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Purple Fleshed Sweet Potato: A Natural Source of Anti Cancer Compounds and Its Pharmacognostic Evaluation

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Abstract
Purple fleshed sweet potato *(Ipomoea batatas)* is a dicotyledonous plant belongs to the family, Convolvulaceae. They are rich in, starch, amino acids, vitamins and minerals. High concentration of anthocyanin and presence of wide variety of antioxidants, including beta-carotene, lutein and zeaxanthin help to protect cells from damage, reduce inflammation and can lower the risk of chronic diseases like heart disease, cancer and cognitive decline. Despite their natural sweetness, they have a relatively low glycaemic index and can regulate blood sugar levels and prevent spikes in insulin levels, making them a good choice for people with diabetes. Sweet potato is a highly valuable food crop in the present scenario and hence phytochemical properties and anti-cancer studies were carried out using the tubers of purple fleshed sweet potato. Results revealed the presence of phytochemicals such as alkaloids, flavonoids, phenol, quinones, saponins, steroids, tannin, terpenoids and protein while using water, ethyl acetate and water extracts. In vitro cytotoxicity assay conducted with cell line DLA (Dalton's Lymphoma Ascites) proved 60.5% efficacy of water extract.

Key words: *Ipomoea batatas*, PFSP, Phytochemicals, In vitro cytotoxicity, DLA cell line.

1. Introduction

Purple Sweet potato (*Ipomoea batatas* L.; Lam.) is a perennial dicotyledonous plant belongs to the morning glory family Convolvulaceae. The intense purple colour in skin and flesh of this sweet potato is due to the richness of anthocyanin (Zhang et al., 2022). Purple fleshed sweet potato pulp following distinct cooking processes, are important sources of nutrients that are beneficial to human health (Basilio et al., 2024). Anthocyanin from purple sweet potatoes were having health properties against several health condition, for instance obesity, inflammation (Wang et al., 2017), hyperglycemia (Matsui et al., 2002), oxidative stress (Zhang et al., 2016) and cancer (Lim et al., 2013). Thus, purple fleshed sweet potato was proposed as functional food and ingredients of nutraceutical to attenuate this major health problem and maintain physiological function in order to promote the human wellness (Das et al., 2012). Research on nutraceutical properties of purple sweet potatoes stated that extracted anthocyanin exhibit strong radical scavenging activity, antimicrobial activity, reduce high blood pressure and liver injury in rats (Zhang et al., 2016). The amino acid Glutamin (Glu) and Aspartic acid (Asp) are the two highest amino acids present in central and west java purple sweet potato cultivars and possess high amount of protein and carbohydrate. So purple sweet potatoes have been established as a source for human nutrition requirements

(Kurnianingsih et al., 2019). Amino acid is very important as a precursor for body function as a regulator of metabolism, growth, development in order to maintain whole body homeostasis (Wu 2013).

The plant encompasses various phytochemical compounds, each harbouring distinct pharmacological properties (Panda et al., 2012). The phytochemicals are the most vital sources for the treatment of destructive diseases. Different phytochemicals have an extensive range of activities, which helps to enhance the immune system and give resistance against long term disease to protect the body from harmful pathogens (Rajasekaran et al., 2003). Phytochemical content in sweet potatoes can be affected by the genetic colour of tuber roots. The purple and red colour of tuber flesh were reported higher flavonoid and phenolic content than other white flesh (Brown 2005). Qualitative phytochemical screening revealed the presence of alkaloids, phenolics, flavonoids, glycosides and tannins in purple sweet potato cultivars of Lawang and Kawai Mountain (Alam et al., 2022). Purple fleshed sweet potatoes are an excellent source of phenolic acids and flavonoid compounds, and some of the most common phenolic acids found in these tubers are chlorogenic acid and ferulic acid (Guclu et al., 2023). This acid can provide the antioxidant properties, anti-inflammatory effects, potential blood sugar regulation and a neuroprotective effect (Gomez-Gomez et al., 2019). Sugata et al., (2015) reported extracts of purple fleshed sweet potato TNG 73 may inhibit the growth of some cancer cell lines, such as human breast cancer (MCF-7), gastric cancer (SNU-1) and colon adenocarcinoma (WiDr), in concentration- and time-dependent manner. Studies conducted by Budiman et al., 2021 also reported anthocyanin in the purple sweet potatoes inhibited breast, colon and stomach cancer cells proliferation with IC50 (50% inhibitory concentration) and they can also promote apoptosis, cell cycle arrest, inhibit proliferation, cell growth inhibition and inhibit cancer progression. Studies done by Vishnu et al., (2019) reported anthocyanin in purple sweet potato leaf were also having anti-cancer properties.

The anticancerous activity of purple fleshed sweet potatoes remains a relatively unexplored area of research, with only a few studies having examined their potential in this regard. Hence for the present study selected purple fleshed sweet potato as the plant material and the phytochemical analysis using ethyl acetate, chloroform and water were carried out and *in vitro* cytotoxicity was conducted using Dalton's Lymphoma Ascites cells (DLA) cell line. The *in vitro* cytotoxicity studies have proved that the tuber of sweet potato can inhibit the growth of Dalton's Lymphoma Ascites cells (DLA).

2. Materials and Methods

2.1. Plant material

Purple Fleshed Sweet Potato (PFSP) belongs to the family Convolvulaceae is selected for the study. The plants were cultivated at the St Dominics college campus of Podimattom, Kanjirappally and tubers were harvested during the month of December 2024. The flesh of the tuber was subjected to Soxhlet extraction, phytochemical screening and in vitro cytotoxicity assay.

2.2. Extraction by Soxhlet apparatus

The flesh of PFSP tuber (1 kg) was air dried under shade at normal temperature then it was powdered in a mixer grinder and further used for Soxhlet extraction process. The bioactive compounds from the plant were extracted by using solvents such as ethyl acetate, chloroform and distilled water in Soxhlet extraction apparatus. The known volume of PFSP powder (800g) was weighed, packed and placed in the Soxhlet apparatus along with 500 ml of solvent at their corresponding boiling points and run continuously for 4-5 hours (until almost no plant residues was left in the recycled solvents) respectively in order to get the bioactive compounds (Parvathy et al., 2025). After 5 hours of Soxhlet extraction the crude extracts of 25

ml were collected by distillation and then stored at 4º C in screw capped tubes. These stored plant extracts were used for the phytochemical screening and anti-cancer study.

2.3. Phytochemical screening

The presence of different phytochemicals extracted in different solvents was confirmed by using standard protocols. Test for alkaloids and protein were conducted following the method of Santhi et al., (2011), test for alkaloids and quinones were conducted following the method of Evans (2002), test for phenol and saponins were conducted following the method of Pradeep et al., (2014), test for steroids was conducted following the method of Khan et al., (2010), test for tannins was conducted following the method of Yusuf et al., (2013) and test for terpenoids was conducted following the method of Siddiqui et al., (2009).

2.4. In vitro cytotoxicity study

The test compound was studied for short term in vitro cytotoxicity using Dalton's Lymphoma Ascites Cells (DLA). The test compound was dissolved in DMSO and concentration range between 1.25 µg/ml to 0.5 µg/ml was used for study. The tumour cells aspirated from the peritoneal cavity of tumour bearing mice were washed thrice with PBS or normal cell lines. Cell viability was determined by trypan blue exclusion method. Viable cells suspension (1×10^6 Cells in 0.1 ml) was added to tubes containing various concentrations of the test compounds and the volume was made up to 1 ml using phosphate buffered cell line (PBS). The control tube contained only cell suspension. These assay mixtures were incubated for 3 hours at 37^0 C. Further cell Suspension was mixed with 0.1 ml of 1% trypan blue and kept for 2-3 minutes and loaded on a haemocytometer. Dead cells take up the blue colour of trypan blue while live cells do not take up the dye. The number of stained and unstained cells were counted separately.

3. RESULTS AND DISCUSSION

3.1. Phytochemical Screening

3.1.1 Test for Alkaloids

2 ml of crude extract in addition with 2 ml of concentrated HCl followed by few drops of Mayer's reagent was subjected to alkaloid screening. The water extract showed positive result due to the presence of yellow precipitate while ethyl acetate and chloroform extract showed negative result (Table 1).

3.1.2 Test for Flavonoids

2 ml of crude extract in addition with 1 ml of 2 N sodium hydroxide was subjected to flavonoid screening. Water and ethyl acetate extract showed positive result due to the presence of yellow precipitate and chloroform extract showed negative result (Table 1).

3.1.3 Test for Phenol

1 ml of crude extract in addition with 2 ml of distilled water followed by few drops of 10% ferric chloride was subjected to phenol screening. Water and ethyl acetate extract showed positive result due to presence of green colour and chloroform extract showed negative result (Table 1).

3.1.4 Test for Quinones

1 ml of crude extract in addition with concentrated sulphuric acid was subjected to quinone screening. Water and ethyl acetate extract showed positive result due to presence of red colour and chloroform extract showed negative result (Table 1).

3.1.5 Test for Saponins

2 ml of crude extract in addition with 2 ml of distilled water and shaken in a graduated cylinder for five minutes length wise was subjected to saponin screening. Water extract showed positive result due to the

formation of 1 cm layer of foam while ethyl acetate and chloroform extract showed negative result (Table 1).

3.1.6 Test for Steroids

1 ml of crude extract in addition with 1 ml of chloroform and few drops of concentrated sulphuric acid was subjected to steroid screening. Chloroform and ethyl acetate extract showed positive result due to the appearance of brown ring while water extract showed negative result (Table 1).

3.1.7 Test for Tannins

1 ml of crude extract in addition with 2 ml of 5% of ferric chloride was subjected to tannin screening. Water extract showed positive result due to the presence of greenish black colour while chloroform and ethyl acetate extract showed negative result (Table 1)

3.1.8 Test for Terpenoids

0.5 ml of crude extract in addition with 2 ml of chloroform and concentrated sulphuric acid was subjected to terpenoid screening. Water extract showed positive result due to the presence of red brown precipitate while chloroform and ethyl acetate extract showed negative result (Table 1).

3.1.9 Test for Protein

1 ml of crude extract in addition with 2 ml of 5% ferric chloride was subjected to protein screening. All the extracts showed positive result due to the presence of violet colour (Table 1).

Table 1: Table showing the results of phytochemical screening of PFSP tuber

Test Performed	Results					
	Water extract		Ethyl acetate extract		Chloroform extract	
	+ve (Present)	-ve (Absent)	+ve (Present)	-ve (Absent)	+ve (Present)	-ve (Absent)
Alkaloids	+ve	-ve	+ve	-ve	-ve	-ve
Flavonoids	+ve	-ve	+ve	-ve	-ve	-ve
Phenol	+ve	-ve	+ve	-ve	-ve	-ve
Quinones	+ve	-ve	+ve	-ve	-ve	-ve
Saponins	+ve	-ve	-ve	-ve	-ve	-ve
Steroids	-ve	-ve	+ve	-ve	+ve	-ve
Tannins	+ve	-ve	-ve	-ve	-ve	-ve
Terpenoids	+ve	-ve	-ve	-ve	-ve	-ve
Proteins	+ve	-ve	+ve	-ve	+ve	-ve

The results of our study clearly showed that the PFSP tubers contains alkaloids, flavonoids, phenols, quinones, saponins, steroids, tannin, terpenoid and proteins (Table 1). The results were supported by some other works. Pharmacological activity and mechanism of action depends on the phytochemical compounds. The study conducted by Laveriano-Santos et al., (2022) provides an in-depth analysis of the phytochemical constituents and highlights the potential of purple sweet potatoes, rich in anthocyanin, as a unique food option and source of functional ingredients for healthy food product. They also emphasized the need for further research on the impact of processing and cooking methods on the retention of these bioactive compounds. Studies done by Alam et al., (2022) showed that qualitative phytochemical profiles of purple sweet potatoes from Lawang and Kawi Mountain revealed the presence of alkaloids, phenolics,

flavonoids, glycosides and tannin. However, the cultivar of Lawang was a relatively low color intensity for alkaloid and higher colour intensity for flavonoid. Giner (2019) and Chia-Lin et al., (2016) reported the various other bioactive compounds in purple sweet potato. These include, alkaloids (Indole-3-aldehyde), triterpenol (Boehmerol and Batatasenol), monoTerpenes (− l-Menthone and (+)-2-Bornanone) and some sesquiterpenes, polysaccharides such as Fructans, pectin, hemicellulose and cellulose has been reported by Gou et al., (2019) and Tang et al., (2019). Ina et al., (2017) reported the highest content of flavonoid at 182.28 ppm and the lowest rate of saponins at 7.99 ppm.

3.2 In vitro cytotoxicity study

0.5 µl/ml of crude extract which was dissolved in DMSO were subjected for short term in vitro cytotoxicity using Dalton's Lymphoma Ascites cells (DLA). The sample showed 26.18±0.5 % of cell death while using 0.5µl/ml extract. 1.0µl/ml, 1.5 µl/ml, 2.0µl/ml, of drug concentration showed 31.6±1.6, 45.2±0.9 and 60.0±1.5 % cell death respectively (Table 2 & Figure 1).

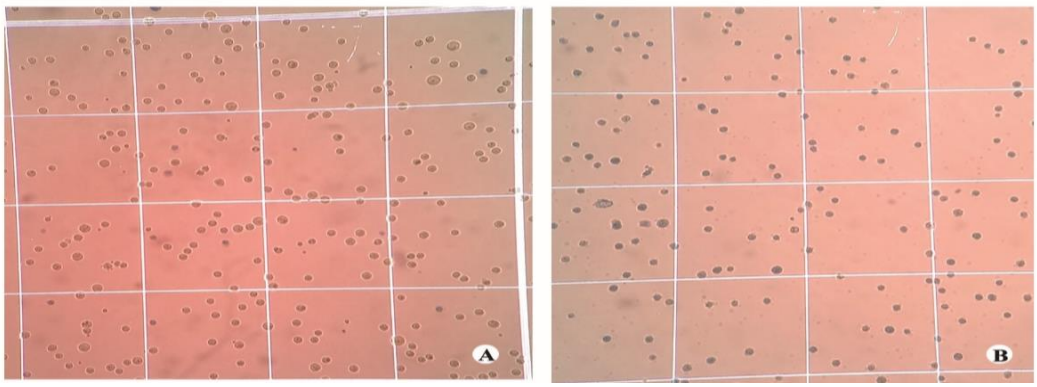
Table 2: Table showing in vitro cytotoxicity results of sweet potato using DLA cell line.

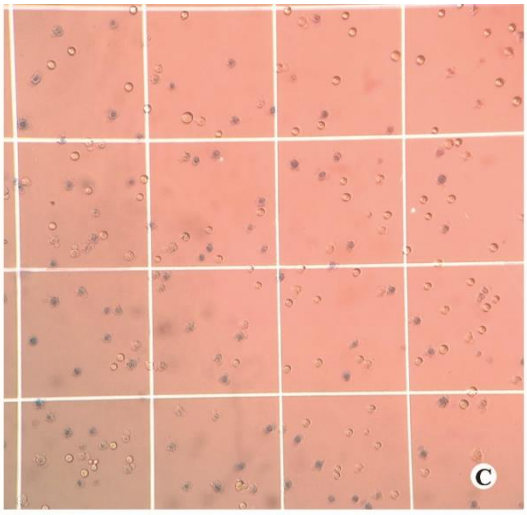
Sl. No.	Drug concentration(µl/ml)	%cell death
1	0.5	26.18±0.5
2	1.0	31.6±1.6
3	1.5	45.2±0.9
4	2.0	60.0±1.5

The present study clearly showed tubers of PFSP have 60.0% of cell death in DLA cell line and it thereby proved highest efficiency of killing the cancer cell lines. There is only one study was reported earlier to prove the anticancer activity of PFSP tubers (Sugata et al., 2015). This is the first report in anticancer activity of DLA cell line using PFSP tuber (Table 2 & Figure 1).

PFSP has been known to possess high amount of anthocyanin which contribute to its antioxidant activity. The studies done by Sugata et al., (2015) showed that PFSP extracts did not have cytotoxic effect on RAW 264.7 murine macrophage cells; they even increased the cell viability (5 × 104 cells/mL) within 24 h. In a higher concentration of RAW 264.7 cells (1 × 105 and 2 × 105 cells/mL), high amount of purple sweet potato extracts could reduce macrophage cells viability. Despite their potential, there have been limited studies on the anti-cancerous activity of PFSP.

Figure 1: *In vitro* cytotoxicity assay result of PFSP tubers with DLA cell lines. (A). 100%live cells control, (B). 100% dead control and (C). 60.0% dead cells with 2.0 µl/ml water extract.





4. Conclusion

The esteemed nutritional density and phytochemical composition of purple fleshed sweet potatoes have garnered considerable interest in their potential health benefits, particularly in the context of cytotoxicity studies, which have led to a deeper understanding of their anticancer properties. The study investigated the cytotoxic potential of PFSP extracts on the mice DLA cell line using invitro cytotoxicity assay, while also examining the correlation between cytotoxic effects and the phytochemical profiles of sweet potatoes. This research emphasizes the potential of incorporating sweet potatoes into diet as a preventive measure against cancer, promoting health and wellness through natural dietary sources.

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