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Preliminary assessment of the antiproliferative potential of *Ananas comosus* (pineapple) fruit juice

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ARTICLEINFO	ABSTRACT					
Article Type:	Introduction: Cancer is a complex genetic ailment and a significant public health disease. Reports					
Original Article	have shown that about 30%–40% of all cancers can be prevented by lifestyle and dietary procedures alone. This study was aimed to evaluate the anti-proliferative effect of <i>Ananas comosus</i> fruit juice					
Article History:	on rapidly proliferating cells.					
Received: 14 November 2015 Accepted: 12 February 2016	Methods: The study was carried-out on rapidly proliferating seeds of <i>Sorghum bicolor</i> . Five groups consisting of 20 seeds each were used. Groups 1-2 were the control and methotrexate groups respectively, while groups 3-5 were the test groups treated with different concentrations of <i>A. comosus</i> . The mean lengths (mm) of radicle emerging from the seeds were measured after 24, 48					
Keywords:	and 72 hours.					
Ananas comosus	Result: The result showed that the control S. bicolor seeds radicle had a very high growth rate					
Pineapple Antiproliferative potential	throughout the study. The methotrexate treated seeds showed a significant (P <0.05) retardation in growth of radicle length when compared with control, after 48 and 72 hours. The inhibition rate was 73.93% and 88.45%. All doses (40%-80% v/v) of <i>A. comosus</i> fruit juice showed a remarkable inhibition of seeds radicle growth, after 48 and 72 hours. The percent inhibition after 48 and 72 hours for 40% v/v was 72.39 and 84.36%, for 60% v/v was 53.22 and 72.58%, while that of 80% v/v was 87.35 and 94.78%, respectively. The percent inhibition at 80% v/v was also higher than that produced by methotrexate throughout the study.					
	Conclusion: The findings from the study indicate that <i>A. comosus</i> fruit juice has an antiproliferative effect.					

Implication for health policy/practice/research/medical education:

Ananas comosus fruit juice effectively inhibited the growth of rapidly proliferating Sorghum bicolor seed radicle. It is, therefore, a potential anticancer agent which may be developed and employed for cancer prevention and treatment.

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Introduction

Cancer is a complex genetic ailment and a significant public health disease. It is caused by a variety of factors which include chemicals, unhealthy diet, genetic mutation and environmental factors. Cancer has transformed into a major public health concern globally, due to the high death rate it produces. In 1996 alone, about 10 million new cases of cancer with an accompanying 6 million cancer deaths were recorded worldwide. In 2004, about 7.4 million death cases by cancer were reported, while 7.6 million deaths caused by cancer were recorded globally in 2008. There are predictions that the death rate may increase to about 12 million by 2020 and even further to about 15 million by 2030 (1,2). The most prominent characteristics of cancer cells include uncontrollable sustained proliferation, evasion of apoptosis, angiogenesis and metastasis (3). Though various medical procedures (such as surgery, the use of radiation and anticancer drugs) are currently being used in taming this ailment, their cost and side-effects have also generated a lot of concern. Therefore, the most effective approach still remains prevention. Reports have shown that about 30%–40% of all cancers can be prevented by lifestyle and dietary procedures alone (4). Therefore, various natural dietary products are now being seen as non-toxic, cost-effective anticancer tools. Literature has even revealed that about 60% of the currently used anticancer drugs were sourced from natural products including plants (5). *Ananas comosus* (pineapple) belongs

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to the family Bromeliaceae. Its fruit is cylindrical in shape, with a scaly brown, green or yellow skin, fibrous yellow flesh and a crown of spiny on it. Its fruit juice is widely consumed in various parts of the world due to its taste, flavor, medicinal and nutritional values. Its medicinal qualities have been employed traditionally in various part of the world, including South America, China and South East Asia (6). Its fruit juice is used as a diuretic agent, antidote for sea sickness, gargle for sore throat and as an anti-inflammatory agent. It has been stated that natural products from fruits including pineapple, may serve as a useful anticancer tool due to their nutritional composition (7). Literature have revealed it to be a valuable source of vitamin C, copper, vitamin B₁, vitamin B₂, vitamin B₃, vitamin B_{6} , vitamin B_{12} , vitamin E and pantothenic acid. Antioxidants (including vitamin C and E) have been known to produce anticancer activity (8). The aim of this study was to evaluate the anti-proliferative effect of A. comosus fruit juice on rapidly proliferating cells, using bench-top assay method.

Materials and Methods

Materials

Methotrexate injection (Korea United Pharm. Inc., Korea) was purchased from Tarhaf Pharmacy, Jos.

Plant material

Collection and authentication

A. comosus fruit was harvested from a local garden, identified and authenticated at the Federal College of Forestry Jos by Mr. Jeff Azila.

Extraction

The fruit was washed and the outer skin covering was removed using a sterile knife. After that, the fruit juice was extracted using a manual juice extractor. The resultant juice obtained was collected in an aseptic manner into a sterile vessel. It was stored at 4°C till used.

Experimental plant (Sorghum bicolor)

The experimental plant, guinea corn (*Sorghum bicolor*), was procured from Angwan-rukuba market, Jos. It was subjected to a viability screening by placing it in a vessel with water. The floating seeds were discarded, while the submerged ones were dried for usage and cleansed with alcohol.

Anti-proliferative assessment

The bench top assay method described previously by Ayinde et al, (9) with some minor modifications, was employed for this study. Diverse concentrations of *A. comosus* fruit juice (40%, 60% and 80% v/v) were prepared. Methotrexate was prepared to a concentration 50 µg/mL. Five Petri dishes were layered with cotton wool and filter paper (Whatman No. 1). Twenty seeds of *S. bicolor* each, were placed in the individual Petri dishes. The control seeds were treated with 10 mL distilled water. The methotrexate seeds were treated with 10 mL of 50 µg/mL methotrexate. The test seeds were treated with the different fruit juice preparations. Specific Petri dishes received 10 mL of a particular concentration (i.e., the seeds in a specific Petri dish were treated with 40% v/v concentration, seeds in a different Petri dish received 60% v/v and seeds in the last Petri dish received 80% v/v concentration). All seeds in the different Petri dishes were incubated in a dark room. The mean radicle lengths (mm) from the seeds were measured after 24, 48 and 72 hours. The percent growth was calculated as [mean radicle length treated/mean radicle length control] \times 100. Percentage inhibition was calculated as 100 – % growth.

Statistical analysis

The data obtained were expressed as mean \pm standard error of mean. Two-way analysis of variance (ANOVA) and Bonferroni post hoc test were used to test for significance. *P*<0.05 was considered significant. Graph pad prism (version 5.02) was used for the analysis.

Results

From the result gotten, the control seeds radicle had a high growth rate throughout the study. The methotrexate treated seeds showed a significant (P < 0.05) retardation in growth of radicle length when compared with control. The inhibition was however significant (P < 0.05) after 48 and 72 hours of experiment, with inhibition rate of 73.93% and 88.45%, respectively. All doses (40%-80% v/v) of *A. comosus* fruit juice showed a remarkable inhibition of *S. bicolor* seeds radicle growth. The inhibition was significant (P < 0.05) after 48 and 72 hours of experiment at all doses. The percentage inhibition after 48 and 72 hours for 40% v/v was 72.39% and 84.36%, for 60% v/v was 53.22% and 72.58%, while that of 80% v/v was 87.35% and 94.78%, respectively (Figure 1, Table 1).

Discussion

Cancer is a global phenomenon which elicits a significant mortality rate to the world's population every year (10). This has led to series of research work by the scientific community, which produced the various anticancer agents currently being used. However, an ideal anticancer agent has not been yet developed. This has rekindled interest in research aimed at discovering better anticancer

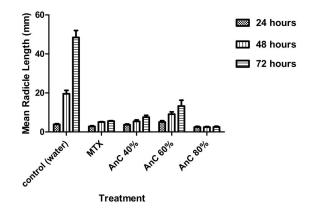


Figure 1. Anti-proliferative effect of *Ananas comosus* (pineapple) juice on the growth of guinea corn (*Sorghum bicolor*) radicle. Values are mean \pm SEM, n = 20. * = P < 0.05 compared with control, MTX = Methotrexate, AnC = *Ananas comosus*.

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Treatment	Percent growth			Percent inhibition		
	24 h	48 h	72 h	24 h	48 h	72 h
Control (water)	100	100	100	0.00	0.00	0.00
MTX	73.27	26.07	11.55	26.73	73.93*	88.45*
AnC 40% v/v	98.83	27.61	15.64	1.17	72.39*	84.36*
AnC 60% v/v	50.13	46.78	27.42	49.87	53.22*	72.58*
AnC 80% v/v	62.98	12.65	5.22	37.02	87.35*	94.78*

Table 1. The growth inhibitory effect of Ananas comosus (pineapple) fruit juice on Sorghum bicolor radicle

Abbreviations: MTX, Methotrexate; AnC, Ananas comosus.

* *P* < 0.05 compared with control.

agents. This research interest has been directed towards plants, especially. This is because plants have proven to be a bankable source for drug discovery (11). Literature has shown that most fruits and vegetables have anticancer property. This effect has, however, been attributed to their nutritional content. A. comosus fruit juice is widely consumed, with speculations of it having anticancer potential. Uncontrollable high proliferation rate is one of the basic characteristic of cancerous cells. Therefore, it has been established that agents capable of producing anti-proliferative effects are potential anticancer agents. Therefore, the effect of suspected anticancer agents on proliferation is now a commonly used parameter in screening for potential anticancer agents. The use of various bench top procedures which involves rapidly proliferating seeds radicles as parameter in the screening of suspected potential anticancer agents have been previously employed by Sogbaike et al, (12) McLaughlin et al, (13) Ayinde and Agbakwuru, (14) and Chinedu et al (15). Just like cancerous cells, meristematic cells of seeds (including S. bicolor) also show a high proliferation rate under favorable conditions (9). This justifies the adoption of this procedure for this study.

The results showed that the control seeds radicle had a high growth rate throughout the study. This proves that at favorable conditions S. bicolor seeds are capable of proliferating rapidly, just like cancer cells. The methotrexate treated seeds showed a significant (P < 0.05) retardation in growth of radicle length when compared with control. The retardation of radicle was due to inhibition by methotrexate. This justifies its current use as an anticancer agent. The inhibition was however significant (P < 0.05) after 48 and 72 hours of experiment, with inhibition rate of 73.93% and 88.45%, respectively. All doses (40%-80% v/v) of A. comosus fruit juice showed remarkable inhibition of S. bicolor seeds radicle growth. The inhibition was significant (P < 0.05) after 48 and 72 hours of experiment at all doses. The percent inhibition after 48 and 72 hours for 40% v/v was 72.39% and 84.36%, for 60% v/v was 53.22% and 72.58%, while that of 80% v/v was 87.35% and 94.78% respectively (Figure 1, Table 1). The result therefore shows that A. comosus fruit juice dose of 80% v/v gave a better anti-proliferative effect than 40% and 60%. The percent inhibition at 80% v/v was also higher than that produced by methotrexate throughout the study. This therefore suggests that A. comosus fruit juice is a potential anti-proliferative agent. Its antiproliferative property may be attributed to the presence of antioxidants (vitamin C and E) as part of its constituent (16). Antioxidants are known to produce anti-proliferative effects and are implicated in cancer prevention (17-19). This has led to encouragement of increase consumption of foods rich in antioxidants such as fruits and vegetables. Therefore, the result gotten from this study is in line with previous reports concerning natural products which contain antioxidants.

Conclusion

The finding from the study indicates that *A. comosus* fruit juice has an anti-proliferative effect. We therefore recommend that, further work should be carried-out on this property using animal models.

Authors' contributions

All authors contributed significantly to the success of the work. All read the final edition and confirmed it.

Conflict of interests

None.

Ethical considerations

Ethical issues (including plagiarism, misconduct, data fabrication, falsification, double publication or submission, redundancy) have been duly observed by authors.

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