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Genotoxic potentials of some medicinal plants on *Drosophila melanogaster*

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ABSTRACT

Genotoxic potentials of three commonly used medicinal plants (*Cola nitida*, *Ocimum gratissimum* and *Monodora myristica*) were assessed on *D. melanogaster*. Eight generations of *D. melanogaster* were cultured on over-ripe banana paste with inclusion levels of 0, 2, 4, 6, 8 and 10% (w/w) of the medicinal plants. Mutation rates both on the wing structure and body colouration were dose-dependent and generation-specific; as increase in the dose of treatment, increases the mutation rates as well as the higher the generations. The degree of effects on these structures was also plant-specific, with followed the trend; *C. nitida* > *O. gratissimum* > *M. myristica*. The result revealed that the medicinal plants have genotoxic potentials with *C. nitida* having the greatest effects; hence they should be consumed with caution.

Introduction

Some medicinal plants have been studied for their anti-mutagenic activities because of their secondary metabolites. [1, 10, 17] Anti-genotoxic agents have also been identified in fresh fruits, vegetables, coffee and tea. [1, 6] Despite the views of these reports, there are other reports pointing to the fact that some of these medicinal plants have toxic and mutagenic properties because of the phytochemical constituents inherent in them. Induced mutation is usually caused by radiation, chemicals, including phytochemicals, errors that occur during meiosis or DNA replication or organism's cellular processes in case of hyper-mutation. [2, 3, 4]

D. melanogaster has relatively simple anatomy and genetically similar to mammals, *hence its use* as a good model to assess the effects of mutations, gene transmission, evolutionary changes in population and chromosomal aberration. [14, 16] It is noteworthy to report that *D. melanogaster* has also been used as a genetic model for several human diseases and to study mechanisms underlying aging and oxidative stress, immunity, diabetes and cancer as well as drug abuse. [26]

Kola nut (*Cola nitida*), Scent leaf (*Ocimum gratissimum*) and African nutmeg (*Monodora myristica*) contain various bioactive compounds, which though therapeutic could be mutagenic or anti-mutagenic. For instance, kola nut contains xanthine stimulants like caffeine, theobromine and theophylline. [21] It is a central nervous system stimulant and has been shown to mediate some physiological effects that are similar to the actions of refined caffeine. [6, 7] It stimulates the cardiac muscle, increase urinary output and relaxation of smooth muscles. [29] On the other hand, scent leaf (*O. gratissimum*) has been reported to possess pharmacological properties such as anti-oxidant, [22] chemotherapeutic, [9] anti-diarrhoeal, [23] anti-contraceptive, [25] insecticidal [11] and anti-helminthes. [13, 24] *M. myristica* contains myristicin among other bioactive compounds. It has been reported to possess anti-diarroheal,[12] anti-microbial activities,[5] reducing effect of cardiac contractions in hypertensive animal. [20]

This study is therefore focused on assessing the genotoxic potentials of these commonly used medicinal plants on *D. melanogaster*.

Materials And Methods

Preparation of culture media: Fresh kola nuts, leaves of scent leaf and African nutmeg were obtained from the Watt market, Calabar, Nigeria and authenticated by Dr. Samuel Udoh, a plant taxonomist. They were washed, finely chopped; oven dried at 37-38ºC and pulverized using electric blender. The powdered samples were mixed with mashed over-ripe bananas at 2, 4, 6, 8 and 10% (w/w), respectively to give the culture media while the 0% inclusion served as the control (Table 1).

Table 1: Protocol for preparation of culture media

Inclusion (%)	Weight of plant sample (g)	Weight of banana paste (g)	Weight of medium (g)
0	0.0	20.0	20.0
2	0.4	19.6	20.0
4	0.8	19.2	20.0
6	1.2	18.8	20.0
8	1.6	18.4	20.0
10	2.0	18.0	20.0

Experimental procedure: *D. melanogaster* (fruit fly) were trapped in clean, labeled large glass jars containing the culture media of the individual plant samples. As the flies came to feed on the culture media, the jars were covered with fine net materials and fasten with tight rubber bands to prevent their escape. The trapped flies constitute the parental generation (P) and were allowed to feed, interbreed and lay eggs.

Eight to nine days after stocking, the eggs hatch to give rise to the next generation. These emerging flies were transferred to fresh culture media according to the different concentration of plant samples. The transferred flies constitute the first mutant generation (M_1) for the study. This procedure was repeated to obtain the successive mutant generation, M_2 to M_7. At every generation, adult flies were removed from the jars after 4 days,

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anaesthetized with dimethyl ether and examine under light microscope.

Data collection and analysis: Morphological aberrations were observed on the wing structure and body colouration while the rates of mutation on the wing structure, body colouration and total mutation were computed and subjected to analysis of variance (ANOVA) using Predictive Analytics Software (PASW), version 18.0.

Results

Various morphological aberrations were observed on the wing structures (Type I aberrations) and on body colouration (Type II aberrations) of *D. melanogaster* after treating with the three medicinal plants. However, the degree of effects on these structures was plant-specific. Generally, the common type I aberrations observed were curved, apterous, curly and miniature wings while type II aberrations were only ebony and yellow body colours.

Mutation rates both on the wing structure and body colouration were dose-dependent and generation-specific as increase in the dose of treatment, increases the mutation rates as well as the higher the generations, the higher the rate of mutation, the medicinal plant notwithstanding (Figs. 1-6). The results revealed that the degree of effects on these structures followed the trend; *Cola nitida* > *O. gratissimum* > *M. myristica*.

Figure 7 shows the percentages of the various aberrations on the wing structure of *D. melanogaster* after exposing them to the three herbal plants. It was observed that *K. nitida* induced more of the curly abnormality (12.25%) of the wing while *O. gratissimum* caused more the miniature wings (3.18%). When exposed to *M. myristica*, apterous wings were observed more (0.93%).

Disscussion

The pharmacological and therapeutic efficacy of any medicinal plant is hinged on the bioactive constituents inherent in it. Undoubtedly, these pharmacological/therapeutic effects have been harnessed positively for the production of drugs or directly in ailment treatment. This notwithstanding, it has been reported that if these plants are not taken with caution they might elicit adverse effects on the same system that they were meant to heal. [18, 19]

Drosophila melanogaster being a model animal to ascertain mutagenicity of substances such as medicinal plants, the results show that the three medicinal plants caused varying degree of effects on the structure of wing and body colouration with *C. nitida* having more mutagenic effect when compared with *O. gratissimum* and *M. myristica*. According to Odebunmi *et al.* [21] *C. nitida* contains caffeine, theobromine and theophylline, which mediate some physiological effects in the system of the recipient organism. [6, 7] These methylxanthines might possess both positive and negative health effects. [15, 27] It is important to understand that though medicinal plants may produce the same bioactive compounds, the likelihood for these bioactive compounds having the same variants may be doubtful. This may also has contributed to the varying degree of effects reported in the present study. Nevertheless, both types of aberrations observed in the fruit flies may have been caused by the singly or synergistic interaction of bioactive compounds in the plants.

Though conflicts in opinion exist concerning the genotoxic and mutagenic effects of caffeine in biological system, [28] it is opined that it interacts chemically with rapidly replicating DNA in growing cells. It is possible that the observed results could be explained by this interaction. Expectedly, the observed increase in the mutation rate both as per concentration and generation might be partly due to the bioactive components of the plants that may have been organ-specific and partly as a result of the mutant flies in M_1 generation reproducing and transferring mutant genes to the next generation, the sterility phenomenon of mutant flies notwithstanding. It is also very likely that these extracts may have distorted the chromosome arm bearing the loci for these genes, which may have been transferred from generation to generation

Though it had submitted that *D. melanogaster* has simple anatomy and remarkable molecular similarity with mammals, implying that any substance that affects them should also affect mammals. This assumption might not be totally correct.

Conclusion

Implicitly, though the results revealed that medicine plants could be mutagenic. It thus suggest that out of the *C. nitida*, *O. gratissimum* and *M. myristica*, *C. nitida* should be consumed with caution while not under-estimating the genotoxic potentials of others.

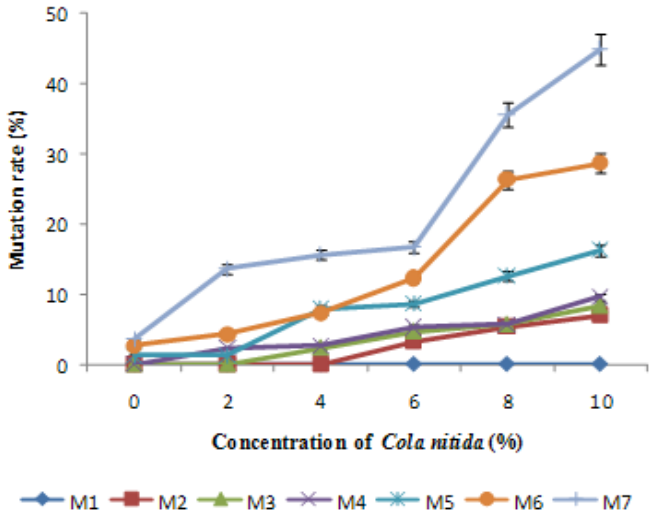

Fig. 1: Mutation rate in wing structure of *D. melanogaster* after treating with *C. nitida*

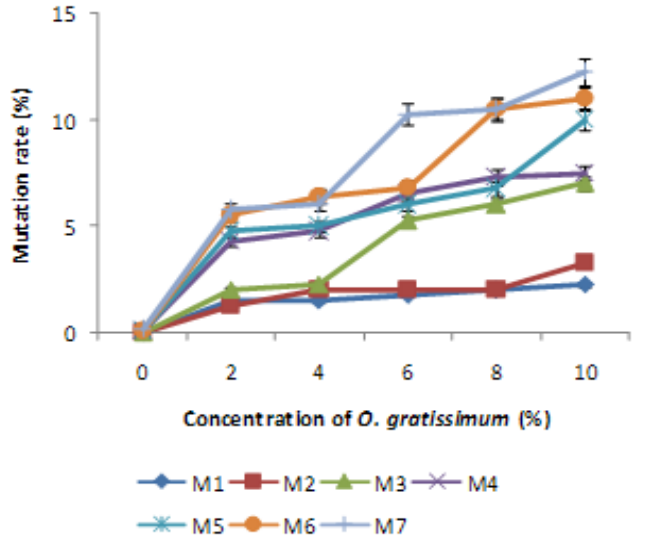

Fig. 2: Mutation rate in wing structure of *D. melanogaster* after treating with *O. gratissimum*

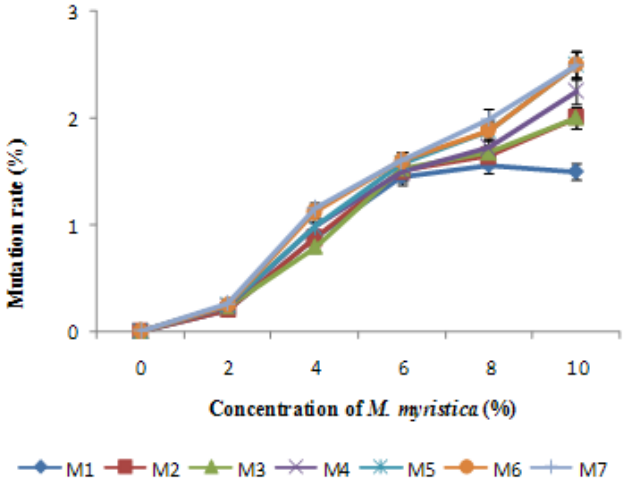

Fig. 3: Mutation rate in wing structure of *D. melanogaster* after treating with *M. myristica*

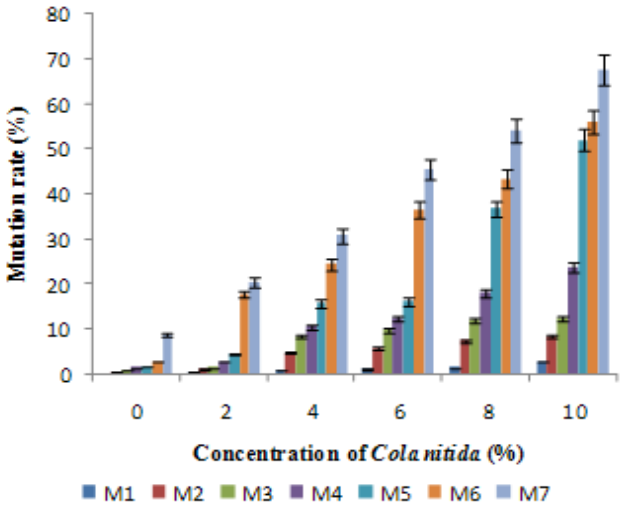

Fig. 4: Mutation rate in body colouration of *D. melanogaster* after treating with *C. nitida*

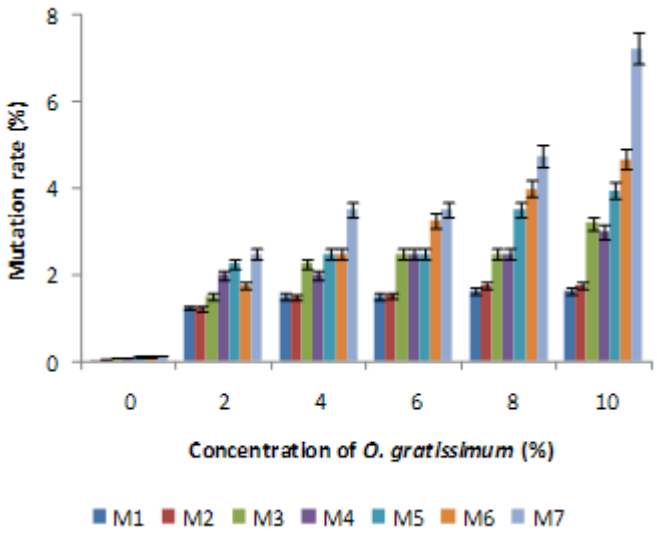

Fig. 5: Mutation rate in body colouration of *D. melanogaster* after treating with *O. gratissimum*

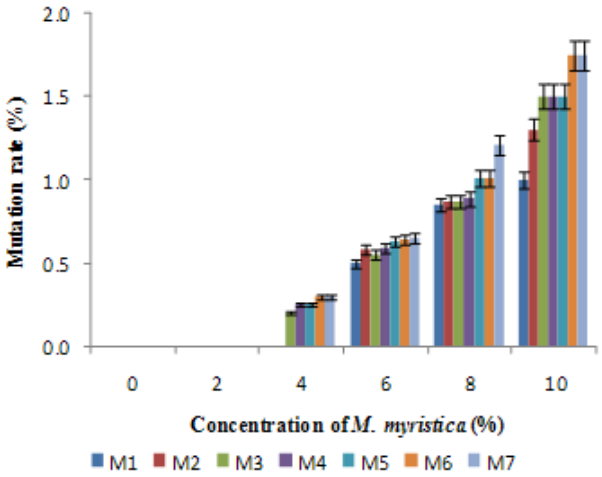

Fig. 6: Mutation rate in body colouration of *D. melanogaster* after treating with *M. myristica*

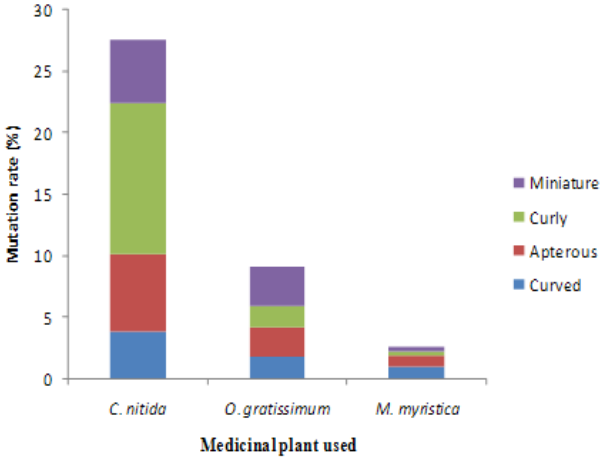

Fig. 7: Effect of medicinal plants on the mutation rate of type 1 aberration

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