Phytochemical and Toxicity of Ethanol extract Sijukkot Leaves (*\textit{Lactuca Indica}*)

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**Abstract.** Phytochemical screening on Sijukkot which determined as *Lactuca indica* L, the plants from an area in the village of Parsaora Sibisa Ajibata, Toba Samosir, North Sumatera has been done. Test was carried out to determine the composition of secondary metabolites contained in these plants. It was started by extracting plant leaves using 96% ethanol. Phytochemical test was used to identify compounds from the ethanol extract of sijukkot leaves. The identification results obtained compounds as Flavonoids, tannins, sapoinns, steroids and triterpenoids. Furthermore, the toxicity test was carried out using the BSLT method and the results showed LC_{50}: 11.644 ppm, thus the ethanol extract of the leaves had a strong toxicity.

1. **Introduction**

*Lactuca Indica* L known as sijukkot (vegetables) is found as a wild plant in the Tobasa district of North Sumatra, long believed to be a medicinal plant.

**Figure1.** Sijukkot (*Lactuca Indica*)

The Sijukkot (*Lactuca Indica*) L family of Compositae found around Tobasa North Sumatra is a wild plant, it likes lots of sunshine. Height can reach one meter. Leaves elongated with irregular edges.
and tapered tips. The color of the leaves is green, the middle stems of the leaves are reddish white, with yellow flowers that have a relatively high water content. Stems that grow fresh have a lot of white sap. Lactuca inermis found in Africa is referred to as a perennial herbal [1][Lactuca sativa L which is classified as lettuce in North America was studied researched and developed[2]. Extracts from Lactuca Indica were found to have good antioxidant activity[3]. In plants there are secondary metabolite compounds such as alkaloids, flavonoids, terpenoids, tannins which have activity as medicinal plants, such as antibacterial, anti-oxidant, anti-diabetes, cholesterol even as anti-cancer but also have toxic properties[4-8]. The Sijukkot (Lactuca Indica L) plant is believed to have medicinal properties such as diabetes, cholesterol, hypertension and can also increase energy. As a plant that has potential as a herbal medicine, it is also necessary to test the determination of its acute toxicity. The most common Artemia Salina Leach determination of toxicity can be done by the Brine Shrimp Lethality Test (BSLT) Method. This method is carried out by determining mortality or acute lethality. Experiments are carried out on shrimp expressed as LD50, namely the concentration or dose which causes mortality of half the population of organisms within a certain time.

2. Materials and Method

2.1 Collection of plant
The sample material was sijukkot leaves (Lactuca Indica) from the forest area of the Sibisa parsaoaran village, Ajibata district, Toba Samosir Regency, North Sumatra. Then the sorting process is carried out in order to obtain 20 kg of fresh Sijukkot plant leaves. Furthermore, washing, drying, and refining were carried out in order to obtain 3 Kg of leaf simplicity powder of Sijukkot plant. The drying process is carried out in a place that is protected from sunlight to avoid damage to the bioactive compounds contained in the sample due to direct contact with sunlight.

2.2 Chemicals Used for the Research Were
NaCl, ammonia (Merck), petroleum ether (Merck), chloroform (Merck), HCl (Merck), Dragendorff reagents (sigma), ammonia (Merck), petroleum ether (Merck), chloroform (Merck), HCl (Merck), Mayer reagents, CH3COOH anhydrous (Merck), H2SO4 (Merck) Lieberman-Buchard reagents (Sigma), magnesium plates (Merck), amyl alcohol (Merck), FeCl3 (Merck), Stiassny reagents (sigma), sodium acetate and Sigma NaOH (Merck), Dimethyl sulfoxide (DMSO) and Artemia Salina Leach.

2.3 Extract Preparation
The sample powder was macerated three-fold with ethanol solvent for 3 × 24 h and then filtered with Buchner funnel to separate the filtrate and residue. The obtained filtrate was concentrated in a vacuum rotary evaporator to get ethanol extract.

2.4 Phytochemical Screening of Secondary Metabolites
Phytochemical screening: The crude ethanol extracts of leaves were tested for the presence of alkaloids, flavonoid, steroids, tannins and saponins. The qualitative results are expressed as (+) for the presence and (-) for the absence of phytochemicals

2.4.1 Flavonoid Compound Test Group. Test for the presence of flavonoid compounds was carried out by taking 0.5gram extracts, adding two drops of 5% FeCl3 solution to the drip plate sample. The color change to greenish or black and blue indicates the presence of flavonoids (+).

2.4.2 Alkaloid Group Compound Test. Test for the presence of alkaloid compounds was carried out by adding 0.5 gram of sample with 1 mL of HCl 2N and supplemented with 9 mL of distilled water and then added with 5 drops of dragendorff reagent, the not presence of brick red precipitate indicated of alkaloids (-).

2.4.3 Saponin Group Compound Test. Test for the presence of saponin compounds is done by adding 1 mL of sample with 10 mL of distilled water until 10 minutes and leaving the foam and then

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waiting for up to 10 minutes if the foam does not disappear add 2N HCl. If the foam does not disappear indicating the presence of compound saponins (+).

2.4.4 Test Steroid and Terpenoid Compounds. Test the presence of steroid and terpenoids compounds is done by adding 0.5 grams of extract with 10 drops of acetic anhydrous plus 2 drops of concentrated sulfuric acid shaken and left for a few minutes, the emergence of red and purple color shows positive triterpenoids, the emergence of green and blue color shows positive steroids (+).

2.4.5 Test the Tannin Group Compounds. Test for the presence of tannin compounds was carried out by adding 0.5 gram extract with 10 mL aquadest and added with 3 drops of FeCl3 1%, the appearance of a blackish green color showed positive containing tannin(+).

2.5 Toxicity Test
The cytotoxic activity of Sijukkot leaf extracts (Lactuca indica L.) was assessed using the Brine Shrimp Lethality Test (BSLT) method. This method is easy and simple. The first time the larvae of Artemia Salina were hatched by inserting shrimp eggs into artificial seawater for 24 hours. A total of 20 mg of shrimp eggs is put into an incubator containing synthetic sea water which is made by weighing 38 grams of pure NaCl salt dissolved in 1 l of distilled water. After 24 hours the shrimp eggs hatch and can be used as test animals. Cytotoxic testing was carried out using concentrations of 250, 125, 75, 50 and 25 ppm and was carried out 4 times for each concentration. This concentration was used to see the difference in the effect of each concentration on the mortality of Artemia Salina Leach larvae. 5 ml of each concentration was taken and the ethanol solvent was evaporated. After drying at each concentration 2 mL of 1% DMSO was added to dissolve the dried sample because 1% DMSO is a universal solvent that is not toxic. After that, 10 Artemia Salina Leach larvae and artificial sea water were added to each concentration. Let stand for 6 hours and observed the effect of each concentration after 6 hours.

In observing the growth and development of larvae up to the test for the toxicity of the extract, an observing tool is used, namely a magnifying glass. The total number of deaths was calculated by adding up the larvae that died at each concentration. The average mortality rate of larvae at each concentration is obtained by dividing the total number of larvae deaths by the number of repetitions for each concentration divided by the total number of initial larvae. The calculation of the percentage of larval mortality at each concentration was obtained by multiplying the average mortality rate for each concentration by 100%.

3. Result and Discussion

3.1 Phytochemical Screening
Phytochemical screening carried out on the ethanol extract of the leaves of the Sijukkot plant included testing for alkaloids, flavonoids, saponins, steroids and terpenoids. The results can be seen in Table 1.

| Phytochemicals   | Positive Test Indicator       | Test |
|------------------|-------------------------------|------|
| Flavonoid        | Greenish / black blue         | +++  |
| Alkaloid         | Red                           | -    |
| Saponin          | Foaming                       | ++   |
| Tannin           | Blackish Green                | +++  |
| Triterpenoid     | Red and Purple                | ++   |
| Steroid          | Green and Blue                | +    |

3.2 Toxicity
Result of calculation of concentration variation of Sijukkot leaf ethanol extract (Lactuca indica L.) according to Probit Log Graph method Concentration and mortality of Artemia Salina Leach larvae. The results can be seen in Table 2.)
Table 2. The concentration variation of Sijukkot leaf ethanol extract (*Lactuca indica* L) according, Probit and Mortality of Artemia Salina larvae.

| Concentration (X) | Concentration Log | Mortality (%) | Probit |
|-------------------|-------------------|---------------|--------|
| 125               | 2.096             | 92.5          | 6.43   |
| 75                | 1.875             | 82.5          | 5.93   |
| 50                | 1.698             | 77.5          | 5.75   |
| 25                | 1.397             | 60            | 5.25   |

![Toxicity graph](image)

Figure 2. Toxicity graph of ethanol extract of sijukkot (*Lactuca Indica* L) leaves

The LC50 value was obtained by looking for the antilog of x, the LC50 value of the Sijukkot leaf extract was 11.6439 ppm. The level of toxicity of an extract can be categorized as very toxic if the LC50 ranges from 0-250μg / ml, 250-500 μg / ml: toxic, 500-750 μg / ml: moderate, 750-1000 μg / ml: non-toxic[9]. Plants that have high toxicity have the potential to be developed as medicinal plants and have potential as anti-cancer properties[10-12]. So that the toxicity of the sijukkot plant needs to be studied to see its anti-cancer activity as the clove plant has relevant toxicity with its anticancer properties. Further research is needed to determine the effectiveness of this plant as a medicinal drug[13-14].

4. Conclusion
Phytochemical results in ethanol extract from the leaves of the Sijukkot plant (*Lactuca Indica* L) secondary metabolites were found, namely: flavonoids, saponins, tannins, triterpenoids and steroids. From the results of the toxicity shows a strong toxicity. The results of this study can be used as a prelude for further research to see its bioactivity so that later it can be used as a herbal medicinal plant. As it has been for a long time, local residents have been trusted to treat diabetes and cholesterol for a long time. As a medicinal plant, it needs further research in vitro and invivo as well as dosage factors and also its toxic effects.
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Acknowledgment
I would like to thank the State University of Medan, who funded this research with a contract 104/UN33.8/PL-PNBP/2020.