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Preparation Of Tridax Procumbens Stem Based Chitosan Gel and Its Antioxidant Activity

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Abstract:

Introduction: *In many countries across Africa, T. procumben L. (Asteraceae) has long been used as a traditional medicine to treat bronchial severe stomach cramps, diarrhoea, dysentery, and liver disorders. For the purpose of removing periodontal pockets, chitosan has been tested in vivo using flap surgery and a shallower probing depth.*

Materials and methods:
T.procumbens stem based chitosan gel was prepared and its antioxidant activity was evaluated. The antioxidant activity was evaluated by using DPPH assay and Hydroxyl Radical Scavenging Assay. Different concentrations (10µL,20µL,30µL,40µL,50µL) of*T.procumbens* stem based chitosan gel were analysed for its antioxidant activity.

Results: The % inhibition of *T.procumbens* stem based chitosan gel was calculated and was found to be maximum at 50µL and lowest at 10µL concentration.

Conclusion: *T.procumbens* stem based chitosan gel can serve as an excellent antiinflammatory agent and can be used as an alternative to commercially available wound healing gel for oral applications

Keywords: antioxidants, anti-inflammatory agent, chitosan gel, *T.procumbens,*

Introduction:

In many countries in Africa, South and Southeast Asia, *T. procumben L.* (Asteraceae) has long been used as a traditional medicine to treat bronchial severe stomach cramps, diarrhoea, dysentery, and liver disorders. [1]It was once a common plant, despite allegations that it invades many crops as a weed.[2] Procumbetin, among other bioactive compounds, have been successfully isolated from this plant.
Puerarin, centaurein, and centaureidin, as well as 8,3′-dihydroxy-3,7,4′ trimethoxy-6-O-d-glucopyranosyl flavone and 6,8,3′ trihydroxy-3,7,4′ hydroxyflavone[3]. This plant has been shown to contain a number of lipid components, including lupeol, fucosterol, 30-methyl-28-oxodotriacont-29-en-l-oic acid, methyl,14-octadecanoate, and methyl,14-oxononanoate.[4] Additionally, phenolic acids from this plant have been found, including guaiacol, benzoic, vanilic, and benzeneacetic acids. T. procumbens performs a wide variety of biological processes[5].The ethyl acetate extract of this plant showed strong larvicidal and allelopathic effects. Methanol and ethanol extracts demonstrated anti-hyperglycemic, anti-fungal, anti-leishmanial, and hepatoprotective activities while ethyl acetate extract had anti-inflammatory, anti-cyclooxygenase, and antioxidant effects.[6] This herb's acetone extract exhibited anticoagulant, anti-herpetic, and antibacterial properties. It has been reported that the hydrophilic biopolymer chitosan, which is produced by alkaline deacetylation of chitin, a vital component of arthropod shells, functions as both a bioadhesive and a permeabilizer. Positive characteristics of chitosan include nontoxicity, biocompatibility, and biodegradability.[7] The antibacterial, wound healing, hemostatic, and tissue regeneration capabilities of chitosan itself have sparked a lot of attention in the dental industry.Chitosan has been researched for periodontal pocket removal using flap surgery in vivo and a reduction in probing depth.A member of the nitroimidazole group of drugs, metronidazole operates only on anaerobes and does not affect the commensal aerobic bacteria, hence resistance seldom arises.[8] It has been observed that the dose used in dentistry, both for local and systemic treatment, is extremely safe. When compared to systemic distribution, local delivery of metronidazole has been proven to be less dangerous (lower serum levels). For both its

above-mentioned bioactive capabilities and the local delivery of metronidazole, chitosan was employed in this investigation as a gel. In addition to scaling and root planing (SRP), the therapeutic efficacy of chitosan gel formulations in the presence or absence of metronidazole was assessed in CP patients.[9]People regularly use traditional medicine systems in underdeveloped and developing nations, yet little is known about the biological function and chemical make-up of medicinal plants. Various nations have employed herbal remedies to treat medical problems Medicinal plants displayed a range of behaviors, including antimicrobial properties. [10]Significant antibacterial and antifungal potential exists in T. procumben. This medicinal plant is found practically year-round in almost all regions of the country and is widely dispersed in Asia, Africa, and Australia. *T. procumbens* is a traditional remedy for typhoid fever, fever, cough, epilepsy, asthma, and diarrhea. Over the last few decades, both the use and popularity of traditional systems of medicine have grown significantly. More than 80% of medical demands are met by the traditional medical system.[11]

The search for naturally occurring antioxidants and antibacterial compounds for application in food or pharmaceutical products from *T. procumbens*, on the other hand, has also sparked an increase in interest. This plant's ethanol and methanol extracts have been found to have antioxidant properties.[10]

Materials and methods:

Preparation of chitosan gel

0.5 ml of chitosan was dissolved in 49 mL of distilled water plus 1mL of glacial acetic acid. Then, it was kept in a magnetic stirrer for 24h for a uniform mixture. *T. procumbens* extract was added and kept again in a magnetic stirrer for another 24h. Then each activity was done using this sample.

DPPH assay

DPPH assay was used to test the antioxidant activity of *T.procumbens* stem based chitosan gel at different concentrations (10µL,20µL,30µL,40µL,50µL). *T. procumbens* stem based extract was mixed with 1 mL of
0.1 mM DPPH in methanol and 450 µL of 50 mM Tris HCl buffer (pH 7.4) and incubated for 30 minutes. Later, the reduction in the quantity of DPPH free radicals was assessed dependent on the absorbance at 517 nm. Ascorbic acid was used as standard.

Hydroxyl Radical Scavenging Assay

All solutions were prepared freshly.1.0mL of the reaction mixture containing 100µL of 28mM of 2-deoxy-2-ribose (dissolved in phosphate buffer,pH 7.4), 500µL solution of various concentrations of the
T. procumbens stem based chitosan gel (10µL,20µL,30µL,40µL,50µL). 200µL of 200µM FeCl3 and 1.04mM EDTA (1:1 v/v),100µL H2O2 (1.0mM) and 100µL ascorbic acid(1.0mM).After an incubation period of 1 hour at 37°C the extent of deoxyribose degradation at about 532nm against the blank solution. Vitamin E was used as a positive control.

Results

The odd electron molecule in DPPH free radical gave a strong absorption at 517 nm where it turned yellow to brown colour.The % inhibition of *T.procumbens* stem based chitosan gel were calculated and was found to be maximum at 50µL for both DPPH assay and hydroxyl radical scavenging assay.

Figure 1:

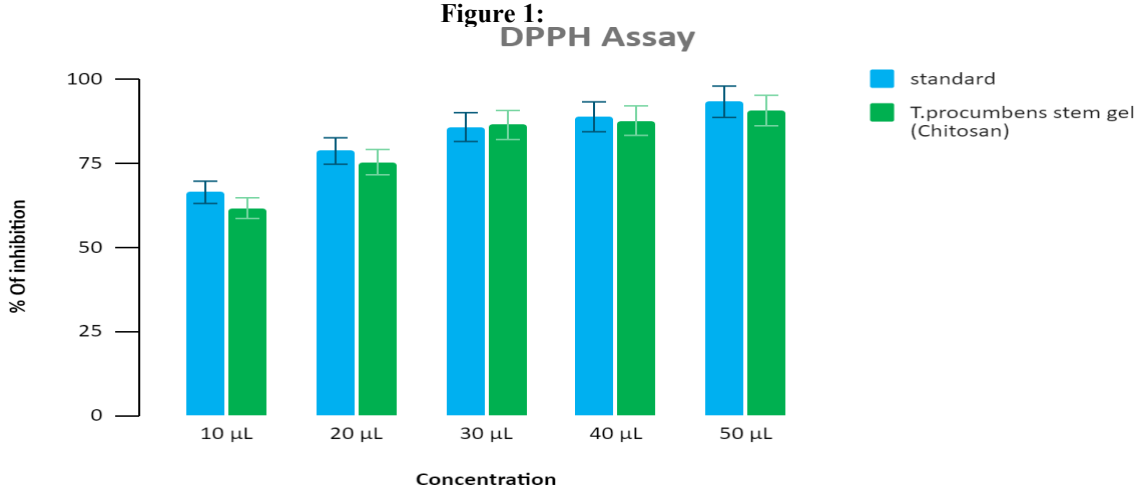

Figure 1 represents DPPH Assay results. *T.procumbens* stem based chitosen gel shows the same effect as the standard drug. At 10µL the effectiveness of standard drugs was higher than . But by increasing the concentration both the drugs had same effectiveness

Figure 2:

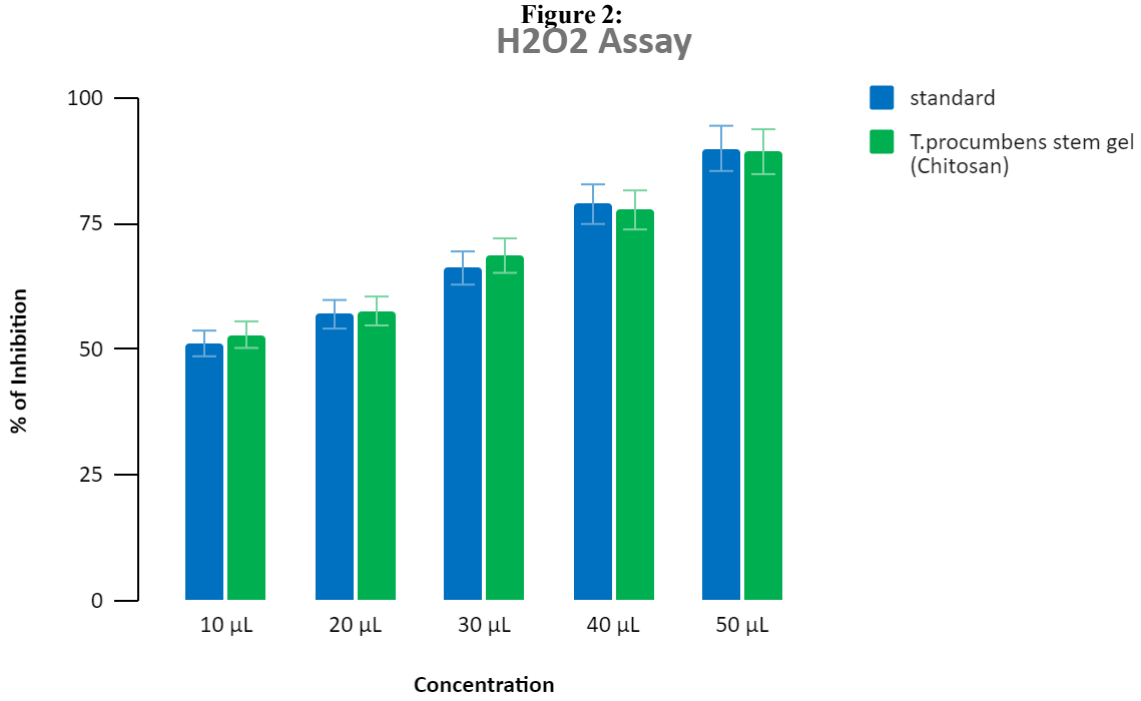

Figure 2 represents H2O2 Assay results. *T.procumbens* stem based chitosen gel shows same effect as the standard drug

Discussion:

One class of flavonoid chemicals derived from T. procumbens is called centaureidin .[4] R Nagao et al .'s revelation that several dietary flavonoids had inhibitory activity on XO is consistent with this study's findings[12]. In general, Lin et al[13]. revealed that hydrophobic interaction was critical in the binding of flavonoids to XO inhibition based on chemical structure and binding affinities of flavonoids serving as XO inhibitors. [14] Flavonoids' planar structure and double bond C2 = C3 are beneficial for their potential to reduce XO Due to the F45–47 fraction's strong inhibitory effects on XO in this work, it was also necessary to establish how fatty acids, glycerides, and flavonoids interact to cause XO inhibition.[15]

In other researches T. procumbens methanol extract was tested for its cytotoxicity against *T. procumben*s human lung cancer cells and breast cancer cell lines. [16]The MTT assay was run to examine the cytotoxic effect. In other T. procumben in vitro antioxidant activity using simultaneous DPPH, ABTS, FRAP, and TRP tests.[17] The results of this investigation showed that T. procumben extracts all possessed potent antioxidant properties against DPPH and ABTS free radicals as well as potent lowering properties against Fe 3+.[17,18] In other study plant leaf extract had more efficacy against breast cancer cell lines than human lung cancer cells. [19]84 2.8% cytotoxicity was demonstrated by a 250 g/ml plant extract against human lung cancer cells, while 68 3.1% cytotoxicity was observed in breast cancer cell lines.[20]

Our team has extensive knowledge and research experience that has translate into high quality publications [21–30] [31] [32–34][33,35]

Conclusion:

Many plants exhibit in vitro and an invivo antioxidant property owing to their phenolics, proteins, vitamin d and pectins contents.

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CONFLICT OF INTEREST:
There was no potential conflict of interest.

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