apparent difference in the nitrosamine contents between the raw and roasted food sample might be due to reconstitution and chemical interactions between the various component effect by heating or boiling during processing [13].



**Fig.1.** The variation in the level of nitrosamine in roasted food sample with respect to roasting methods.

The results suggested that the use of wood, coal and oven roasting method may cause some small increase in the natural nitrosamine level. The size of this increase is markedly affected by the roasting procedure viz: the increase when fish is roasted in oven or coal or wood is greater than raw; similarly placing every other food samples, the level in wood roasting nitrosamine levels is greater than coal, and oven. The differences between the levels in food samples of different species on different occasion were at least great as these attributed to roasting. Earlier work of Kawabata et al [14] reported that coal of Japanese dried fish results in significantly higher levels of nitrate, nitrite and nitrosamine (up to 300ppm). Various conditions can affect nitrosamines formation, for example spices such as pepper can be a source of formation.

### 5. Conclusion

The study revealed that a significant relationship exist between nitrosamine value of coal, wood and oven roasting methods. The gases produced during wood roasting contributed to high

value of nitrosamine wood roasted food materials than the coal and oven roasted. Based on the findings the use of direct roasting (smoking) using wood, coal of food materials should be discouraged and that processing of the food materials be modified to reduce the presence of toxicants.

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