

UNVEILING THE TOXICOLOGICAL PROFILE OF DIFFERENT PARTS OF YACON (*Smallanthus sonchifolius*)

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ABSTRACT: *Smallanthus sonchifolius* (yacon) is a functional food and medicinal plant recognized for its rich phytochemical composition and diverse biological activities. However, information regarding the cytotoxicity of its different plant parts remains limited. This study evaluated the preliminary cytotoxicity of methanolic extracts from the flowers, leaves, stems, tubers, and tuber peels of *Smallanthus sonchifolius* using the Brine Shrimp Lethality Assay (BSLA). Mature yacon plants were collected from Doalnara Aposkahoy, Claveria, Misamis Oriental, Philippines. Approximately 50 g of each powdered plant sample was extracted with 95% methanol, and crude extracts were prepared at concentrations of 125, 250, 500, 1,000, and 2,000 ppm. Each concentration was evaluated in triplicate using *Artemia salina* nauplii. Mortality was recorded after 24 h of exposure, and the median lethal concentration (LC_{50}) was determined through Probit analysis. The flower extract exhibited the highest cytotoxic activity, producing 73.33% and 90.00% mortality at 1,000 and 2,000 ppm, respectively. In contrast, the leaf, stem, and tuber peel extracts caused only minimal mortality (3.33–10.00%), whereas the tuber extract produced no mortality at any concentration tested. The estimated LC_{50} values were 1437.72 ppm for flowers, 2319.56 ppm for leaves, 2428.77 ppm for stems, and 2056.38 ppm for tuber peels, while no LC_{50} was obtained for the tuber extract because no mortality was observed. According to Meyer's toxicity criterion (Meyer et al., 1982), crude plant extracts with LC_{50} values greater than 1,000 ppm are classified as non-toxic; therefore, all methanolic extracts evaluated in this study were classified as non-toxic. These findings indicate that the tested yacon plant parts possess low acute cytotoxicity under BSLA conditions and support the potential safety of yacon, particularly its tubers, while suggesting that the flowers contain biologically active constituents worthy of further phytochemical and toxicological investigation.

Keywords: *Smallanthus sonchifolius*, yacon, cytotoxicity, Brine Shrimp Lethality Assay, LC_{50} , *Artemia salina*

1. INTRODUCTION

Yacon (*Smallanthus sonchifolius*) is a perennial plant native to the Andes Mountains of South America [1–3]. It is characterized by dark green leaves with numerous glandular trichomes and yellow, daisy-like flowers [4,5]. Traditionally, yacon has been cultivated as a food crop because of its sweet-tasting tuberous roots, which have long served as a staple food among the Andean people [6,7]. In recent years, yacon has attracted considerable scientific interest because of its rich phytochemical composition and potential health benefits. Previous studies have reported that yacon contains bioactive compounds with antioxidant, anti-inflammatory, and antidiabetic properties, highlighting its potential as a functional food and medicinal plant.

Despite its nutritional and medicinal value, certain plants or specific plant parts may contain toxic compounds that can produce adverse effects when consumed in excessive amounts. Plant toxicity is influenced by several factors, including the type and concentration of secondary metabolites, the amount consumed, duration of exposure, plant part, environmental conditions, and genetic variation among species [8]. Therefore, evaluating the safety of different plant parts is essential before recommending their use for food, medicinal, or pharmaceutical applications.

Previous studies have demonstrated that high concentrations of yacon extracts may produce toxic effects. In particular, yacon leaf extract has been reported to exhibit cytotoxic activity by inducing apoptosis in 3T3 cells, while both leaf and root extracts have shown genotoxic effects at elevated concentrations [9]. These findings suggest that although yacon possesses beneficial bioactive compounds, some plant parts may exhibit adverse biological effects when used at high doses. Consequently, toxicity assessment is necessary not only to establish safe levels of use but also to identify

biologically active compounds with potential pharmaceutical value.

Although the pharmacological properties of *Smallanthus sonchifolius* have been widely investigated, toxicological evidence remains limited to only a few plant parts and experimental models. Most studies have focused primarily on the leaves or tubers, whereas the cytotoxic potential of other plant parts, such as the flowers, stems, and tuber peels, has received relatively little attention. Furthermore, comparative studies evaluating the preliminary cytotoxicity of multiple yacon plant parts under the same experimental conditions are scarce. This limitation has resulted in insufficient information regarding the relative cytotoxicity and safety profiles of the different anatomical parts of the plant.

To address this gap, the present study evaluated the preliminary cytotoxicity of methanolic extracts from the flowers, leaves, stems, tubers, and tuber peels of *Smallanthus sonchifolius* using the Brine Shrimp Lethality Assay (BSLA), a simple, rapid, and widely accepted bioassay for the preliminary screening of cytotoxicity in natural products. The median lethal concentration (LC_{50}) of each extract was determined through Probit analysis to provide a comparative assessment of the toxicological profiles of the different plant parts under a common experimental framework.

The findings of this study are expected to contribute to a better understanding of the toxicological properties of different parts of the yacon plant. The results may provide scientific evidence supporting the safe utilization of yacon as a functional food while serving as a basis for future pharmacological and toxicological investigations of plant parts that may contain biologically active compounds with potential therapeutic applications.

2. MATERIALS AND METHODS

Plant Sample Collection and Preparation

Mature yacon (*Smallanthus sonchifolius*) plants were collected from Doalnara Aposkahoy, Claveria, Misamis Oriental, Philippines, using purposive sampling. This is why parts of yacon are selected and the site are chosen where the plant is abundant. Only plants that were four to five months old after planting were selected for the study.



Figure 1. Map of Brgy. Aposkahoy Claveria, Misamis Oriental (PhilAtlas)

The collected plant samples were washed thoroughly and cut into smaller pieces to facilitate drying. The flowers, leaves, and stems were air-dried at room temperature in an air-conditioned room. The tubers and tuber peels were dried in an oven at low temperature under a nitrogen gas atmosphere to minimize oxidation. After drying, all plant parts were ground into a fine powder using a laboratory blender. The powdered samples were stored in sealed containers and kept in a freezer until extraction and analysis.

Toxicological Analysis: Brine Shrimp Lethality Assay

The preliminary cytotoxicity of methanolic extracts from different parts of the yacon plant (*Smallanthus sonchifolius*) was evaluated using the Brine Shrimp Lethality Assay (BSLA) following a standard toxicity testing procedure [10]. Each treatment was performed in triplicate, with ten *Artemia salina* nauplii per replicate, to ensure the reliability of the results.

Approximately 50 g of powdered sample from each plant part was extracted with 95% methanol at a ratio of 1 g of sample to 4 mL of solvent for 48 h at room temperature. The extracts were filtered through Whatman filter paper to remove solid

residues. To maximize extraction efficiency, the plant materials were re-extracted twice using 100 mL of fresh methanol for each extraction. The combined filtrates were concentrated under reduced pressure at 40°C using a rotary evaporator and stored at -20°C until further analysis.

The crude extracts were weighed by gravimetric analysis and used to prepare stock solutions. Serial dilutions of each stock solution were prepared to obtain final concentrations of 125, 250, 500, 1,000, and 2,000 ppm. Each concentration was evaluated using the BSLA, and the mortality of the brine shrimp nauplii was recorded after 24 h of exposure.

The percentage mortality was calculated using Abbott's formula:

$$\% \text{ Mortality} = (Po - Pc) / (100 - Pc) \times 100$$

where:

% Mortality = corrected percentage mortality of *Artemia salina* nauplii

Po = observed percentage mortality in the treatment group

Pc = percentage mortality in the control group

The corrected mortality data were subjected to Probit analysis to estimate the median lethal concentration (LC₅₀), defined as the concentration of extract required to cause 50% mortality of the brine shrimp nauplii [11]. The cytotoxicity of the extracts was evaluated based on the percentage mortality at each concentration and the corresponding LC₅₀ values.

3. RESULTS AND DISCUSSION

Cytotoxic Activity

The preliminary cytotoxicity of methanolic extracts from selected parts of *Smallanthus sonchifolius* was evaluated using the Brine Shrimp Lethality Assay (BSLA). The assay is widely employed as a rapid and reliable bioassay for the preliminary screening of cytotoxicity in natural products, where toxicity is assessed based on the mortality of *Artemia salina* nauplii following exposure to plant extracts [10,12]. The median lethal concentration (LC₅₀), determined through Probit analysis, was used to estimate the acute cytotoxicity of each extract. According to Meyer's toxicity criterion, crude plant extracts with LC₅₀ values below 1,000 ppm are considered toxic, whereas extracts with LC₅₀ values greater than 1,000 ppm are classified as non-toxic. This criterion was adopted throughout the present study for the interpretation of cytotoxicity [10].

The percentage mortality of *A. salina* nauplii exposed to different concentrations of the methanolic extracts is presented in **Table 1**. Mortality varied among the different plant parts, indicating differences in their preliminary cytotoxic activity. The flower extract exhibited the greatest cytotoxic effect, producing no mortality at concentrations up to 500 ppm but causing 73.33% mortality at 1,000 ppm and 90.00% mortality at 2,000 ppm. In comparison, the leaf extract produced only 6.67% and 10.00% mortality at 1,000 and 2,000 ppm, respectively, while the stem extract resulted in 3.33% and 6.67% mortality at the same concentrations. The tuber peel extract showed a maximum mortality of 6.67% at 500 ppm, whereas the tuber extract produced no mortality throughout the concentration range tested.

The LC₅₀ values and preliminary cytotoxicity classifications are summarized in **Table 2**. The flower extract exhibited the lowest LC₅₀ value (1437.72 ppm), followed by the tuber peel (2056.38 ppm), leaf (2319.56 ppm), and stem (2428.77 ppm) extracts. No LC₅₀ value was calculated for the tuber extract because no mortality was observed during the assay. Although the flower extract demonstrated the highest cytotoxic activity among the plant parts evaluated, its LC₅₀ value remained above the toxicity threshold of 1,000 ppm. Consequently, all methanolic extracts from the selected yacon plant parts were classified as non-toxic according to Meyer's toxicity criterion [10].

Table 1. Percentage Mortality of Brine Shrimp (*Artemia salina*) Exposed to Various Concentrations of Extracts from Selected Parts of the Yacon Plant (*Smallanthus sonchifolius*)

Yacon Parts	Concentration (ppm)	Total Number of Survivors	Total Number of Dead	Average Mortality, %
Flowers	Control	30	0	0.00
	125	30	0	0.00
	250	30	0	0.00
	500	30	0	0.00
	1000	8	22	73.33
	2000	3	27	90.00
Leaves	Control	30	0	0.00
	125	30	0	0.00
	250	30	0	0.00
	500	30	0	0.00
	1000	28	2	6.67
	2000	27	3	10.00
Stems	Control	30	0	0.00
	125	30	0	0.00
	250	30	0	0.00
	500	30	0	0.00
	1000	29	1	3.33
	2000	28	2	6.67
Tubers	Control	30	0	0.00
	125	30	0	0.00
	250	30	0	0.00
	500	30	0	0.00
	1000	30	0	0.00
	2000	29	0	0.00
Tuber peels	Control	30	0	0.00
	125	29	1	3.33
	250	30	0	0.00
	500	28	2	6.67
	1000	30	0	0.00
	2000	30	0	0.00

Note: Three replicates with ten (10) nauplii in each trial.

Table 2. Median Lethal Concentration (LC₅₀) and Preliminary Cytotoxicity Classification of Methanolic Extracts from Selected Parts of *Smallanthus sonchifolius* Based on Meyer's Toxicity Criterion (Meyer et al., 1982)

Yacon Parts	Average LC ₅₀ (ppm)	Toxicity*
Flowers	1437.72	Non-toxic
Leaves	2319.56	Non-toxic
Stems	2428.77	Non-toxic
Tubers	--	Non-toxic
Tuber peels	2056.38	Non-toxic

Classification based on Meyer's toxicity criterion (Meyer et al., 1982): LC₅₀ < 1,000 ppm = toxic; LC₅₀ > 1,000 ppm = non-toxic.

The observed differences in cytotoxic activity among the plant parts may reflect variations in their phytochemical composition. The flower extract exhibited the lowest LC₅₀ value (1437.72 ppm) and the highest mortality, indicating comparatively greater biological activity than the other extracts. However, because its LC₅₀ remained above the 1,000

ppm threshold established by Meyer's toxicity criterion [10], it was still classified as non-toxic. Similarly, the leaf, stem, and tuber peel extracts all exhibited LC₅₀ values greater than 1,000 ppm and were therefore classified as non-toxic. No LC₅₀ value was calculated for the tuber extract because no mortality occurred throughout the assay, further supporting its lack of detectable acute cytotoxicity under the experimental conditions.

These findings are consistent with previous reports indicating that yacon leaf extracts exhibited cytotoxic and genotoxic effects only at relatively high concentrations [9]. Likewise, the absence of detectable cytotoxicity in the tuber extract agrees with previous studies demonstrating that yacon extracts did not exhibit cytotoxic effects on normal human cells [13]. Overall, the present findings indicate that the cytotoxicity of *S. sonchifolius* varies among its anatomical parts, with the flowers showing the greatest biological activity. However, all extracts were classified as non-toxic based on their LC₅₀ values, supporting the potential safety of yacon, particularly its tubers, for food and medicinal applications. The comparatively higher cytotoxic activity of the flower extract suggests that it may contain bioactive compounds worthy of further phytochemical characterization and more comprehensive in vitro and in vivo toxicological investigations.

4. CONCLUSION

This study evaluated the preliminary cytotoxicity of methanolic extracts from the flowers, leaves, stems, tubers, and tuber peels of *Smallanthus sonchifolius* using the Brine Shrimp Lethality Assay (BSLA). Among the extracts tested, the flower extract exhibited the highest mortality of *Artemia salina* nauplii, whereas the tuber extract produced no mortality at any concentration evaluated. The leaf, stem, and tuber peel extracts showed only minimal mortality throughout the assay. Probit analysis showed LC₅₀ values of 1437.72 ppm for flowers, 2319.56 ppm for leaves, 2428.77 ppm for stems, and 2056.38 ppm for tuber peels, while no LC₅₀ was calculated for the tuber extract because no mortality occurred. According to Meyer's toxicity criterion [10], all methanolic extracts were classified as non-toxic because their LC₅₀ values exceeded 1,000 ppm. These findings indicate that the tested yacon plant parts possess low acute cytotoxicity under the conditions of the BSLA and support the potential safety of yacon, particularly its tubers, for food and medicinal applications. Nevertheless, the comparatively higher biological activity observed in the flower extract suggests the presence of bioactive constituents that merit further phytochemical characterization and comprehensive toxicological evaluation using in vitro and in vivo models.

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