



A PRELIMINARY STUDY ON THE ANTIPROLIFERATIVE AND CYTOSTATIC EFFECT OF NIGERIAN HONEY

ENEKIDE CHINEDU^{1*}, DAVID AROME¹, DABUM LUKA JACOB²

¹Department of Science Laboratory Technology (Physiology & Pharmacology Technology), University of Jos, Jos, Nigeria,

²Department of Human Physiology, Faculty of Medical Sciences, University of Jos, Jos, Nigeria. Email: chinex.snow@gmail.com

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ABSTRACT

Background: Cancer is a significant health problem and also a major cause of death in both developing and developed countries all over the world. The World Health Organization (WHO) has ranked it among the leading causes of death, as about 7.4 million (13%) deaths in 2004 were as a result of cancer. Chemoprevention simply refers to the use of pharmacological or dietary agents to prevent induction, inhibit or even delay the advancement of cancer. These agents should have antiproliferative or even cytostatic effect to achieve this aim. There have been recent claims of various naturally occurring products having anticancer effect, with honey being one of these products. **Aim:** The aim of this study is to evaluate if Nigerian honey has antiproliferative and cytostatic potentials. **Materials and Method:** Honey solutions A-E were prepared from pure honey. The study was carried-out on rapidly proliferating seeds (*Sorghum bicolor*) and the mean radicle lengths (mm) were taken after 24, 48 and 72 hours. **Results:** The seeds treated with methotrexate showed a remarkable retarded radicle growth compared to control. While those treated with honey solutions A-C showed a high degree of inhibition of proliferation of the seeds radicle. Their effects were significant ($P < 0.05$) at 48 and 72 hours. Honey solutions D and E showed a total cytostatic effect on the seeds throughout the study. **Conclusion:** The study reveals that Nigerian honey has a potential for causing antiproliferative and cytostatic effects on rapidly proliferating cells.

Keywords: Nigerian honey, antiproliferative, cytostatic effect, *Sorghum bicolor*.

INTRODUCTION

Cancer occurs as a result of series of events that basically alter the properties and functioning of cells. Cancer is a significant health problem and also a major cause of death in both developing and developed countries all over the world. The World Health Organization (WHO) has ranked it among the leading causes of death, as about 7.4 million (13%) deaths in 2004 were as a result of cancer. It has also been estimated that this death ratio will rise to about 11.5 million deaths in the year 2030. [1] Some of the hallmark of cancer are uncontrollable and sustained proliferation, with others being evasion of apoptosis, angiogenesis and metastasis. [2] The use of the current agents and methods available for treatment cannot reverse these processes and their effects once initiated, but may only control/terminate the process if diagnosis was at the early stage. Therefore, prevention of cancer is better than treatment. Chemoprevention simply refers to the use of pharmacological or dietary agents to prevent induction, inhibit or even delay the advancement of cancer. [3] Therefore, these agents should have antiproliferative or even cytostatic effect to achieve this aim. There have been recent claims of various naturally occurring products having anticancer effect, [4] with honey being one of these products. Honey is a naturally occurring product from bee, and its health benefits have long been identified as far back as 1700 BC. [5] In Nigeria, a lot of its pharmacological application have been explored which includes its wound healing property, antimicrobial property and antiulcer property. [6] But however, the antiproliferative and cytostatic property of Nigerian honey have not been proven scientifically. The aim of this study is to evaluate if Nigerian honey has antiproliferative as well as cytostatic potentials. The study was carried-out on rapid proliferating seeds (*Sorghum bicolor*).

MATERIALS AND METHOD

Materials

Pure honey from the African honey bee (*Apis mellifera adansonii*) was purchased from a retail outlet in Terminus, Jos, while methotrexate injection (Korea United Pharm.Inc) was purchased from Tarhaf pharmacy, Jos.

Experimental Plant

Guinea corn (*Sorghum bicolor*) was purchased from Angwan-rukuba market, Jos. They were then subjected to viability test by putting them in a container having water in it. The floating ones were discarded, while those that submerged were dried and cleansed with alcohol before usage.

Antiproliferative and Cytostatic Evaluation

The modified bench-top assay method of Ayinde *et al.*, [7] was adopted for this study. Different honey preparations (i.e Solutions A-E) were made from the pure honey. This was done by diluting different volumes of honey (0.2, 0.6, 1, 2 and 4 ml respectively) with 10 ml of distilled water each. Solution A constitute of honey 0.2 ml and water 10 ml, Solution B contains 0.6 ml honey and 10 ml water, Solution C contains 1 ml honey and water 10 ml, Solution D contains 2 ml honey and 10 ml water, while Solution E contains 4 ml honey and water 10 ml. Methotrexate was prepared to a concentration 50 µg/ml. The petri-dishes were layered with cotton-wool and filter paper (Whatman No. 1). Twenty (20) seeds of *Sorghum bicolor* were placed in each of the petri-dish. The control seeds were treated with 10 ml distilled water, while the methotrexate seeds were treated with 10 ml of 50 µg/ml methotrexate. The test seeds were treated with the various preparations of honey, 10 ml for the seeds in each petri-dish. The first test group was treated with 10 ml of Solution A, the second group 10 ml of Solution B, the third group 10 ml of Solution C, the fourth group 10 ml of Solution D, and the last group 10 ml of Solution E. The seeds were incubated in a dark room and the mean lengths (mm) of radicle emerging from the seeds were measured after 24, 48 and 72 hours respectively. The percentage inhibition was calculated as $[(\text{mean radicle length control} - \text{mean radicle length treated}) / \text{mean radicle length control}] \times 100$. Percentage growth was calculated as $100 - \% \text{ inhibition}$.

Statistical analysis

The data obtained were expressed as mean \pm SEM (Standard Error Mean). Two way ANOVA (Analysis of Variance) and Bonferroni post-test were used to test for significance. $P < 0.05$ were considered significant. Graph pad prism (version 5.02) was used for the analysis.

RESULTS

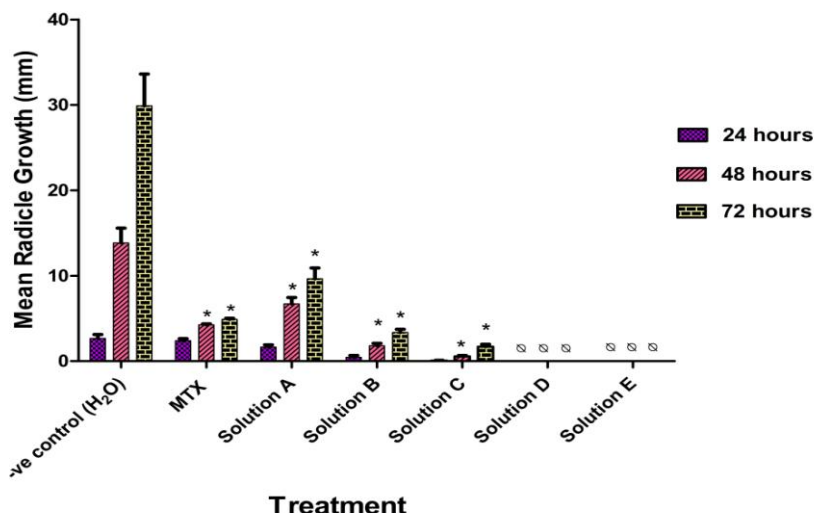


Fig 1: Antiproliferative and cytostatic effects of Nigerian honey on the growth of guinea corn (*Sorghum bicolor*) radicle. Values are Mean ± S.E.M, n=20. * Significant difference from control P<0.05. Ø= Cytostatic (i.e no growth), MTX=Methotrexate.

Fig 2

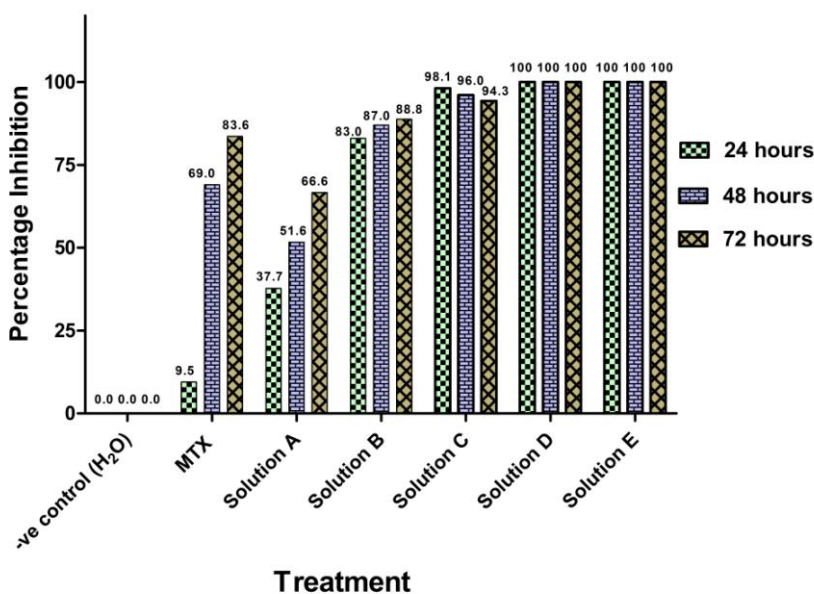


Fig 2: Percentage inhibition of guinea corn (*Sorghum bicolor*) radical by Nigerian Honey. MTX=Methotrexate.

DISCUSSION

Cancer has proven to be a global threat, unleashing death to a significant proportion of the world’s population yearly. [8] History has revealed that natural products are good and reliable source in terms of drug discovery including anticancer agents. Literature has also shown that more than 60% of the anticancer agents presently used were derived from natural sources. [9, 10] Several cancer therapies have been developed but however, these have not been able to effectively curtail the ailment though a couple of successes have been achieved. Also with the fact that most of these therapies elicit a high degree of adverse effects, have led to the search for a more effective and reliable anticancer agent, with minimal adverse effects. This search have also encompassed for agents that may be used for both curative and prophylactic purposes. The use of the radicles of growing seeds as a parameter for the initial screening of

suspected anticancer agents have been demonstrated by McLaughlin et al., [11] Obuotor and Onajobi, [12] Shogbaiké et al., [13] and Chinedu et al., [14] previously. Cancer cells show a high degree of proliferation and this also occurs with meristematic cells of seeds (including *Sorghum bicolor*) at favourable conditions. [15] These facts justify the adoption of this method for this study.

The results gotten from the study shows that the control seeds radicle grew unhindered for the 72 hours of study, with very remarkable growth every 24 hours. This shows that the seeds of *Sorghum bicolor* are capable of proliferating fast under suitable conditions without any form of interference. But however, the seeds treated with methotrexate showed a remarkable retarded radicle growth compared to control. This effect was significant (P<0.05) at 48 and 72 hours of the experiment [Figure 1]. This shows that methotrexate is a potent anticancer agent and is

capable of inhibiting fast growing radicles of *Sorghum bicolor* just as it does to cancer cells in the body. The inhibitory effect was highest at 72 hours [Figure 2]. The seeds treated with Solutions A, B and C also showed a high degree of inhibition of the proliferation of the seeds radicle. Their effects were significant ($P < 0.05$) at 48 and 72 hours, but the inhibition was highest at 72 hours of experiment [Figure 1, Figure 2]. However, solution D and E showed a total cytostatic effect on the seeds throughout the period of the experiment as no growth was observed. This indicates 100% inhibition [Figure 1, Figure 2]. The results indicate that Nigerian honey at lower doses has a very good antiproliferative effect and at higher doses is capable of causing a cytostatic effect. This indicates that it has a potential not just for managing, but also preventing cancer. This finding on its antiproliferative effect correlates with the finding by other researchers who noted the antiproliferative effect of the honey from their region.[3] The presence of antioxidants such as vitamin c and polyphenols (flavonols and phenol) in Nigerian honey, have been documented in scientific chronicles. [15] Antioxidants have however been known for their anticancer property. [16, 17] Therefore, the antiproliferative and cytostatic effects demonstrated in this study by Nigerian honey can be largely attributed to the antioxidants present in it.

CONCLUSION

The study has revealed that Nigerian honey has a potential for causing antiproliferative and cytostatic effects on rapidly proliferating cells. We propose that further works on it using human cell lines should be done.

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