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RESEARCH PAPER

THE COMBINED INCIDENCE OF GRADE II AND GRADE IV ASTROCYTOMA IN THE BRAIN OF RATS FED WITH DIET CONTAINING YAJI: A COMPLEX NIGERIAN SUYA MEAT SAUCE

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ABSTRACT

Available scientific evidence has shown that some of the active principles of *Yaji* -capsaicin, piperine and monosodium glutamate, have excitotoxic, apoptotic or tumourigenic potentials. The focus of this histological study however, is to determine the tumourigenic potentials of *Yaji* in the brain of rats. Eight weeks old white albino rats were used. They were divided into eight groups (A – H), each of which has three subgroups (n=5). The subgroups represent experimental durations of 2 weeks, 4 weeks, and 6 weeks respectively. Group A served as control while groups B – H served as the test groups. The control received only normal feed daily, while the test groups received normal feed plus graded levels of *Yaji* daily. Microscopic examination of the stained brain tissue sections showed emerging histological features similar to those described for grade II astrocytoma in test group H3 (6 weeks; 70%), and grade IV astrocytoma in test groups D3 (6 weeks; 30%). These observed incidence of astrocytoma appeared to be high-dosage/duration dependent, indicating therefore, that the call for the regulation of *Yaji*-production and consumption, is now more pertinent.

Keywords: Suya, Yaji, Additives, Spices, Astrocytoma

INTRODUCTION

Yaji is a complex mixture of groundnut cake powder, additives, spices and salt (Okonkwo, 1987). It is a sauce for the meat delicacy called *Suya* (Nwaopara *et al*., 2004, 2007a, 2007b, 2008a, 2008b, 2009; Omojola, 2008; Uzeh *et al*. 2006). Historically, the sauce is named after a 14th century Hausa ruler called "Yaji (meaning the 'hot one')" (Betumiblog, 2006). According to Igene and Mohammed (1983), "*Suya* is a popular, traditionally processed, ready to eat meat product, which may be served or sold along streets, in club houses, at picnics, parties, restaurants and within institutions". Omojola *et al*. (2008), describes *Suya* as "one of such intermediate moisture products that is easy to prepare and highly relished". On their part, Uzeh *et al*. (2006) described *Suya*, as "a mass consumer fast food whose preparation and sales along the streets, are not done under strict hygienic conditions".

The spices in *Yaji* include ginger, clove, red pepper and black pepper (Nwaopara *et al*., 2004) and they contain gingerol (Witchtl, 2004), eugenol (Krishnaswamy and Raghuramulu, 1998), capsaicin (Collier *et al*., 1965), and piperine (McGee, 2004) as active principle respectively. The main additive in *Yaji* is known as 'white maggi' (or Ajinomoto) and it contains monosodium glutamate (MSG) (Omojola, 2008). The other two constituents –salt and groundnut, contain sodium chloride (Carson *et al*. 1998) and fat (Fageria *et al*. 1997) as active principle respectively.

Of interest however, is the growing concern about the indiscriminate production and consumption of *Suya* and *Yaji*. This has become the subject of several scientific investigations (Nwaopara *et al*. 2004; 2007a; 2007b; 2008a; 2008b; 2009) and some of the findings have shown that an excessive consumption of *Yaji* is capable of inducing pancreatic, liver and kidney damage (Nwaopara *et al*. 2004; 2007b; 2008a). These findings indicate that the unregulated nature of *Yaji* production and consumption portends serious danger to the health of consumers. In fact, the potential health hazards of *Yaji* have been highlighted (Nwaopara *et al*., 2007a).

Furthermore, there are reports that some the active principles in *Yaji*, like capsaicin, piperine and monosodium glutamate have excitotoxic, apoptotic or tumourigenic potentials (Choi, 1988; Blaylock, 1997; Lipton and Rosenberg, 1994; Whetsell and Shapira, 1993; Olney, 1989; Olney, *et al*., 1997; Sugimoto *et al*., 1998; Ankarcrona *et al.* 1998; Martin *et al.* 2000; Bellamy, 2008). The focus of this histological study therefore, is to determine the tumourigenic potentials of *Yaji* in the brain of rats.

MATERIALS AND METHODS

The Substance of Study: The constituents of *Yaji* used for this study, were purchased from Aduwawa Cattle market, Benin City, Edo State, Nigeria, and subsequently mixed together in powdery forms. The mixing process was done as directed by the dealers of *Yaji* at the market. Unlike the dealers, the constituents to be mixed were measured to determine their respective quantities in a given measure of *Yaji*.

The weighing balance used for the measurements, was manufactured by Denver Company USA (Model 200398.1REV.CXP-3000). The measured quantities include Ajinomoto (150g), black pepper (30g), clove (39g), ginger (78g), groundnut cake powder (230g), red pepper (22g), and salt (100g). The total weight of these constituents was calculated and summed up to 649g.

Pellets were produced by mixing appropriate quantities of *Yaji* and feed with little quantity of water. The resultant paste was split into bits and allowed to dry under the sun.

The Subjects/ Substance Administration: Eight weeks old white albino rats of an average weight of 170g were used for this study. They were divided into eight groups (A – H) of three subgroups (n=5) each. The three subgroups represent experimental durations of 2 weeks, 4 weeks, and 6weeks respectively. The subgroups of A (A1, A2 and A3) served as the control, while the subgroups of B – H (B1 – H1; B2 - H2; and B3 – H3) served as the test groups. Group A rats were fed with normal feed (growers mash) only. The feed was purchased from Bendel Feeds and Flour Mills (BFFM), Ewu, Edo State, Nigeria. Test groups B - H rats were fed with growers mash from the same source plus graded quantities of *Yaji* (B, 10%; C, 20%; D, 30%; E, 40%; F, 50%; G, 60%; H, 70%) daily as follows:

1. For two weeks, subgroups 1 (B1, C1, D1, E1, F1, G1 and H1) were fed with feed plus graded levels of *Yaji* (10%, 20%, 30%, 40%, 50%, 60%, and 70%) on daily basis respectively.

2. For four weeks, subgroups 2 (B2, C2, D2, E2, F2, G2 and H2) were fed with feed plus graded levels of *Yaji* (10%, 20%, 30%, 40%, 50%, 60% and 70%) on daily basis respectively.

3. For six weeks, subgroups 3 (B3, C3, D3, E3, F3, G3 and H3) were fed with feed plus graded levels of *Yaji* (10%, 20%, 30%, 40%, 50%, 60% and 70%) on daily basis respectively.

The total daily feeding allowance for each experimental group was 30g while the feeding allowance per rat was 6g. Test groups B (10%) received 3g of *Yaji* daily (0.6g per rat), C (20%) received 6g of *Yaji* daily (1.2g per rat), D (30%) received 9g of *Yaji* daily (1.8g per rat), E (40%) received 12g of *Yaji* daily (2.4g per rat), F (50%) received 15g of *Yaji* daily (3g per rat), G (60%) received 18g of *Yaji* daily (3.6g per rat), and H (70%) received 21g of *Yaji* daily (4.2g per rat).

Tissue Processing: The animals in subgroups 1, 2 and 3 were sacrificed after two weeks, four weeks and six weeks respectively and the tissues obtained, were immediately fixed in formaldehyde to prevent autolysis and putrefaction. The tissue sections were produced by standard routine histological procedures (fixation, dehydration, impregnation, embedding, sectioning and staining with Haematoxylin and Eosin) as described by David (2004). The micrographs of the relevant slides were subsequently taken with the aid of a light microscope at magnification x40.

RESULTS

The result of this study showed several degenerative changes amongst which included the presence of vacuolations, eosinophilic cells, pyknotic nuclei, and gliosis. Of particular relevance are the observed histological features that were similar to those described for:

1. Grade II astrocytoma (as described by Stevens *et al*., 2007) in the micrographs obtained from stained brain tissue sections of rats within test group H3 (6 weeks; 70%) in a manner that appears to be high dosage/duration dependent (See plates 1A and B).
2. Grade IV astrocytoma (as described by Stevens *et al*., 2007) in the micrographs obtained from stained brain tissue sections of rats within test group D3 (6 weeks; 30%) in a manner that appears to be high dosage/duration dependent (See plates 2A and B).

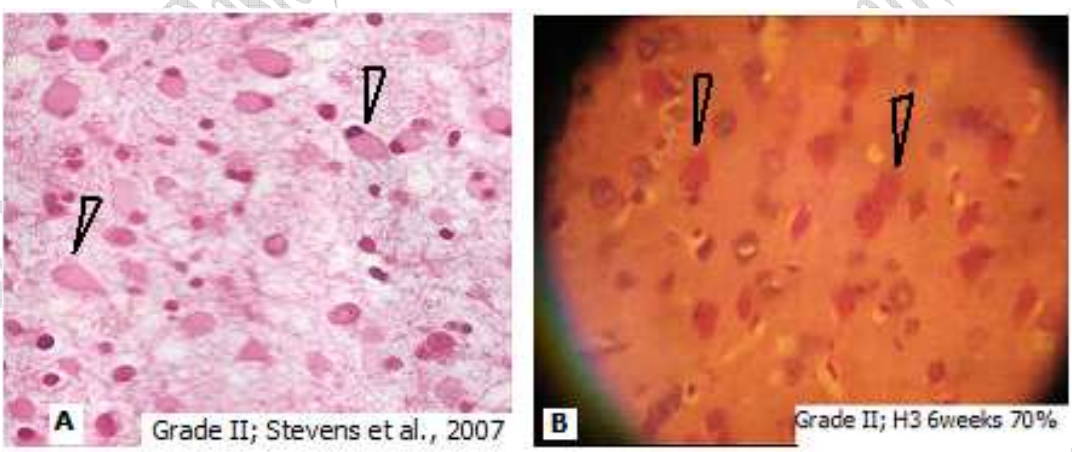

Plate 1 (A and B)**: (**Brain Section; H&E x40**)** showing distortions in cellular architecture of the brain of rats in test group **H3** (6 weeks; 70%) with emerging signs of astrocytoma (**Grade II**). Note the numerous deep 'pinkish' bodies in plate A and compare with plate B adapted from Stevens *et al*., 2007.

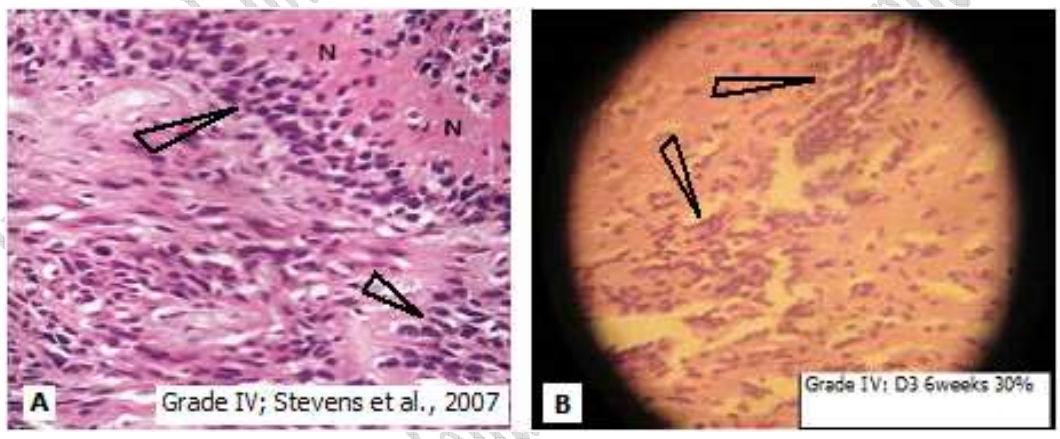

Plate 2 (A and B)**: (**Brain Section; H&E x40**)** showing distortions in cellular architecture in the brain of rats within test group **D3** (6 weeks; 30%) with emerging signs of astrocytoma (**Grade IV**). Note the proliferated cells in plate B and compare with plate A adapted from Stevens *et al*., 2007.

DISCUSSION

The findings of this study fit in perfectly with the histological description for grade II and IV astrocytoma (tumour types of astroglial origin). According to Stevens *et al*. (2007), grade II astrocytoma is characterised by cells with

pink staining cytoplasm and cellular processes that are typical of astrocytic cells, while grade IV astroglioma (also known as *glioblastoma multiforme)* is composed of pleomorphic glial cells of varying sizes and are associated with necrosis, high cellularity and proliferation of endothelial cells in blood vessels (Stevens *et al*., 2007). Also, the high dose/duration dependent manner by which the incidence of astrocytoma was observed implies that *Yaji* at sustained high-dose consumption has the capacity to unleash its tumourigenic potentials. This incidence cannot be unconnected with the high-dose consumption of the constituents in *Yaji*, which, on individual basis, are potentially harmful when consumed in excess (Southgate, 1993).

Of cause, the role of diet types in the induction of cancer has been established and existing scientific evidence show that less than one percent of cancer deaths in industrialized nations are attributable to food additives and industrial products (Trichopoulos and Li, 1996). Dietary factors have also been estimated to account for about one third of cancer deaths in the United States (American Cancer Society 2000; Ames *et al*. 1995; Doll and Peto 1981; Ries *et al*. 2000). Attention has also been drawn to the fact that an excessive consumption of alcoholic beverages is associated with cancers of the breast, oral cavity (primarily in smokers), and liver (International Agency for Research on Cancer 1988; Willett 2001).

Specifically, the influence of MSG cannot be ruled out as there are reports that it is a potential cause of brain tumour (Bellamy, 2008; Rothstein and Brem, 2001). Red pepper is also implicated by the report that capsaicin has tumourigenic and mutagenic potentials (Azizan and Blevins, 1995). The excitotoxic elements in *Yaji* such as MSG, capsaicin and piperine are implicated as well, by the report that excitotoxic destruction facilitates brain tumour growth (Rothstein and Brem, 2001). Of greatest concern is the report that roasted and charcoaled grilled meat delicacy like '*Suya*' (served with *Yaji*), has inherent carcinogenic potentials (Ferguson, 1999).

According to Kleihues *et al*. (2002), the malignancy of gliomas (tumours of glial origin) is assessed by histological grades (II – IV) depending on the presence of cellular pleomorphism, mitosis, vascular proliferation and necrosis. Of greater concern, is the fact that local treatments of tumours of all the grades fail because gliomas are highly invasive (Giese et al, 2003) and surgery fails since the tumour recurs inevitably at the infiltrated margin, no matter how much margin has been resected (Chicoine and Silbergeld, 1995; Silbergeld and Chicoine, 1997). It is our opinion therefore, that *Yaji* has tumourigenic potentials and as such, excessive consumption should be avoided and the need to regulate the production and consumption of *Yaji* cannot be overemphasized.

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AUTHOR(S) CONTRIBUTION

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