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HYPOURICEMIC PROPERTY OF THE SEMI-PURIFIED FLAVONOIDS FROM TALONG-TALONGAN (*SOLANUM TORVUM* LINN. FAMILY SOLANACEA) LEAVES

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ABSTRACT
Solanum torvum commonly called as Talong-talongan is a plant that is currently used in Cameeron ethnomedicine for treatment of anti-gout. This plant is said to have a presence of flavonoids which has both a sedative and a diuretic therapeutic effect. Gout is a form of rheumatic disease caused by deposits of uric; Flavonoids display pharmacological properties as cytoprotective and antioxidant agent. The contents of the flavonoid were extracted by macerating about 100 grams of the dried leaves were place in a 500mL glass jar and macerated with a 500mL of 80% Ethyl Alcohol for 48 hours. Samples are subjected to ICR mice, divided into five groups a negative control, which are no treatment, positive control which are administered with allopurinol, the 100mg/mL, 200mg/mL, 300mg/mL of Talong-talongan samples. After a week of induction of high protein diet an increase of blood uric acid level base line was observed. All treatment groups and positive control showed decrease in uric acid levels. Higher dosage can significantly cause injury to the kidney. Therefore the researchers conclude that the flavonoid extract from the Talong-talongan (*Solanum torvum* Linn. A family Solanaceae) leaves has potential hypouricemic property.

KEYWORDS: Hyperuricemic, hypouricemic, flavonoids, uric acid, therapeutic effect.

INTRODUCTION
In the Philippines, the latest prevalence of hyperuricemia leading to gout in nationwide survey conducted in 2003 is 1.6 percent. Gout, also called metabolic arthritis is a disease caused by the buildup of uric acid in excessive amounts in the human body. Hyperuricemia occurs in about two percent of hospitalized patients and less than 0.5 percent of the normal population. In this condition, monosodium urate or uric acid crystals are deposited on the articular cartilage of joints, tendons and surrounding tissues due to high concentration of uric acid in the blood. Gout is most common in men. Gout is caused by too much uric acid in the blood. Almost all of the time, having high uric acid isn't harmful. Many people with high levels in their blood never get gout. But when uric acids levels in your blood are too high, the uric acid can form hard crystals in your joints. Your chances of getting gout or high uric acid are higher if you are overweight by eating too much high, drinking too much alcohol, or eating too much meat and fish that are high in purines.

Xanthine oxidase inhibitors (XOI) are known to be used for the management of gout. Plants and its natural products are worthy to be utilized as a xanthine oxidase inhibitor as they have provided individuals with many possible beneficial effects and found safe for human bodies. Flavonoids are a group of polyphenolic compounds that are distributed in various plant origins with essential beneficial effects. Xanthine oxidase is one of the most important enzymes that are inhibited by some flavonoids.

Talong-talongan leaves were collected from Bureau of Plant Industry (BPI) Malate, Manila. And undergone extraction using 80% ethanol. A fraction was utilized for phytochemical screening, and other portions were subjected for the isolation of semi-purified flavonoids that was used in treatment of induced hyperuricemia mice. Blood collection was done thrice ad was evaluated through blood uric acid strip test.

There were studies that *Solanum torvum* commonly called as Talong-talongan is a plant that is currently used in Cameeron ethnomedicine for treatment of anti-gout (Journal of Ethnopharmacology, 2008). It is an ascending or spreading weed, somewhat branched, hairy herb, 30 to 60 centimeters in height. Stems, petioles, and leaves are armed with scattered, sharp, rather stout spines, 3 to 6 millimeters in length. Leaves are oblong-ovate, 4 to 12 centimeters long. Pointed at the tip, inequilaterally at the base, with

irregularly undulate-lobed margins. Flowers, 1 to 5, are borne in racemes in the axils of leaves. Calyx is green, with a slightly spiny tube. Corolla is violet or purplish, rotate, and shallowly 5-lobed, about 2 centimeters in diameter. And it is found in the waste places throughout the Philippines at low and medium altitude.

The researcher selected ICR mice due to its non-sensitive characteristics and they have a rather similar biological and genetic consistency in human. The same mice are being used too in other countries and in other studies.

Research Methodology

The researchers conducted an experimental method of research. It was conducted in Centro Escolar University, Mendiola, Manila, Philippines.

Solanum torvum Linn. Leaves underwent extraction and were subjected for a physical and chemical test to identify the presence of the flavonoids. The researchers used Fourier Transform Infrared (FTIR) Spectroscopy to identify the presence of constituents from the extract.

Authentication of the Plant Sample

The sample was verified and authenticated in the Bureau of Plant Industry (BPI) Manila, Philippines on June 17, 2017.

Research Procedure

1. Collection of the Plant sample

The *Solanum torvum* leaves used in this study were collected from Bureau of Plant Industry (BPI) Malate, Manila, Philippines during the month of June 2017. The leaves were air-dried and eventually put in the oven to further eliminate the excess moisture.

2. Extraction and Purification of Flavonoids

About 100 grams of the dried leaves were placed in a 500 mL glass jar and macerated with a 500mL of 80% Ethyl Alcohol for 48 hours. The leaves were strained using a muslin cloth and filter paper, and was subjected to Rotary evaporator to remove the solvent from the extract in 40-60˚C and 140-150 rpm.

An equivalent amount of 10 grams of the plant extract was evaporated over a water bath. The residue was triturated with 10 mL 2M HCL and 10mL ethyl acetate and was separated with separatory funnel. This was repeated until the solvent is almost colorless. The lower portion solution was discarded after obtaining the defatted residue.

The defatted residue was dissolved in 5 mL petroleum ether and 20 mL of 80% Ethanol. Any insoluble residue was filtered off. The filtrate was divided into 6 groups. – Group A, B, C, D, E, and F. Group A served as the control.

3. Physicochemical Test

Physical Test

3.1 Test for Solubility

The solubility of the substance was determined using water, ethyl alcohol, chloroform, petroleum ether, and hexane.

3.2 Organoleptic Evaluation

The isolated extract determines its physical appearance, color and odor.

3.3 Chemical Test

3.3.1 Bate-Smith and Metcalf Test Method

Treat one portion of the sample filtrate and treated with 0.5mL of concentrated hydrochloric acid (12M); and it was observed for any color changed. Warm the solution for 15 minutes in a water bath. Observed for further color change in color within an hour and compared with the control. Positive result would be a strong red or violet indicated the presence of leucoanthocyanins.

3.3.2 Wilstatter "Cyanidin" test

Take another portion of the alcohol filtrate and was treated with 0.5mL of concentrated hydrochloric acid (12M). Place three to four pieces of magnesium turnings and observe any color change within 10 minutes; compared the result with the control tube. Positive result was observed colors ranging from orange to red, crimson, and magenta and occasionally to green or blue.

3.3.3 Lead Acetate Test

In test tube, a few mL of Lead Acetate solution were added drop by drop. Formation of yellow precipitate would indicate the presence of flavonoids.

3.3.4 Ammonia Test

In test tube 5 mL of diluted ammonia solution was added to the extract followed by the addition of concentrated sulfuric acid. Yellow coloration which disappears on standing indicates the presence flavonoids.

1.3.5 Shinoda Test

In test tube, a piece of Magnesium ribbon and 2mL of concentrated Hydrochloric acid were added to the extract. Positive result observed was change in red coloration.

2 Instrumental Analysis

4.1 Fourier Transform Infrared Spectrophotometer (FTIR)

In Potassium Bromide disc, a drop liquid sample was placed and was left free standing. The sample holder containing the sample was place in the instrument and scanned by the computer. The printed result was then interpreted and the functional group was determined.

3 Biological Test- Determination of the Hypouricemic Property

5.1 Acclimatization of Experimental Analysis

Twenty-five (25), 20-30grams ICR mice were used in the experiment. The mice will be cleaning of the cages was performed every other day, or as needed. The mice was stored in an air-conditioned room with constant temperature of 23˚C +/- 2˚C and 12 h light/ 12 h darkness photo period and relative humidity of NMT 65% and allowed free access to water. All animals were feed with high protein dog food supported with beans, and legumes twice a day (morning and evening), and were provided a sufficient amount of water daily. A water feeder was placed on the side of the cage for easy access of the mice at any time and it was changed daily.

5.2 Preparation of Experiment to Animals

Twenty-five (25) ICR Mice were used in the experiment. Five (5) of them represent the negative control group untreated, fifteen (15) represented the hyperuricemia group, and the five (5) represented with Allopurinol drug group (positive). The hyperuricemia group was equally divided to represented 2, 3 4 and 5. The treatment models were represented by 3 trials, five mice each.

General description of animal manipulation methods (including method of conditioning) the mice were subjected first top screening by weighing. The researchers used ICR Mice Strain as a subject and was group into five, each group containing 5 mice. Group 1 served as the negative control, group 2 served as the control of the study. The control was treated with Allopurinol and group 3, 4 and 5 were treated with the flavonoid extract of *Solanum torvum* vial oral gavage (100mg/kg, 200mg/kg, and 300mg/kg) respectively for one week. During the induction of the semi purified flavonoid extract from *S. torvum*, the mice were picked up gently. For oral treatment the mice were held with one hand by the scruff of the neck with the mouth held so that the esophagus is as straight as possible. The oral gavage needle fitted to an attached syringe was then carefully inserted between tongue and the roof of the mouth. After 2 hours of inducing the isolated Flavonoids and the positive control Allopurinol. Using Blood Uric Acid Test, collection of blood was conducted after the treatment to test the Uric acid level, and then histopathological study will be conducted.

Statistical Treatment of Data

The researchers conducted a statistical analysis using repeated measures analysis of variance (ANOVA). This type of analysis is appropriate since the researchers are measuring the same rats on different periods (baseline, post induction, post treatment.) Blood uric acid levels that were gathered served as the data. The blood uric acid levels of the rats from the three varying doses were compared to the control group, allopurinol. Statistical significance is set at 0.05 level of probability.

The researchers woul likely use the following formula.

	SS	Df	MS	F
Between	SS(B)	k-1	$\frac{SS(B)}{k-1}$	$\frac{MS(B)}{MS(W)}$
Within	SS(W)	N-k	$\frac{SS(W)}{N-k}$	
Total	SS(W) + SS(B)	N-1		

Presentation, Analysis and Interpretation of Data

This chapter presents the presentation, analysis and interpretation of data obtained by the researchers. It includes the percentage yields, the constituents in physical and chemical testing, with the confirmatory test and determination of the anti-gout activity of the flavonoid content of talong-talongan leaves. The results of the flavonoids content obtained were to accordance on the methods and procedures employed in the study.

Isolation and Calculation of Percentage Yield

The collected plant samples had undergone isolation and purification. The residue was obtained and weighed and calculated to measure the percentage yield of Flavonoids from the plant extract. The results were shown.

Table 1: Percentage Yield of Semi-Purified Flavonoids.

Weight of the Sample	100grams
Weight of the Residue	16.11grams
Percentage Yield	16.11%

Table 1 show that the percentage yields of the Flavonoids of Talong-talongan (*Solanum torvum* Linn. Family Solanaceae) leaves were 16.11%.

Organoleptic Evaluation

The color, odor, appearance and physical state of the Flavonoids extract were determined and the results are shown in Table 1.

Table 2: Results for Organoleptic Evaluations of Flavonoid Extract

Physical appearance	Viscous dark-green
Color	Dark Green
Odor	Characteristics

The Table 2 shows that the Flavonoid extract from the leaves of *Solanum torvum* had a brown in color, with characteristics odor and a Green color liquid solution in Physical appearance.

Test for Solubility

The solubility of Flavonoids extract from Talong-talongan leaves was performing using Distilled Water, Ethyl Alcohol, Chloroform, and Hexane. The results are shown in Table 2.

Table 3 Results for Solubility.

Solvent	Result
Distilled Water	Soluble
80% Ethyl alcohol	Soluble
Chloroform	Insoluble
Hexane	Insoluble

Table 3 shows that the Flavonoids extract of *Solanum torvum* leaves was soluble in Distilled water and Ethyl Alcohol.

Chemical Test

The chemical test performed for the confirmatory of the Flavonoids from the leaves of Talong-talongan and was performed using Bate-Smith and Metcalf test, Wilstatter "Cyanidin" test, Lead Acetate Test, Ammonia Test, and Shinoda test. The results are shown in Table 3.

Table 4: Results of Chemical Evaluations of Flavonoids.

Test perform	Expected result	Actual result	Remarks
Bate-Smith and Metcalf test	Strong red or violet color	Red color solution	Presence of Flavonoids
Wilstatter "Cyanidin "test	Orange-red precipitation	Red precipitate	Presence of Flavonoids
Lead Acetate Test	Formation of yellow precipitate.	Yellow precipitate formed	Presence of Flavonoids
Ammonia Test	Disappearance of yellow precipitate	Disappearance of yellow precipitate	Presence of Flavonoids
Shinoda Test	Red precipitate	Red brown color precipitate	Presence of Flavonoids

Table 4 shows the presence of Flavonoids from the leaves of Talong-talongan.

Semi-purified Flavonoid Extract by Fourier Transform Infrared Spectroscopy (FTIR)

The Absorption of Aromatic ring is C-C=Aromatic Ring stretch is between the wavenumber of 1000-1400 cm^{-1}. The C-O bond had a weak absorption of the wavenumber of 900-1100 cm^{-1} because it was bonded to an aromatic ring and the CH_2 stretch was below the wavenumber of 3000 cm^{-1} there was a weak absorption noticed.

Table 5: FTIR Spectra Interpretation.

Absorption Location (cm-1)	Functional Groups in Flavonoids	Absorption Intensity	Remarks
3200- 3000	sp2 C-H bond (-C-H)	Variable and sharp, may be doublet	Absent
3000 – 2830	sp3 C-H bond (-C-H)	Strong doublet	Present
1900 – 1600	Aromatic	Several weak overtones	Absent
1725 – 1680	Ketone (C=O)	Very strong, sharp	Present
1620 – 1500	Olefin (C=C)	Weak to medium, must be confirmed by the presence of the sp2 C-H bond	Absent

Table 5 shows that the Talong-talongan Extract has sp3 C-H bond (-C-H), and Ketone (C=O) functional group that indicate the presence of flavonoids.

1. Biological Test Result

Table 6: Results of Uric Acid Level in Day 1 the First day of Dose Treatment.

Treatment group		Baseline	Uric Acid Level after
Negative	Mean N Standard Deviation	4.64 5 0.24	4.92 5 0.61
Positive Control	Mean	8.8	3.36

	N	5	5
	Standard Deviation	2.81	0.44
100mg	Mean	7.7	3.48
	N	5	5
	Standard Deviation	2.27	1.29
200mg	Mean	4.24	3
	N	5	5
	Standard Deviation	0.48	0
300mg	Mean	6	3.48
	N	5	5
	Standard Deviation	2.40	0.48

Table 6 shows that there was an increased of uric acid level in negative control and decreased of uric acid level in the positive control and flavonoid extract at 100mg, 200mg and 300mg/mL concentration.

Table 7: Results of Uric Acid Level in Day 2 the Second day of Dose Treatment.

Treatment group		Baseline	Uric Acid Level after
Negative	Mean	5.56	6.7
	N	5	5
	Standard Deviation	0.87	2.53
Positive Control	Mean	7.6	3.36
	N	5	5
	Standard Deviation	4.04	0.44
100mg	Mean	7.5	1.5
	N	5	5
	Standard Deviation	2.78	1.87
200mg	Mean	4.26	3.18
	N	5	5
	Standard Deviation	1	0.22
300mg	Mean	4.92	3.3
	N	5	5
	Standard Deviation	0.61	0.37

Table 7: shows that there was an decreased of uric acid level in the positive control and flavonoid extract at 100mg, 200mg and 300mg/mL concentration.

Treament Group	X	SD	F-value	Significance	Remarks
Negative Control	4.92	0.61	14.13	P=0.05 <0.05 Significant	Negative vs Positive
Positive Control	3.36	0.44	14.13	P=0.05 >0.05 Not Significant	Positive vs 100mg/kg
Talong-talongan Extract (100mg)	3.48	1.29	14.13	P=0.05 >0.05 Not Significant	100mg/kg vs 200mg/kg
Talong-talongan Extract (200mg)	3	0	14.13	P=0.05 >0.05 Not Significant	200mg/kg vs 300mg/kg
Talong-talongan Extract (300mg)	3.48	0.48	14.13	P=0.05 <0.05 Significant	Negative vs 300mg/kg

Table 8 Comparison of Uric Acid Level after First Day Treatment of Talong-talongan Extract and the Positive Control

Table 8 shows that there is a significant difference result in uric acid level after the first day treatment of the test sample between positive control and Talong-talongan extract at 100mg, 200mg and 300mg, whereas the Talong-talongan extract at 300mg shows comparable to positive control after the first trial which means that the Talong-talongan extract exhibit potential in lowering uric acid.

Table 9: Comparison of Uric Acid Level after Second Day Treatment of Talong-talongan Extract and the Positive Control.

Treament Group	X	SD	F-value	Significance	Remarks
Negative Control	6.7	2.53	14.13	P=0.05 <0.05 Significant	Negative vs Positive
Positive Control	3.36	0.44	14.13	P=0.05 >0.05 Not Significant	Positive vs 100mg/kg
Talong-talongan Extract (100mg)	1.5	1.87	14.13	P=0.05 >0.05 Not Significant	100mg/kg vs 200mg/kg
Talong-talongan Extract (200mg)	3.18	0.22	14.13	P=0.05 >0.05 Not Significant	200mg/kg vs 300mg/kg
Talong-talongan Extract (300mg)	3.3	0.37	14.13	P=0.05 <0.05 Significant	Negative vs 300mg/kg

Table 9 shows that there is a significant difference result in uric acid level after the second day treatment of the test sample between positive control and Talong-talongan extract at 100mg, 200mg and 300mg, whereas the Talong-talongan extract at 300mg shows comparable to positive control after the first trial which means that the Talong-talongan extract exhibit potential in lowering uric acid.

Review of related literature and studies

Talong-talongan is found occurring in open, waste place at low and medium altitudes in most islands and provinces. The forms with white flowers are perhaps typical. It is now pantropic in distribution. This weed is a coarse, erect, branched, half-woody herb 1 to 3 meters in height. The branches are covered with short, scattered spines and in most parts with stellate hairs. The leaves are ovate to oblong ovate and 10 to 20 centimeters long, with sinuate-lobed margins, and pointed tip. The flowers are white, many, about 1 centimeter long, and borne on lateral and usually extra-axillary inflorescences. The fruit is yellow, smooth, rounded, and about 1 centimeter in diameter.

The family Solanaceae represents one of the most economically and medicinally important families of angiosperm. *Solanum torvum Linn* is a small solanaceous shrub, widely distributed in the tropical and sub-tropical areas, with a small number in temperate areas (Jennifer et. al., 1997) widely in Pakistan, India, Malaya, China, Philippines, and tropical America (Nasir, 1985). Among the major chemical constituents of *S. torvum* steroids, steroids saponin, steroid alkaloids, and phenols. Pharmacologically studies indicate that the stem and roots of *S. torvum* have anti-tumor, anti-bacterial, anti-viral, anti-inflammatory, and other medicinally important effect. This plant species is very good source of alkaloids, flavonoids, saponins, tannins, and glycosides (Zubaida et al., 2013)

The pharmacological property of *Solanum torvum* has both a sedative and a diuretic therapeutic effect. The compounds isolated from the fruits of *Solanum torvum* is antiviral isoflavonoids sulfate and steroidal glycoside. Arthan et. al., 2002 has investigated methyl alcohol extracts of fruits and found new isoflavonoids sulfate named tovanol A, and a new steroidal glycoside torvoside A. In the phytochemical studies that the fruit of this specie have a good concentration of various alkaloid, flavonoid, tannins, and glycosides as sufficient to have pharmacological effect. Therefore, it is not only used for nutritive purposes but are effective for cough ailments and are considered to be effective medicine in cases of liver and spleen enlargement. The ripened fruits are used in the preparation of tonic haemopoetic agent and also for treatment for pain, according to Kala 2005.

According to Abas et al., 2006 *Solanum torvum* exhibit some anti-oxidant activity and DNA-Repair capability in oxidative DNA damage caused by free radicals. And according to Chah et al 2000 the methanolic extract of fruits and leaves were reported to have antimicrobial activities against human and clinical isolates. Recently, a novel protein was isolated from water extract of seed that has been proved to be an effective anti-oxidant. Sicapriya et al 2007 concluded that these proteins are effective even at low doses when compared to well-known standard synthetic anti-oxidant.

Flavonoids have been reported to have multiple biological including anti-inflammatory, antibacterial, anti-viral, anti-allergies and also can be utilized for treatment protocols for cardiovascular disease, asthma, cancer, liver disease, periodontal disease, macular degeneration and cataracts (Arreola et al., 2012).

The contents of the flavonoid extracted for 2 hours reached its maxima. Furthermore a decrease in the flavonoids content was noticed for 3hrs extraction and a sudden increase in their content was observed for 4hrs extraction time. This increase in the flavonoids content may be due to the synergistic effect of other parameters involved. A decrease in the flavonoids content was notice furthermore, i.e beyond 75%. Considering one of the aims of this work is to propose a suitable solvent for extracting the raw flavonoids.

In Yucatan, according to Martinez, Filipinos are the people who lived in the South East Asia, the Philippines. Prasad and Krishnan reviewed different studies regarding the hyperuricemia of the Filipinos within or outside the country. 2.5% of incidence of gout among Filipinos in contrast to 0.13% incidence in non-Filipino group was observed in the Kind Country Hospital in Honolulu, Hawaii. Different factors for the disparity of the level of

hyperuricemia of the Filipinos in the U.S. compared to Filipinos in the Philippines. In the study of Bayani Siason et. al., hyperuricemia is more common in Filipinos in the US than in the Philippines because Filipino immigrants to the U.S. are largely from the Ilocano region of the Philippines, where hyperuricemia is more widespread but in other conducted by the same author it was observed that there is no significant difference in mean serum urate levels between Ilocano and non-Ilocano's, negating the theory of selective migration and was also supported by the study of Healey et. al., Maladaptation of renal function, lifestyle such as diet, adiposity and exercise, health care utilization and diagnosis, genetics, and comorbidities also serves as an important factors in the difference of uric-acid levels of the Filipinos in the U.S. and in the Philippines. (Prasad, 2014).

According to the study Hyperuricemia and gout has been acknowledge among the Filipinos in other countries for almost two decades. Several studies have been reviewed. Gout is a disease that has become so prevalent in the Philippines that Filipinos tend to use it as butt of jokes. We often joke around about the victim's old age or his or her unhealthy eating habits. Gout is a form of rheumatic disease caused by deposits of uric acid crystals in tissues and fluids within the body. The awareness if these observations prompted an extension to the Filipinos in his natural environment. This is due to process caused by an overproduction or under excretion of uric acid. Aside from genetic factor, Filipinos hyperuricemia may become manifest because of environmental stress, including dietary stress, and among the most doctors advise patients to prevent gout is to avoid alcohol and soft drinks to prevent uric acid to build up. Foods that have purines must also be removed from patients' diet. Ultimately you can prevent gout by trying to lose weight. The clinical profile of gout as it exists in the Philippines has been compared and found to be similar generally to that of other series. The control of the hyperuricemia and gout has been satisfactorily accomplished in the Filipino patients with the long-term used of Allopurinol, and sometimes complemented with Colchicine taken daily.

Gout is more likely to affect men than woman, especially men between the ages of 40 to 50. Women become more susceptible to gout after menopause. Gout appears to run in some families. A number of factors may cause the body to produce too much uric acid or deter the kidneys from eliminating enough of it. Some factors include excess weight, certain medical conditions (high blood pressure, diabetes, and elevated fat levels) excessive alcohol intake, excessive intake of foods high in purine, certain medications (diuretics, low-dose aspirin, niacin, and the organ transplant anti-rejection drug, cyclosporine), surgery, and severe illness or injury.

Typically, uric acid is dissolved in the blood as urate then passes on through the kidneys and is the eliminated in the urine. Sometimes, blood uric acid levels may be elevated (a condition known as hyperuricemia) because the body either produces too much uric acid or the kidneys don't eliminate enough.

High levels of uric acid in the blood, or other triggers, can cause urate crystals to from in joint spaces and other tissues. The resulting inflammation, which is due to one's white blood cells response to these crystals, is a gout attack. In some people, the crystals can form kidney stones (calculi).

Gout is a kind of arthritis caused by a buildup of uric acid crystals in the joints. Uric acid is a breakdown product of the purines that are part of many foods we eat and drink. . An abnormality in handling uric acid and crystallization of these compounds in joints can cause attacks of painful arthritis, kidney stones, and blockage of the kidney filtering tubules with uric acid crystals, leading to kidney failure. Gout is one of the most frequently recorded medical illnesses throughout the world and history. The prevalence of gout in the U.S. has risen over the last twenty years and now affects 8.3 million (4%) Americans. Gout is more common in men than in women and more prevalent to African-American men than to white American men. The chances of having gout or hyperuricemia rises with age, with a peak age of 75 to all people. In women, gout attacks usually occur after menopause. Among the United States population, about 21% have elevated the blood uric acid levels, a condition known as hyperuricemia. However, only a small portion of those with hyperuricemia or high uric acid will actually develop gout. If your parents have gout, then you have a 20% chance of developing and getting it. Acute gout attacks can also be characterized by a rapid onset of a pain in the affected joint or in the big toe followed by a warmth, swelling, reddish discoloration, and marked tenderness. The small joint at the base of the big toe is the most common site for an attack and it usually starts there. Other joints that can also be affected include the ankles, knees, wrists, fingers, and elbows. In some people, the acute pain is so intense to other people that even a bed sheet touching the toe causes severe pain. These painful attacks are usually subside or be lessen in hours to days, with or without taking any medication. In rare instances, an attack can last for weeks. Most people with suffering gout will experience repeated bouts over the years. Obesity, excessive in weight gain, especially in young people, moderate to heavy alcohol intake, high blood pressure, and abnormal kidney function are among the risk factors for developing the gout. Certain drugs and diseases can also cause elevated levels of uric acid or hyperuricemia. Also, there is an increased prevalence of abnormally low thyroid hormone levels (hypothyroidism) in patients with gout. (Webmd.com Feb. 2016)

According to Zubaida et al., 2013 in Journal of Applied Pharmaceutical Science 3 there are a lot of traditional uses of *Solanum torvum*, the leaves after drying in shade it is powdered mixed with hot water or cow milk and administered orally in relief from colds and coughs it used in the community in Puducherry, Karaikal, Mahe and

Yanam, India. The extracted lead juice is taken orally to reduce body heat. In kurichyas, Kannur District the roots of *Solanum torvum* is externally applied on cracks to cure cracked foot. The fruit of it is used in India is cooked as an important ingredients of soups and sauces, and in Mexico, the aqueous extract of fruits is known lethal to mice. In Indonesia the whole plant is use to control bacterial and fungal disease of *S. melongena*. The roots and leaves in Garo tribe Bangladesh is used for asthma, diabetes and hypertension. While the roots off it is used in Northeastern Brazil is juiced to treat liver disease, tuberculosis and as anti-emetic.

Solanum torvum has an effect on systolic blood pressure according to Mohan et. al., 2009 Its ethonolic extract acts by preventing and reversing the development of hyperinsulinemia to control the rise of systolic blood pressure. They investigated its effect on blood pressure and metabolic alteration in fructose hypertension condition generated in rats.

Flavonoid represents a highly diverse class of secondary metabolites with potential beneficial human effect that is widely distributed in the plant kingdom and currently consumed in large amounts in the diets. Flavonoids display several pharmacological properties in gastro protective area, as acting as anti-secretory, cytoprotective and antioxidant agent.

As it stated in the study entitled "Optimization of flavonoids extraction from the leaves of Tabernaemontana heyneana" the main factors that affect the extraction of flavonoids are temperature, extraction time, materials ratio (weight of the leaves; volume of extracting agent) extracting agent (%) and the number of extraction were studied individually.

In the study of K. P. Sampath Kumar et al, studies have shown that quercetin protects against cataracts, cardiovascular disease, and cancer. The researcher mentioned that it also possess anthelmintic, anti-inflammatory, antiseptic, antispasmodic, carminative, diuretics, expectorant, febrifuge, hypoglycemic, hypotensive, lithontripic, stomachic and tonic activity.

In the study "The Isolation and Determination of the Antioxidant Property of the Flavonoid Extract of Asparagus (*Asparagus officinalis)* family Liliaceae" by Caballes, et. Al., it stated that after the Asparagus sample was subjected to phytochemical screening which yielded the presence of flavonoids, carbohydrates, volatile oils, protein, leucoanthocyanins, condenses tannins, saponins and alkaloids. Flavonoids were obtained by macerating 400 grams of fresh Asparagus shoots for 48 hours in hot methanol. Then, the residue was re-extracted with ether, evaporated in incipient dryness then with ethyl acetate and likewise evaporated to incipient dryness to obtain flavonoid extrat was used in the determination of it antioxidant activity by using DPPH 91, 1-diphenyl 1-2pictyl hydrozyl) method. Six concentration (1, 5, 10, 50, 100 ug/mL) of both sample taken (Flavonoid extract from Asparagus shoots) and the standard (Ascorbic Acid) were taken. Using the DPPH method, the researchers concluded that 500 ug/mL has the highest percentage inhibition of free radical DPPH, wherein as the concentration increases, the percent inhibition also increases. (Caballes, et. al., 2012)

This review provides an updated and extensive overview of methods and their applications in natural product matrices and sample biological origin. In addition, it critically appraises recent development and trends and provides selected representative bibliographic examples. Among various solvents, ethanol was selected as a right choice because it is environmentally benign and relatively safe to human health. Ethanol reacts through non-covalent interactions and promotes a rapid diffusion into the solution. Various concentration of ethanol used exhibited different effect in changing the fluid polarity and thus had diverse effect on the solubility enhancement of the sample. The optimal extraction yield may be fulfilled when the polarity on the fluid and the constituent to be extracted. The results of the study indicated that the optimal ethanol concentration for extraction of the flavonoid was found to be 75% (Satkishkumar, 2008).

Allopurinol is used to treat gout and certain types of kidney stones. It is also used to prevent increased uric acid levels in patients receiving cancer chemotherapy. These patients can have increased uric acid levels due to release of uric acid from the dying cancer cells. Allopurinol works by reducing the amount of uric acid made by the body. Increased uric acid levels can cause gout and kidney problems. (webmd.com)

Allopurinol reduces the production of uric acid in your body. Uric acid buildup can lead to gout or kidney stones. Allopurinol is used to treat gout or kidney stones, and to decrease levels of uric acid in people who are receiving cancer treatment. Allopurinol may also be used for purposes not listed in this medication guide. (www.drugs.com)

In the study conducted by Aala. Emma et. al., "A comparative stability of Allopurinol tablets", confirming the shelf-life stability safety and effectiveness of Allopurinol tablets namely Llanol, Zyloprim, and Purinase based on expiry dates provided was cited. Samples which contained Allopurinol were analyzed by spectrophotometric method under ultraviolet region. To confirm that the said Allopurinol was really present, identification test was performed. Based on the investigation made on the results obtained, all the results were within USP requirement which Allopurinol tablet contains NLT 93% and NMT 107% of the labelled amount per tablet (Aala. 1990).

According to an article published in Phil. Care, gout has become one of the most common diseases in the Philippines but Filipinos don't take it seriously. However, gout can be more serious that what one would

like to think. As reported to the Center for Disease Control, gout may render people immobilized for days or even weeks. In some situations, gout may also be an indication of increase risk of kidney stones.

An article in the Philippines Star entitled "About Gout" by Tyrone M. Reyes M.D. last December 2011 explained the nature of gout, the people at risk, its symptoms, and complications, diagnosis and prevention, and lastly its treatment.

Gout is a joint disorder characterized by acute inflammation commonly affecting the big toe. The pain and swelling associated with gout are result of the body's inflammatory responses to the accumulation of urate crystals in or around the joint. Uric acid is formed when the body breaks down molecules called purines, which are building blocks to the DNA. Purines are found in high quantities in certain foods, including anchovies, sardines, and organ meats, such as liver. But most if the uric acid produced in the body is a result of a breakdown of naturally occurring purines found in one's system.

In the study of Dela Cruz, et al., who investigated the antioxidant content and activities in a variety of a locally grown guava entitled "The Antioxidant property of flavonoids in methanolic extract from unripe guava peel (*Psidium guavaja L.*) Family Myrtaceae" the sample was prepared throughout maceration method in 80% ethyl alcohol and extract was collected and evaporated in the rotary evaporator.

In the study "The Isolation and Determination of the Antioxidant Property of the Flavonoid Extract of Asparagus (*Asparagus officinalis)* family Liliaceae" by Caballes, et al., it stated that after the Asparagus sample was subjected to phytochemical screening which yielded the presence of flavonoids, carbohydrates, volatile oils, protein, leucoanthocyanins, condenses tannins, saponins and alkaloids. Flavonoids were obtained by macerating 400 grams of fresh Asparagus shoots for 48 hours in hot methanol. Then, the residue was re-extracted with ether, evaporated in incipient dryness then with ethyl acetate and likewise evaporated to incipient dryness to obtain flavonoid extrat was used in the determination of it antioxidant activity by using DPPH 91, 1-diphenyl 1-2pictyl hydrozyl) method. Six concentration (1, 5, 10, 50, 100 ug/mL) of both sample taken (Flavonoid extract from Asparagus shoots) and the standard (Ascorbic Acid) were taken. Using the DPPH method, the researchers concluded that 500 ug/mL has the highest percentage inhibition of free radical DPPH, wherein as the concentration increases, the percent inhibition also increases. (Caballes, et al., 2012)

In a study was made at the University of the Philippines the mice used in the assay were 6-8 weeks old, Swiss Albino mice (ICR stain) purchased from the Food and Drug Administration (FDA) Philippines, Department of Health, Alabang, Muntinlupa City. The animals were acclimated for at least one week in standard cages. The mice were fed with commercial pellets with free access to purified drinking water and libitum, standard condition of 12 hours light and 12 hours dark cycle, and temperature (23˚C - 25˚C). The protocol used for the anti-ulcerogenic assay was approved by the college of Science Animal Care and Use Committee (CSACUC) of the University of the Philippines Diliman with assigned protocol number IC 2011-06.

SUMMARY OF FINDINGS

Comprehensive presentations of data gathered from the study conducted.

1. The crude extract of *Solanum torvum* was found to contain Flavonoids.
2. The computed percentage yield of flavonoids in 100 grams of Talong-talongan was 16.11%
3. The semi-purified flavonoid extract obtained was viscous dark-green with characteristic odor.
4. It was soluble in solvents such as Water, and 80% Ethyl alcohol.
5. Intrumental Analysis for the Classification of Flavonoids; Ketone and sp^3 C-H bond (-C-H) was found present in Talong-talongan. Occurance of ketone and sp^3 C-H bond (-C-H) could not lead to presence of flavonoids, hence it may be concluded that Talong-talongan may *not* contain flavonoids.
6. Allopurinol and the doses of the semi purified flavanoid of 100 mg/mg 200 mg/kg and 300 mg/kg of the Talong-talongan extract, decrease in the level of the blood uric acid level was obtained.

CONCLUSIONS

Therefore the researchers concluded that the flavonoid extract from the Talong-talongan *(Solanum Torvum Linn.* Family Solanaceae*)* leaves produce a hypouricemic property. The researchers also conclude that all doses and the positive control are equally effective. Also, the 200 mg/kg and 300 mg/kg dose can cause injury to the kidneys.

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REFERENCE

1. Agrawal, A., Bajpei, P., Patil A., Bavaskar, S. *Solanum torvum* Sw. – A Phytopharmacological review. Scholards Research Library. Sciences, 2010.
2. Burkill, and Haniff (*Solanum torvum* Linn. Family Solanaceae) Medicinal Plants of the Philippines, 2012.
3. Caballes, et. Al., The Isolation and Determination of the Antioxidant Property of the Flavonoid Extract of Asparagus (*Asparagus officinalis)* family Liliaceae. CEU Graduate School Library.
4. Jaiswal, B.S., *Solanum torvum;* Review of its traditional uses, Phytochemistry and Pharmacology. International Journal of Pharma and Bio, 2012.
5. Quisumbing, E. PhD. Medicinal Plants of the Philippines. Katha Publishing Co. Inc., 2010; 864.
6. Tang, E. Preparation of an Antimicrobial Ointment from the Semi purified Alkaloids of the leaves of Talong-talongan (*Solanum torvum* Sev., Family Solanacea). CEU Graduate School Library, 1997.
7. Yousaf, Z. Wang, Y., Baydoun, E. Phytochemistry and Pharmacological Studies on *Solanum torvum* Swartz, 2013.