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## A Review on Biological and Chemical Properties of Cyperus Species.

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#### ABSTRACT

*Cyperus* species are monocotyledonous, perennial, potent, multipurpose medicinal plants which belongs to Cyperaceae family. These are commonly known as nagarmotha, nut grass, umbrella sedges and sweet sedges. The leaves, roots and tubers of these whole plant species are used in astringent, diuretic, diaphoretic, cordial, brodycardiac properties and used to treat more diseases therapeutically. The steam distillation of essential oil extracted from rhizomes of these species used in anti-inflammatory diseases. Our present study in this review encompasses on biological and chemical properties of *Cyperus* species. Complete information regarding these plant species has been collected from various books and journals since the last 30 years, internet databases *etc* were also searched. This compiled data reflects the therapeutic properties of these herbs. This review article helpful for researchers to focus on natural plant products to find out new therapeutically important compounds responsible for its claimed traditional importance. Several therapeutically important natural compounds have been isolated from these herbs and they can serve as very potent and reliable drug candidate for treatment of various disorders. These compounds have indentified for its effectiveness against number of activities in *in vitro* and *in vivo* mechanisms.

Keywords: Anti-inflammatory, Anti diuretic, Antinociceptive, Multi potential bioactivities.



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#### INTRODUCTION

*Cyperus* species belongs to family Cyperaceae, these are monocotyledonous graminoid flowering plants known as sedges, which superficially resemble grasses or rushes. They are popularly known as *Nagaramotha* is an important herb in the *Ayurveda* [1]. In *Ayurveda*, *Nagaramotha* is *tikta*, *katu*, *kashaya* and *sheetala*, pacified deranged *kapha*, beneficial in the treatment of fever caused by aggravated pitta, in diarrhea, anorexia thirst burning sensation and fatigue [2]. In Southern India, its essential oil is employed in the perfume industry and the nut grass is used in the formulation of hair and skin care products; it stimulates sebaceous glands near hair roots [3]. The dried tuberous roots of these herbs are used in traditional medicine [4]. Tubers are credited with astringent, diaphoretic, diuretic, desiccant, cordial, and stomachache properties [5]. In traditional medicine, the rhizomes of these plant species are used in the treatment of inflammation [6].

These are erect, perennial, grass like sedges, single-stemmed, graminoid, up to 3 ft. tall underground. Along with fibrous roots, there are many slender rhizomes which form a tuber at each end. The leaves become grass-like and the blades are light green, smooth, glossy, and glabrous in texture. These species has an extensive and complex system of fine, fibrous roots and scaly rhizomes with small hard, spherical tubers and basal bulbs (swelled rhizome tips which produce stems and leaves) attached. Two types of rhizomes are produced, short rhizomes from germinating tubers which end in basal bulbs, and long wiry rhizomes from basal bulbs, which can end in tubers or basal bulbs. Tubers are unevenly globose, 0.3-1.9 cm in diameter, black to brown, hard, smooth (scales shed with maturity), with buds at the apex only. The tubers taste mildly almond-like, with a nutty flavor. The inflorescence is terminal, umbellate, umbel simple to compound, loose, with 1-10 narrow, unequal rays 0-6 in. (0-15 cm) long, and with 2-6 subtending leaf-like, unequal bracts forming an involucre with the longest involucral bract much exceeding the umbel, and often wider than basal leaves. Sedges (Cyperus) have grass-like leaves, having no hairs. They can be distinguished from grasses, even while young, by their triangular stems and triangular inflorescence stems, three-ranked leaves and their leaves lacking ligules, auricles, and collar regions while the grass stems are flat or round. There are several native Cyperus species that have distinct similarities to Cyperus esculentus. Purple nutsedge (Cyperus rotundus), another weedy non-native sedge found in southern Arizona, is very similar to vellow nutsedge (Cyperus esculentus). These two nutsedges can be found growing on the same site. The two species are difficult to distinguish from each other before blooming. Purple nutsedge has purplish spikelets, and the tubers that form on the rhizomes are often multiple and form chains, while in C. esculentus there is one at the tip of the rhizome. The tubers are oblong and covered with persistent reddish scales in C. rotundus, while roundish and smooth on C. esculentus.

#### Compounds of Cyperus species

Cyperotundone and  $\alpha$ -cyperone compounds was separated from essential oil of *C.rotundus* rhizomes by high speed counter current chromatography separation with two phase solvent system composed of n-hexane-acetonitrile-acetone-water (7:6:0.5:1.5v/v) [7]. The steam distilled essential oil of *C.rotundus* contains cyperene, caryophyllene, cyperol, rotundiene and cyperene [8]. Two novel natural phenolic compounds  $1\alpha$ -methoxy- $3\beta$ -hydroxy- $4\alpha$ -(3,4-dihydroxyphenyl)-1,2,3,4-tetrahydronaphthalin (Methoxy cyperotundol) and  $1\alpha$ , $3\beta$ -dihydroxy-

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 $4\alpha$ -(3,4-dihydroxyphenyl)-1,2,3,4-tetrahydronaphthalin (cyperotundol) were isolated from rhizomes of *C.rotundus* [9]. Oxo-isolongifolene,  $\alpha$ -gurjunene, z-valerenyl acetate,  $\alpha$ -salinene compounds extracted from hydrodistillation of *C.rotundus* tubers [10]. The chemistry of volatile components of *C.rotundus* has been studied by three methods hydro distillation (HD), pressurized liquid extraction (PLE) and supercritical fluid extraction (SFE). For extraction of  $\alpha$ copaene, cyperene,  $\beta$ -selinene,  $\beta$ -cyperone and  $\alpha$ -cyperone were quantitatively estimated using  $\alpha$ -cyperone as standard [11]. Quercetin, kamperol, catechin and myricetin have reported as flavanoids in leaf and root and callus of C.rotundus. The maximum amount of total flavonoid in six weeks old callus tissue 1.96mg/g.d.w and minimum 0.28mg/g.d.w in two weeks old callus tissue. In vivo studies showed higher flavanoids content in leaf in free form 0.58mg/g.d.w. and bound form 0.48mg/g.d.w. when compared to root in free form 0.19mg/g.d.w and bound form 0.11 mg/g.d.w [12]. There is a report on isolation of sesquiterpenes  $\beta$ -selinene, isocurcumenol, nootkatone and aristolone and triterpene oleanolic acid from the ethylacetate fraction of the rhizomes of *C.rotundus* [13]. Bioactive natural sesquiterpenes cyperene 16.9%, caryophyllene oxide 8.9%,  $\alpha$ -longipinane 8.4% and  $\beta$ -selinene 6.6% were the major compounds have been isolated from dried tubers essential oil extract of C.rotundus by GC/MS analysis [14].  $\alpha$ cyperone, myrtenol, caryophyllene oxide,  $\beta$ -pinene were major chemical compounds in the essential oil of C.rotundus rhizomes were obtained by hydro distillation and analyzed these compounds by capillary GC and GC/MS [15]. The new terpenoid 2, 3-diacetoxy-19-hydroxy-urs-12-ene-24-o-β-D-xylopyranoside isolated from rhizomes of C.scariosus by using chemical and spectroscopic studies [16]. Physico chemical parameters such as loss on drying, crude fiber content, total ash, acid insoluble ash, water soluble ash, sulphated ash and successive extractives values were observed with solvents of petroleum ether, n-hexane, acetone, alcohol and aqueous these values used to detect adulteration from rhizome of *C.rotundus* [17].

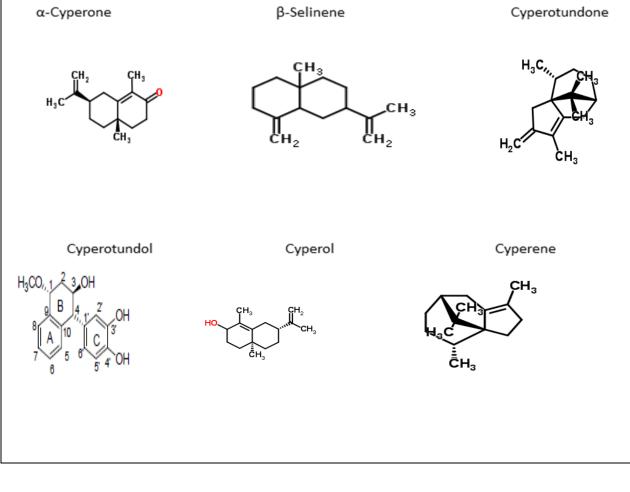
Plant	Extract	Source	Bioactivity	References
C.rotundus	Methanol	Rhizome	Antioxidant activity	[23]
C.rotundus	Hexane	Rhizome	Mosquito repellency activity	[40]
C.rotundus	Ethanol	Rhizome	Antioxidant activity	[22]
C.rotundus	Ethanol	Aerial parts	Antioxidant and Freeradicle scavenging	[20]
C.rotundus	Aqueous methanol & Aqueous ethanol	Stem, leaf and roots	Antioxidant activity	[19]
C.rotundus	Ethanol	rhizomes	Lipid lowering activity	[18]
C.rotundus	Aqueous	Tuber	Antiobesity potential	[36]
C.rotundus	Methanol	Rhizomes	Anti-inflammatory	[32]
C.rotundus	Ethanol	Rhizomes	Antimicrobial	[25]
C.rotundus&C.esc ulentus	Ethanol	Rhizomes	Anti-inflammatory, Antiarthritic, analgesic and anticonvulsant activities	[31]
C.rotundus	Ether & ethanol	Rhizomes	Anti-inflammatory	[29]
C.scariosus	Aqueous & chloroform	Rhizomes	Inhibits T cell response in Balb/C mice.	[35]

Table 1: The biological activities of the extracts	s of Cyperus species are listed as follows.
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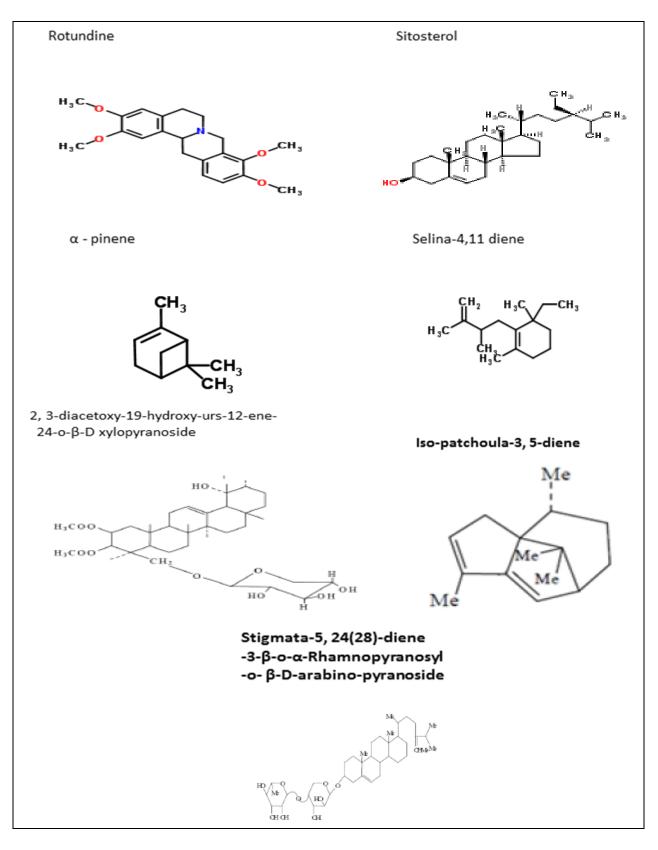


C.scariosus	Methanol	Leaves	Antinociceptive& Antihyperglycemic activity	[34]
C.scariosus	n-hexane	Roots & aerial parts	Antidepressant activity in mice.	[39]
C.rotundus	Petroleum ether, chloroform, ethanol & water	Rhizomes	Antimicrobial activity	[27]
C.rotundus	Ethyl acetate & methanol	Aerial parts	Pharmacological, antioxidant,genotoxic studies & rat splenocyte modulation function.	[33]
C.rotundus	Ethylacetate	Rhizomes	Hepatoprotective activity against carbon tetrachloride induced hepatotoxicity.	[37]
C.esculentus,C.rot undus, C.papyrus	Ethanol	Rhizomes	Antioxidant and Cytoprotective activity.	[21]
C.rotundus	Ethanol & aqueous	Rhizomes	Antimicrobial activity	[10],[26].
C.rotundus	Ethanol	Rhizomes	Wound healing activity	[43]
C.rotundus	Aqueous	Rhizomes	Infectious diarrhea	[42]
C.rotundus	Ethanol	Rhizomes	Age associated changes in glucose and lipids	[41]



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Total ash	12.87
Acid insoluble ash	4.56
Water soluble ash	6.4
Sulphated ash	10.22

#### Table 2: Physicochemical parameters of C.rotundus Linn Rhizome-Ash values.

#### Hypolipidaemia

The alcoholic extract of *C. rotundus* rhizomes used to detect hypolipidaemic activity on high fat diet induced hyperlipidaemic rats of wistar strain. Treatment with the standard drugs simvastatin and fenofibrate and different doses of extract exerted statistically significant (p<0.05) reduction in serum total cholesterol, LDL, TG, HDL levels at the end of 15 days of observation [18].

#### Antioxidant activity

Natural antioxidants either in the form of raw extracts or their chemical constituents are very effective to prevent the destructive processes caused by oxidative stress. Antioxidants stabilize or deactivate free radicals, often before they attack targets in biological cells. The role of free radical reactions in disease pathology is well established and is known to be involved in many acute and chronic disorders in human beings, such as diabetes, atherosclerosis, aging, immunosuppression and neurodegeneration.

An imbalance between ROS and the inherent antioxidant capacity of the body, directed the use of dietary and /or medicinal supplements particularly during the disease attack. Studies on herbal plants, vegetables, and fruits have indicated the presence of antioxidants such as phenolics, flavonoids, tannins, and proanthocyanidins. The antioxidant contents of medicinal plants may contribute to the protection they offer from disease. The antioxidant activity from flavanoids in different plant parts of *C.rotundus* by using methanol and ethanol solvents extraction. Inhibition of linoleic acid percentage in methanol extract (51.50-61.73%) was higher as compared to ethanol extracts (38.37-47.86%) and also reducing power in methanol extract (0.754-1.112) were also higher as compared to ethanol extracts (0.711-0.837) at concentration of 2.5-10.0 mg/mL [19]. The hydro alcoholic extract of *C.rotundus* exhibited high reduction capability and powerful free radical scavenging against DPPH and superoxide anions as well as a moderate effect on nitrous oxide. It will also inhibit lipid peroxidation in rat liver homogenate induced by Fe<sup>2</sup>+ or ascorbic acid and prevented deoxyribose degradation in both non site specific and site specific assays [20]. Various extracts of C.esculentus, C.rotundus and C.papyrus showed antioxidant and cytoprotective properties in two chemical assays FRAP and DPPH and a cell based bioassay in hepa1c1c7 cells using t-butyl hydroperoxide as the inducer of cytotoxicity. EtOH extract of *C.papyrus* was the most potent with direct antioxidant activity in both DPPH (EC<sub>50</sub>:5:1µg/ml and FRAP (FE: 48.7 µg/ml) assays and a significant full cytoprotection against TBHP at 100 µg/ml [21]. C.rotundus rhizomes ethanolic exhibits its scavenging effect in concentration dependent manner on superoxide anion radicals, hydroxyl radicals, nitric oxide radical, hydrogen peroxide and it had a property of metal chelating and reducing power. This



extract also studied for lipid peroxidation assay by thiobarbituric acid reactive substances (TBARS) using young and aged rat brain mitochondria. It showed effective response in preventing mitochondrial lipid peroxidation induced by Feso<sub>4</sub>/ ascorbate in concentration dependent manner [22]. Methanolic extract of *C.rotundus* reduce the most Fe<sup>3+</sup> ions, which had a lesser reductive activity than the standard of ascorbic acid. The total antioxidant capacity of *C.rotundus* was found to be 985.6mgGAE/gm extract [23].

#### *Cyperus* species and their role in Antimicrobial activity

Plants are important resource to combact serious diseases in the world. The traditional medicinal plants still play an important role to cover the basic health needs in the developing countries. In the recent years, infections have increased to a great extent and antibiotics resistant effects become an ever increasing therapeutic problem. In the past few years use of plant derived drugs have been reported to be safe and without side effects. Essential oils are chiefly used for the flavors and fragrances but they also possess antibacterial, antifungal, antiviral and antioxidant properties. The antibacterial activity of essential oil of aromatic plants against staphylococcus aureus, staphylococcus epidermidis, staphylococcus hominis, pseudomonas aeroginosa, Klebsiella pneumonia and proteus vulgaris [24]. In vitro antimicrobial activities of the aqueous and ethanolic extracts of Launaea procumbens (Labiateae), Vitis vinifera L. (vitaceae) and C.rotundus L (Cyperaceae) against a number of bacteria Alcaligens faecalis, Bacillus cereus, Bacillus subtilis, Enterobacter aerogenes, Eschericia coli, Klebsiella pneumonia, proteus mirabilis, proteus vulgaris, pseudomonas aeroginosa, pseudomonas alcaligenes, Salmonella typhimurium, Staphylococcus aureus and Candida tropicalis was performed by agar disc diffusion and agar well diffusion method. The ethanolic extracts of all the plants were active against all bacterial strains, while all the aqueous extracts were inactive except for Vitis vinifera. Among all the extracts S.typhimurium was the most resistant bacteria [25]. The steam distilled essential oil of *C.rotundus* contains cyperene, caryophyllene, cyperol, rotundiene and cyperene compounds showed antibacterial activity in various microorganisms' staphylococcus aureus, Klebsiella pneumonia, proteus vulgaris, streptococcus pyogenes, Escherichia coli and pseudomonas aeroginosa using inhibition zone method. It shows a remarkable activity against gram-positive bacteria, less antibacterial activity was found against gram negative bacteria and no activity was observed with the oil against pseudomonas aeroginosa and proteus vulgaris [26]. Oxo-isolongifolene,  $\alpha$ -gurjunene, z-valerenyl acetate,  $\alpha$ salinene compounds extracted from hydrodistillation of *C.rotundus* tubers. These compounds showed positive effect against Bacillus subtilis, Escherichia coli, pseudomonas aeroginosa and Staphylococcus aureus, Candida parapsilosis, Aspergillus flavus, Aspergillus fumigates and Fusarium oxysporum in different concentrations. At low concentration the oil was effective against S.aureus. Oil shown good antifungal activity against Candida parapsilosis and Aspergillus fumigates. The oil inhibits spore formation of Fusarium oxysporum and Aspergillus flavus [10]. The antimicrobial activity of ethanolic extracts of C.rotundus against Staphylococcus epidermidis, Bacillus cereus, Pseudomonas aeroginosa, Escherichia coli, Aspergillus niger and Candida albicans was most effective antimicrobial agent as compared to other extracts. B. cereus was the most susceptible gram positive bacteria followed by S.epidermidis, whereas



*E.coli* was the most resistant gram negative bacteria against all extracts. None of the strains exhibited antifungal activity against *Aspergillus niger* and *Candida albicans* [27].

#### Cyperus species importance in Inflammation

Inflammation is a disorder involving localized increase in the number of leucocytes and a variety of complex mediator molecules. Inflammation has shown to associate with numerous environmental and genetic factors. Environmental factors include allergens, infectious agents, toxins and chemicals. Whereas, genetic factors include prostaglandins, cyclooxygenases (COX), interleukins, cytokines, tumor necrosis factor alpha and interferon-gamma. Among those, some COXs have shown to play a major role in triggering the inflammation caused by both genetic and environmental factors. COX are two distinct isoforms, such as COX-1 and COX-2, they have shown to play a vital role in conversion of arachidonic acid to prostaglandins. Generally the expression levels of COX-2 in normal tissues are below the level of detection, but enhanced expression of COX-2 was detected by proinflammatory cytokines, growth factors and exposure of several carcinogens. Therefore, regulation of COX-2 is very important for therapeutic approaches against inflammatory associated disorder.

The anti-inflammatory activity of various *Cyperus rotundus* extracts was determined by using carrageenan induced rat paw edema assay [28]. 0.1ml of 1% carrageenan in saline was injected into the sub plantar region of the left hind paw of each rat to induce edema. The paw volume was measured initially at the intervals of 15, 30, 60,120 and 180 minutes. When compared to indomethacin (standard anti-inflammatory drug) paw edema % of inhibition 67.57% against test aqueous CR extract 200mg/kg=60% and 400 mg/kg=61.62% had shown partial inhibition of paw edema, whereas ether CR extract 200=61.62%,400mg/kg=63.78% which was shown alternatively good paw edema inhibitory action after 180min, it has standard deviation 0.67±0.01 when compared with indomethacin 0.60±0.02. The ethanolic extract of CR 200mg/kg=62.70%, 400mg/kg=65.40% this extract showed similar action as standard indomethacin and the standard deviation was 0.64±0.02. When compare to different solvent system the maximum % inhibitory action potential revealed by ethanolic extract at 400mg/kg dose against control and standard groups [29]. The anti-inflammatory potential of sesquiterpenes nootkatone,  $\alpha$ -cyperone, valencene and  $\beta$ -selinene isolated from C.rotundus was evaluated in lipolysaccharide (LPS)-stimulated RAW264.7cells, murine macrophages. Among the four sesquiterpenes,  $\alpha$ -cyperone and nootkatone showed stronger anti-inflammatory and a potent NF-kB inhibitory effect on LPS stimulated RAW264.7 cells. Molecular analysis revealed that various inflammatory enzymes (iNOS and COX-2) were reduced significantly and this correlated with down regulation of the NF-kB signaling pathway. Electrophoretic motility shift assays (EMSA) elucidated that nootkatone and  $\alpha$ -cyperone dramatically suppressed LPS –induced NF-kB-DNA binding activity using [32] p-labelled NF-kB probe these two compounds are potential therapeutic agents for inflammatory diseases [30]. The effect of Cyperus esculentus and Cyperus rotundus essential oils in anti-inflammatory (carrageenan induced), antiarthritic (formaldehyde induced), analgesic (formalin induced writhing) and anticonvulsant (MES produced convulsion). The results showed dose dependent activity, indicated by reduction in paw edema in anti-inflammatory and antiarthritic activity and significant reduction (p<0.01) in the maximal electroshock induced (MES) induced convulsion in comparision to Diclofenac sodium [31]. The anti-inflammatory activity of methanolic extract of C.rotundus rhizome on carrageenan induced paw edema in rats. The extract exhibited dose

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dependent anti-inflammatory activity at the dose of 1000mg/kg the methanolic extract showed maximum inhibition of the edema 57.5% as compared to standard drug indomethacin. This extract significantly inhibited p<0.01, p<0.05 carrageenan induced rat paw edema at 500mg/kg and 1000 mg/kg body weight [32].

#### Analgesic activity

The aqueous, ethyl acetate, methanol and total oligomer flavanoid (TOF) enriched extracts of *C.rotundus* were able to decrease the mouse ear edema induced by xylene, further these extracts reduce the number of abdominal contractions caused by acetic acid in mice, revealing the peripheral analgesic activity of these extracts. It is worth nothing that mice treated with doses upto 300mg/kg b.w. of *C.rotundus* extracts did not exhibit any toxicity. These extracts significantly enhance lymphocyte proliferation at 1mg/ml [33].

#### Antinociceptive activity

The methanolic leaf extract of *Cyperus scariosus* activity was determined by using acetic acid induced gastric pain in mice and antihyperglycemic activity through glucose tolerance test using glucose loaded mice. The extract showed dose dependent significant pain inhibition compared to aspirin in acetic acid induced writhing assay. The maximum writhing inhibition 46.62% was found at a dose of 200mg/kg body weight which was less than that of the positive control aspirin 56.74%, when used at the same dose. Antihyperglycemic activity of the extract was also found to be significant in mice loaded with glucose at doses of 200 and 400 mg/kg body weight. Maximum tolerance 42.86% was showed at 400mg extract/kg body weight, which compared favourably with that of glibenclamide at 10mg/kg body weight 57.62% [34].

#### Hypersentivity

*C.scariosus* chloroform fraction inhibits T cell responses in Balb/c mice in both humoral and cell mediated immune responses on p.o administration significantly p<0.01 by suppressing primary 26.8% and secondary 29.7% antibody titres and also inhibited cell mediated delayed type hypersensitivity immune response 45.9% at 600mg/kg dose,phagocytosis both *in vitro* 37.4% and *ex vivo*37.8% and delayed the graft rejection time 45.8%,thus confirming marked immunosuppression. Chloroform fraction significantly p<0.01 suppressed CD8+/CD4+T cell surface markers (14.0/25.3%) and intra-cellular Th1 cytokines,viz IL-2(34.4%) and IFN- $\gamma$  (34.7%) compared to cyclosporine-A, a standard T cell inhibitor 53.6% which was given to Balb/c mice at 200 mg/kg dose. *C.scariosus* did not show significantly p<0.01 suppress Th2 (IL-4) system [35].

#### Anti-obesity

Overweight and obesity are the most common nutritional problems resulting mainly from an energy imbalance caused by an increased ratio of caloric intake to energy expenditure represent rapidly growing threats to the health of population worldwide. Obesity decreases the quality of life and life expectancy and is also a strong risk factor for diseases such as type 2



diabetes, heart disease, stroke, certain types of cancers, osteoarthritis and depression. A large section of world population relies on traditional remedies to treat various diseases. The aqueous tuber extract of *C.rotundus* (ATECR) in high fat cafeteria diet fed obese rats (HFCD). Administration of HFCD for 40 successive days to experimental rats the body weight, organ and fat pad weights, serum total cholesterol LDL cholesterol, VLDL cholesterol, triglycerides and glucose levels was significantly increased and HDL cholesterol decreased as compared to normal control animals fed with normal pellet chow. If treatment with ATECR showed a significant reduction in the body weight gain, organ weight of the liver, kidney spleen, weight of fat pads and the levels of serum triglycerides, total cholesterol, LDL cholesterol, VLDL cholesterol, VLDL cholesterol, and increase in HDL cholesterol in dose dependent manner. The levels of aspartate transaminase, alanine transaminase and alkaline phosphatase were elevated in serum of obese rats, also resumed to normal on treatment with different concentrations of ATECR [36].

#### Hepatoprotective activity

Ethyl acetate extract and two crude fractions, ether solvent and ethyl acetate of the rhizomes of *C.rotundus* were tested to hepatoprotective activity against carbon tetra chloride induced hepatotoxicity. The ethyl acetate extract at an oral dose of 100mg/kg exhibited a significant protective effect by lowering serum levels of glutamic oxaloacetic transaminase, glutamic pyruvic transaminase, alkaline phosphatase and total bilirubin [37].

#### **Cytotoxic activity**

The oils of *C.rotundus* and *C.alopecuroids* tubers rich in oxygenated sesquiterpenes and hydrocarbons showed significant cytotoxic activity against Ehrlich ascites carcinoma and antimicrobial activity against *Staphylococcus aureus* and *Streptococcus* species and moderate antimicrobial activity against *Sarcina lutea, Mycobacterium* and Bacillus species. But they showed negative effect against *pseudomonas aeroginosa* and *E.coli. C. alopecuroids* oil showed a moderate activity against *proteus* species while *C.rotundus* oil was inactive [38].

#### **Antidepressant activity**

Depression is considered as an affective disorder characterized by change in mood, lack of interest in the surroundings. The prevalence of depression in general population is estimated to be around 5%. The efficacy of the drugs for depression is very limited so the need for newer, better-tolerated and more efficacious treatments is remaining high. Therefore, herbal therapies should be considered as alternative medicines. The n-hexane extract of *C.scariosus* oil exhibited antidepressant activity in mice. With two dose level at 100mg/kg and 200mg/kg anti-depressant activity was screened using forced swim test and tail suspension test in mice and results were compared with standard drug imipramine 15mg/kg. *C.scariosus* n-hexane extract oil significantly p<0.001 reduced the immobility time in both dose level at FST and TST which is similar to standard drug imipramine. The antidepressant activity of n-hexane extract of *C.scariosus* oil may due to increase of nor epinephrine level in synapses [39].



#### Mosquito repellent activity

Malaria resist is the major disease burden and its control hampered many operational and technical reasons and among the technical reason insecticidal resistance namely development of insecticidal resistance namely development of insecticide resistance in malaria vectors to the commonly used synthetic chemical insecticides in public health sprays has made the disease control more difficult. The majority of commercial repellent products contain the DEET, it is not the ideal product as allergic and toxic effects have been documented and its solvent characterstics can demage plastic and other synthetic materials. The hexane extract of tuber of *C.rotundus* used to determine their effect on mosquito vector and comparison with DEET. The tuber extracts showed more effective at all the dose. The tuber extracts are more effective for repellency of all the mosquito vector even at low dose. Highest dose of 10% tuber extract evoking 100% repellency. 100% repellency obtained against *An.culicifacies* in 4 and 6 hours, *An.stephensis* 100% repellency in 6 hours and *Cx.quinquefasciatus* was 100% repellency in 6 hours at the 105 concentration. Against DEET *An.*culicifasciatus have shown 100% repellency in 1, 2 and 6 hours with 2.5% of *C.rotundus* extract [40].

#### Age associated changes

Aging is a multifactorial biological process which is accompanied by general decrease in biochemical and physiological functions that leads to the decreased ability of an individual to respond to a wide range of stresses and increased susceptibility to age associated degenerative diseases and death. It increases total cholesterol and decreases phospholipids, leading to increased cholesterol-phospholipids molar ratios in hepatic mitochondria, brain and cerebral synaptic membranes. Medicinal plants have high density of important nutrients such as minerals, vitamins, phytochemicals and natural antioxidants that can prevent chronic diseases such as cancer, arteriosclerosis, diabetes and improving the quality of life. The ethanolic extract of *C.rotundus* rhizomes on age associated changes in glucose. Total cholesterol, triglycerides, LDL cholesterol and a decrease in HDL cholesterol was observed in aged rats when compared to young rats. Administration of CRRE to aged rats prevented the age associated changes on glucose, total cholesterol, triglycerides, LDL cholesterol and VLDL cholesterol level was found to be increased significantly in both young and aged rats after treatment with CRRE [41].

#### Antidiarrhoeal activity

Diarrheal diseases are most common infectious worldwide, are predicted to remain a leading health problem. The decoction of *C.rotundus* tubers on adherence of enteropathogenic E.coli and invasion of enteroinvasive *E.coli* and *shigella flexnerito* HEp-2 cells was evaluated as a measure of effect on colonization. Effect on enterotoxigenic *E.coli* (ETEC) heat labile toxin, heat stable toxin and cholera toxin was assed. The decotion showed antigiardial activity reduced bacterial adherence to and invasion of HEp-2 cells and affected production of cholera toxin and action of labile toxin. The decotion of *C.rotundus* does not have marked antimicrobial activity



and exerts its antidiarrhoeal action by mechanisms other than direct killing of the pathogen [42].

### Wound healing activity

Wounds are physical injuries that result in an opening of the skin. Wound healing is an important biological process involved in tissue repair and regeneration. A wound is described as 'a break in the continuity of tissue' from violence or trauma and is regarded as healed if there is restoration of the wounded or inflamed tissue to normal condition. Proper healing of wounds is essential for the restoration of disrupted anatomical continuity and disturbed functional status of the skin. The exact pathogenesis of wound healing in diabetic wounds is not clearly understood. Evidence from studies involved in both human and animal reveal several abnormalities in the various stages in healing process.

It is an enigmatic and debilitating complication and posesses of a serious of challenges in clinical practice. Mainly the healing process begins immediately following injuring when the platelets coming to contact with exposed collagen as platelet aggregation proceeds, clotting factors are released and resulting in the deposition of a fibrin clot serves as a provisional matrix and sets the stage for the subsequent events of healing. Many researchers have reported the improvement in the wound healing process by various plant extracts and isolated compounds in animal models. An alcoholic extract *C.rotundus* tubers showed considerable difference in response in all the excision, incision and dead space wound model as compared to those of standard drug nitrofurazone ointement (0.2% w/w NFZ) in terms of wound contracting ability, wound closure time and tensile strength [43].

### **Micro propagation**

*C.scariosus* axillary bud explants inoculated on SH medium, supplemented with different concentrations of Benzyl adenine, Kinetin and Indole-3-butyric acid for *in vitro* regeneration. Maximum numbers of shoots observed on media containing1.0mg/lt BA and 1.0mg/lt Kn after 2 weeks of culture inoculation. Kn 1 mg/lit with Adenosine 1mg/lit and Charcoal 500mg/lit gave best results in rooting from shoots. <sup>[44]</sup> Efficient flowering 80% was recorded on SH media with Kn 0.75mg/lt+ADS 1.0mg/lt+activated charcoal 500 mg/lt. Multiple shooting and multiple rooting was effective on full strength SH medium supplemented with Kn1.5mg/lt+ADS1.0mg/lt+activated charcoal 500 mg/lt.

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