Review Article

Phyto-Pharmacology of *Cichorium intybus* as Hepatoprotective Agent

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ABSTRACT

Cichorium intybus Linn. family Asteraceae is commonly known as chicory and is used in Indian System of Medicine as cardiotonic, anti-inflammatory, digestive, stomachic, liver tonic and diuretic. The chicory plant is used in Indian medicine as a tonic, curative in acne, opthlamia and inflamed throat. The plant is bitter, acrid, thermogenic, anti-inflammatory, appetizer, digestive, stomachic, cholagogue and cardiotonic. Historically, chicory was grown by the ancient Egyptians as a medicinal plant, coffee substitute, and vegetable crop and was occasionally used for animal forage. Its various plant parts have beneficial role in treating liver diseases, enlargement of spleen, as bitter tonic effective in jaundice, liver enlargement etc. The present review focuses on the constituents and phyto-pharmacological uses of the plant, particularly in imparting hepatoprotection against various types of liver toxicities.

Keywords: Hepatoprotection, *Cichorium intybus*, Liver toxicity, Herbal hepatoprotectants, Hepatotoxicity.

INTRODUCTION

Cichorium intybus Linn, family Asteraceae, is commonly known as chicory and is used in Indian System of Medicine as cardiotonic, anti-inflammatory, digestive, stomachic, liver tonic and diuretic. The phytoconstituents are distributed in the whole plant. These are sesquiterpene lactones, lactucin, 8-deoxylactucin, lactopicrin, cichoriolide A, B and C.

Occurrence

The genus *Cichorium* (Asteraceae) consists of six species with major distribution areas in Europe and Asia[1].

Taxonomical Classification

Kingdom: Plantae; **Subkingdom:** Tracheobionta; **Superdivision**: Spermatophyta; **Division:** Magnoliophyta; **Class**: Mangnoliopsida; **Sub class**: Asteridae; **Order:** Asterales; **Family**: Asteraceae; **Genus**: Cichorium.

Botanical Features

Cichorium intybus L., commonly known as chicory is a erect, usually rough and more or less glandular herb stems 0.3-0.9m, angled or grooved, branches tough, rigid spreading radical and lower leaves 7.5-15 cm, pinatified, lobes toothed, pointing downwards, upper leaves alternate[2]. The flowers blooms from May to the summer time.

The colors of the flowers are deep sky-blue. It is a capitate flower and its diameter is about 3-4 cm, and has a brilliant bluish-purple (sometimes pink or white), radially symmetrical bloom.

Flowers are singularly arranged along the length of a fibrous and rigid, dark-green stem.

This wildflower has two types of leaves: large-dandelion-shaped leaves near the base of the stem and small lanceolate-to-oblong-shaped leaves along the length of the stem.

Vernacular Names

Cichorium intybus is called as Hindubar, Indyba in arabic, Zral in Baluchisthan, Chicory in California, Bunk, Chicory in English, Kichora, Kikori in Greek, Kasani in Gujrathi, Kasni in Hindi, Kasani in Persian, Gul, Hand in Punjabi, Kasni, Tsikorie, Kashini virai in Tamil, Kasini vittulu in Telugu, Kasani in Urdu[3].

These various common or local names signify the widespread use of this plant by different folkloric group.

History

Historically, chicory was grown by the ancient Egyptians as a medicinal plant, coffee substitute, and vegetable crop and was occasionally used for animal forage.

In the 1970s, it was discovered that the root of *C. intybus* contained up to 40% inulin, which has a negligible impact on blood sugar and thus is suitable for diabetics[4].

Varieties

Cichorium intybus is cultivated for numerous applications and can be divided into four main varieties or cultivating groups according to their use[5] "industrial" or "root" chicory, predominantly cultivated in northwestern Europe, India, South Africa, and Chile, produces the taproot as a coffee substitute or for inulin extraction; "Brussels" or "witloof" chicory is commonly cultivated around Europe as industrial chicory for etiolated buds (chicons) by forcing; "leaf " chicory is used as fresh or cooked vegetables; and "forage" chicory, initially derived from wild chicory commonly found along roadsides and waste areas, has been used since the mid-1970s to

intensify herbage obtain ability in perennial pastures for livestock.

Uses

Figure 1: Leaves of *Cichorium Intybus*

The chicory plant is used in Indian medicine as a tonic, curative in acne, opthlamia and inflamed throat. The plant is bitter, acrid, thermogenic, anti-inflammatory, appetizer, digestive, stomachic, cholagogue, cardiotonic, depurative, diuretic, emmenagogue, febrifuge, alexeteric and tonic. It is useful in vitiated conditions of kapha and pitta, cephalalgia, hepatomegaly, inflammations, anorexia, dyspepsia, flatulence, colic, gout, burning sensation, allergic conditions of skin, insomnia, jaundice, skin disease, leprosy, chronic and bilious ever, vomiting, asthma and general debility[6,7]. The seeds are reported to be carminative and cordial, and brain tonic and useful in headache and asthma. A decoction of seeds is used in obstructed menstruation and for checking bilious vomiting.

C.intybus roots are roasted and used to add a bitter, millow taste to coffee and tea or used as a substitute for coffee[8]. Added to coffee, it counteracts with caffeine and helps in digestion. In South Africa although it is considered as widespread weed, leaves, stems and roots are made into tea for jaundice and cichory syrup is used as a tonic and for purifying medicine for infants. Seeds of cichory are carminative, agglutinating and cholagogue[9,10].

The ethanobotanical studies have reported the use of leaves in curing jaundice, liver disorders, vomiting, loose motion, fever and pleurisy[11]. Leaves and roots are used to disperse the swelling of joints. In Turkey, an ointment is made from the leaves for wound healing[12].

Its various plant parts have beneficial role in treating liver diseases, enlargement of spleen, as bitter tonic effective in jaundice, liver enlargement etc.

Chemical Constituents

The main constituents of *Cichorium intybus* reported to be present in the root are inulin, reducing sugars and sucrose[13].

Sesquiterpene lactones such as cichoriosides A, B and C, guanine type sesquiterpene lactones such as 8-deoxy lactucin, lactucopicrin, crepidiaside B, and 11 beta, 13 dihydrolactucin, two known germacrene type sesquiterpene glycoside- Picriside B and Sonchuside A and Eudesmane type sesquiterpene glycoside Sonchuside C were reported by Seto[14].

Du H reported seven compounds from the roots of *Cichorium intybus* and identified four of them as alpha-amyrin, taraxerone, baurenyl acetate and beta-sitosterol[15](Fig. 2).

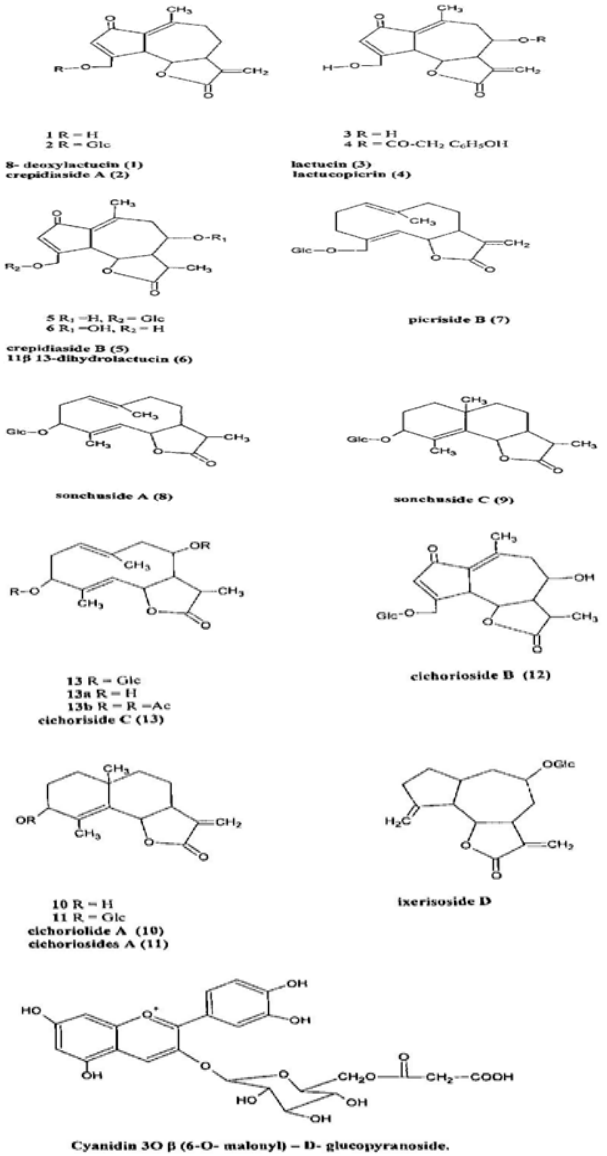

Figure 2: Main Constituents Reported in *C.intybus* Plant

Hepatoprotective Activity of *C.intybus* Against Various Hepatotoxins

Table 1: Hepatoprotective Activity of *C.intybus* against Various Hepatotoxins

S. No	Plant Part Used	Extract	Hepatotoxic Agent	Dose	Reference
1	Roots and root callus		CCl_4		15
2	Seeds	Alcoholic	CCl_4		16
3	Leaves	Hydroalcoholic	CCl_4	50mg/Kg and 100mg/Kg	17
4	Roots		CCl_4 and d-Galactosamine	Pretreatment 800mg/Kg body weight	18
5	Whole plant	Ethanol	Thioacetamide	25mg/Kg	19
6	Leaves	Ethanol:water (1:1v/v)	CCl_4	200, 400, 500 mg/Kg body weight	20
7	Whole plant	Ethanolic	CCl_4		21
8	Leaves	Methanolic	CCl_4	250 and 500mg/Kg	22
9	Whole plant	Ehanolic	Ethanol	300mg/Kg	23,24
10	Esculetin –a phenolic compound		Paracetamol, CCl_4	Esculatin (6mg/Kg wt)	25
11	Seeds	Cichotyboside, a new guainolide sesquiterpene glycosides.	CCl_4		26
12	Roots	Aqueous	Oxytetracycline	75 mg/ Kg by gastric tube	27
13	Liv 52	Petroleum ether, chloroform, butanol and water	CCl_4	1 mg of water-extractables per 0.2 ml of liver homogenates	28
14	Liv 52, Livokin		CCl_4	2.6 ml/Kg wt, 5.6 ml/Kg Wt	29
15	Liv 52 (seeds of C.intybus 65 mg/tablet)	Syrup	CCl_4	0.5 ml of syrup	28
16	Liv 100	Syrup	Antitubercular drugs	400mg/Kg	30
17	Liv 52 (seeds of C.intybus 65 mg/tablet)	Syrup	Antitubercular drugs	500mg/Kg, p.o	31
18	PHF A	Powder suspended in distilled water using 1 % sod. CMC	Antitubercular drugs (isoniazide and Rifampicin)	200 mg/Kg, 400mg/Kg, 600mg/Kg	32
19	Livina	A poly herbal liquid formulation	Ethanol	2ml of syrup	33
20	Liv 52	Syrup	Antitubercular drugs	Different doses for children and adults	28

Table 2: Various Liver Intoxicants and their Mechanism of Action

S. No.	Liver Intoxicant	Mechanism of action	Dose for hepatotoxicity	References
1	Carbontetrachloride (CCl_4)	♦ It is metabolized in endoplasmic reticulum and mitochondria with the formation of CCl_3O^- a reactive oxidative free radical, which initials lipid peroxidation. ♦ $CCl_4 \rightarrow CCl_3O^- + O^-$ ♦ Within 24hrs of administration centrilobular necrosis and fatty changes are observed. ♦ The max. conc is achieved within 3hrs of administration after which the level falls with almost no CCl_4 left in 24hrs	0.1 to 3 ml/Kg body weight, intraperitoneally	59,60,61
2	Galactosamine	♦ Liver injury as in Viral hepatitis. ♦ It disturbs the synthesis of essential uridylate nucleotides resulting in organelle injury and ultimate death. ♦ Histologic changes include necrosis and inflammatory infilteration of periportal areas, mitoses and cell proliferation, appearance of Councilman bodies, and an increased no. of Kupper cells. ♦ Depletion in these nucleotids affects the normal synthesis of RNA and thus protein synthesis. ♦ This further causes increase in cell permeability leading to enzyme leakage and finally cell death. ♦ The oxygen consumption and the no. of viable hepatocytes is reduced	400mg/Kg body weight, intraperitoneally	30,62
3	Thioacetamide	♦ Its metabolite (s-oxide) causes hepatic injury. ♦ Interferes with the movement of RNA from the nucleus to the cytoplasm causing membrane injury. ♦ Decreases the volume of bile and its contents, and also the no. of viable hepatocytes.	100mg/Kg body weight, subcutaneously	60
4	Paracetamol	♦ Its oxidative product, acetyl-P-benzoquinoneimine, binds with suphydryl groups of protein resulting in cell necrosis in the liver. ♦ It causes necrosis of centrilobular hepatocytes followed by large hepatic lesions.	1gm/Kg body weight P.O	61
5	Lipopolysaccharides	♦ It releases several inflammatory mediators like tumor necrosis factor alpha, prostaglandins nitric oxide, interleukins, & leukotrienes causis hepatocellular damage and apoptosis. ♦ d-GalN mainly inhibits hepatic protein synthesis. ♦ When co-administered with LPS, d-GalN inhibits hepatic cytoprotective protein synthesis.		30
6	Alcohol	♦ Causes an increase in hepatic lipid peroxidation which causes alterations in membrane lipid composition. ♦ Enhances the generation of oxy free radicals during its oxidation in liver, which further results in loss in membrane integrity and structure. ♦ Ethanol inhibits glutathione peroxidase, decrease activity of superoxide dismutase, catalases, with the increase in glutathione levels in liver. ♦ Ethanol exposure causes damaging effects due to free radical generation or may be due to acetaldehyde formed by its oxidation.		63

7	Antitubercular drugs	♦ Isoniazide, Rifampicin and Pyrazinamide are potentially hepatotoxic. They are potentially hepatotoxic when given alone or in combination. ♦ INH is metabolized to monoacetyl hydrazine, which is further metabolized by cytochrome P450 to a toxic product leading to hepatotoxicity. ♦ Rifampicin also increases the metabolism of INH to isonicotinic acid and hydrazine, both of which are hepatotoxic. ♦ Pyrazinamide decreases the bioavilability of rifampicin and increases its clearance. ♦ Pyrazinamide in combination with INH and rifampicin causes increased incidences of hepatotoxicity.		31
8.	Arsenic	♦ Arsenic forms strong complexes with sulfhydryl groups and generates ROS (reactive oxygen species) such as superoxide, hydroxyl and peroxyl radicals during its metabolism in cells. ♦ Its exposure causes depression in antioxidant defense systems leading to various oxidative damage.		64
9	NDEA/DEN (N-nitrosodiethyl amine)	♦ It forms DNA carcinogen adducts in the liver and induces hepatocellular carcinomas without cirrhosis through the development of putative pre neoplastic enzyme-altered hepatocellular focal lesions.		65

Other Pharmacological Activities of the Plant

Cichorium intybus is an extremely versatile plant and is known to possess a number of activities like anti-ulcer, hepatoprotective, cardioprotective, antibacterial, antioxidant, antifungal, gastroprotective, antihelmintic, free radical scavenging, analgesic, tumour protective, anti-allergic and other miscellaneous activities[34].

Preliminary pharmacological study of different varieties of *Cichorium intybus* showed that alcoholic extracts of all 8 varieties had a guanidine like action (marked depression of amplitude and rate) on the isolated toad heart but with variable potency, showing some promise for the use of *Cichorium* extracts to treat diseases characterized by tachycardia, arrhythmia and fibrillation[35].

Both the alcoholic and aqueous extracts of *Cichorium intybus* (200-800 mg/kg, i.p) exhibited anti-MES and anti-metrazole but limited anti strychnine activity. These extracts potentiated pentobarbitone and ethanol induced hypnosis in mice, exhibited analgesia and potentiated morphine analgesia in rats.

Dose related antipyretic effect against LSD induced hyperpyrexia was observed in rats. Both the extracts showed anti-inflammatory activity against formalin induced edema[36].

The spermatogenic inhibition in mice is caused by aqueous root suspension of *Cichorium intybus* L. It has also been reported to possess cytotoxic activity towards cultured cancer cells[37,38].

Clinical trials conducted on alcoholic extract of *Cichorium intybus* against pyorrhoea in 40 dental patients revealed that when alcohol dried extracts of chicory weighing 250 mg were brushed by massaging the inflammed gums twice a day for three weeks, it significantly ($p< 0.0\ 1$) reduced the gingival inflammation and bleeding[39].

The presence of plant extracts of *Cichorium intybus and Solanum nigrum,* in the reaction mixture containing calf thymus DNA and free radical generating systems, protects DNA against oxidative damage to its deoxyribose sugar moiety effect[40]. *Cichorium intybus* effect was observed to be much pronounced as compared to the effect of *S.nigrum.*

Inhalative occupational and ingestive immediate type allergy has been reported to be caused by chicory[41].

Aqueous extract of *Cichorium intybus* (plant drug from Bombay market) was fractionated to yield a compound (A) identified as 8, 15-dihydroxy-2-oxo-l-(10)3, 11(13) guaiatrienel2.6-olide. The extract and the compound (A) exhibited antihepatctoxic activity *in-vitro* testing[42,43].

Pharmacological studies of the root extracts from *Cichorium intybus* have shown their anti-inflammatory and hepatoprotective activities. Oral pretreatment of Jigrine was studied on hepatic damage induced by paracetamol in rats. Alcohol, CCl_4 and paracetamol treatment produced an increase in serum transaminase, bilirubin, plasma prothrombintime amd tissue lipid peroxides in liver. These effects were progressively reduced by pretreatment doses of Jigrine. The activity of Jigrine was also compared with Liv-52, a known Ayurvedic hepatoprotective formulation[9,44]. Aqueous or butanol extracts of Liv-52 stimulated the activity of aminopyrine N-demethylase and aniline hydroxylase while the petroleum ether extract stimulated the activity of cytochrome oxidase, succinate dehydrogenase and total

ATPase in the mitochondrial fraction, when added to the post mitochondrial fraction of the liver of normal and CCl_4 treated rats; chloroform extract, inhibited the activities of lysosomal acid phosphatase, acid ribonuclease and cathepsin B in total liver homogenate[28].

The methanolic fraction and a phenolic compound of seed of *C.intybus* were found to possess a potent antihepatotoxic activity comparable to the standard drug silymarin. The results were supported by biochemical parameters and histopathological studies of the liver[45].

Crude ethanolic extracts of seeds of *Cichorium intybus* and the aerial parts of *Guetterda adamanisca* etc. when administered orally on days 1-10 post coitum showed significant contraceptive activity in rats invariably associated with a significant reduction in number of implants[46].

Paracetamol induced rise in serum enzymes was protected by pretreatment of rats with Esculetin (6 mg/kg), a phenolic compound found in *Cichorium intybus* and *Bougainvllra spectabillis*. It has also been reported to prevent the CCl_4 induced rise in serum enzymes, indicating thereby that esculetin possesses anti-hepatotoxic activity[25].

Methanol extract of the root of *Cichorium intybus* has been reported to possess significant anti-inflammatory activity against carrageenin induced edema in rats hand paw at the dose of lOOOmg/kg. Methanol, ethyl acetate and butanol extracts were shown to possess hepatoprotective activity. A significant reduction of blood glucose levels was observed in methanol fraction[44].

The leaves of *Kasini (Cichorium intybus)* have been reported to possess considerable amounts of antioxidants. They were also found to inhibit lipid peroxidation to a significant extent, revealing their candidature for use in the preparation of anti-oxidant formulations[47].

Kasini (Cichorium intybus) extract (1 ml of 10% extract/kg body weight) when given orally for 28 days has been shown to decrease the levels of magnesium and phosphorus in urine in hyperoxaluric rats. The urine output was high in the extract treated rats[48].

The water soluble antioxidant properties of *Cichorium intybus* var. Silvestre were evaluated *in vitro* as antioxidant activity (AA) and *ex vivo* as protective activity (PA) and it was shown that the vegetable contained both biologic antioxidant and pro-oxidant compounds[49].

70% ethanolic extract of a group of medicinal plants, including *Selenium marianum, Matricaria chamomilla*, *Calendula officinalis, Cichorium intybus* and *Dracocephalum kotschyi,* growing in Iran have been examined by Amirghofran for mitogenic activity on human peripheral blood lymphocytes and thymocytes. The results have indicated that none of the extracts had a direct mitogenic effect on human lymphocytes or thymocytes (stimulation index, S.I<0.07). Among the plants studied, *Cichorium intybus* and *Taraxacum officinale* have shown a complete inhibitory effect on the proliferation of lymphocytes in the presence of PHA[50] (SI range 0.01-0.49).

Recently a molecular mechanism of anti-inflammatory action of sesquiterpene lactones, via inhibition of transcription factor NF-a p, has been proposed[51,52]. The tumour-inhibitory effect of an ethanolic extract of chicory root was studied against Ehrlich ascites carcinoma in mice. Significant results were obtained at doses from 300 to 700mg/kg[53].

The effects of the ethanol extract of *Cichorium intybus* (CIEE) on the immunotoxicity of ethanol (EtOH) in ICR mice. The combination of CIEE and EtOH showed significant increase in the circulating leukocytes and the relative weights of liver, spleen and thymus as compared with those in mice treated with EtOH alone. The findings indicated that the immunotoxicity induced by EtOH is significantly restored or prevented by CIEE treatment[23].

The hypoglycemic and hypolipidemic properties of an ethanolic extract of *Cichorium intybus* (CIE) for its use in the treatment of diabetes mellitus. Hypoglycemic effects of CIE were observed in an oral glucose tolerance test (OGTT) in which, a dose of 125 mg of plant extract/kg body weight exhibited the most potent hypoglycemic effect. Also, daily administration of CIE (125 mg/kg) for 14 days to diabetic rats attenuated serum glucose by 20%, triglycerides by 91% and total cholesterol by 16%. However, there was no change in serum insulin levels, which ruled out the possibility that CIE induces insulin secretion from pancreatic β-cells. In addition, hepatic glucose-6-phosphatase activity (Glc-6-Pase) was markedly reduced by CIE when compared to the control group.

Some constituents of Chicory plant extract may serve as a source of drugs useful in chemotherapy of some infections caused by bacteria and also as an antioxidant agent[54]. However, the alcoholic extract of *C. intybus* had no antibacterial effect on Gram +ve bacteria including *Streptococcus pyrogen*, *Staphylococcus aureus* & *Enterococcus*.

Similarly, Khakzadine found that chicory leaf extract with different solvents had no antibacterial effect on *Staphylococcus aureus* and *E. coli*[57].

An extract containing crude sesquiterpene lactones (CSL) from chicory was found to be effective against the motility of third stage (L3) lungworm larvae in abdominal fluid. The vasorelaxant activity of caffeic acid derivatives from *Cichorium intybus* and *Equisetum arvense*[55,56] was also reported.

Various Live Hepatotoxins

Several chemicals are known to cause hepatotoxicity in experimental animals, like carbon tetrachloride (CCl_4), Galactosamine, Lipopolysaccharide, d-Galactosamine/Lipopolysaccharide (GaIN/LPS), Thioacetamide, Paracetamol, Alcohol, Anti-tubercular

drugs, arsenic and N-Nitrosodiethylamine (NDEA) (Table 2).

In Vitro Hepatoprotective Activity of the Plant

In vitro and *in vivo* hepatoprotective properties of *Cichorium endivia* L. extract (CEE), and identified its chemical constituents[65]. CEE significantly blocked the oxidative stress and cytotoxicity induced by *tert*-butyl hydroperoxide (*t*-BHP) in HepG2 cells. Oral administration of CEE to mice before the treatment of *t*-BHP exhibited a markedly protective effect by lowering serum levels of ALT and AST, inhibiting the changes in liver biochemistry including MDA, SOD, GSH and GST, as well as ameliorating the liver injuries according to the histopathological observations. Phytochemical analysis of CEE showed the presence of five compounds identified as 2-furanmethanol-(5'→11)-1,3-cyclopentadiene-[5,4-c]-1*H*-cinnoline, which is a new cinnoline derivative derived from a natural source but not synthesis, 2-phenylethyl-*β*-D-glucopyranoside, kaempferol-3-*O*-β-D-glucoside, kaempferol, and adenosine.

The activity of HD-03, a polyherbal formulation, on two hepatitis B surface antigen (HBsAg) expressing human hepatocellular carcinoma cell lines, PLC/PRF/5 and HepG2/A2 was studied. They observed that HD-03 downregulates HBsAg expression from these cell lines. Further their studies also show that this effect is neither due to cytotoxicity on the cell lines nor due to blockade of the release of the antigen from the cells nor due to binding of the substance with the antigen[66].

The potential cytoprotective effect of Liv.52 against toxicity induced by copper (Cu^{2+}) in HepG2 cells was studied. Cu^{2+} at 750 µM induced cytotoxicity to HepG2 cells as determined by MTT assay. The toxicity was brought about by increased lipid peroxidation, DNA fragmentation and decreased GSH content. But, upon treatment with Liv.52 cell death induced by Cu^{2+} was significantly abrogated by inhibition of lipid peroxidation by 58% and DNA fragmentation by 37%. Liv.52 increased the GSH content by 74%. Activities of the antioxidant enzymes catalase, glutathione peroxidase and superoxide dismutase were increased by 46%, 22% and 81% respectively in Liv.52 treated cells[67].

The biological activity, mode of action, toxicity and forthcoming applications of some of the potential lead molecules for hepatoprotective activity was discussed by Gosh. The hepatoprotective potential of several herbal medicines was clinically evaluated. Significant efficacy was observed with silymarin, glycyrrhizin and Liv-52 in treatment of hepatitis, alcoholic liver disease and liver cirrhosis[68].

The anti-hepatitis B property of cichoric acid by the D-galactosamine (D-GalN)-induced normal human HL-7702 hepatocyte injury model, the duck hepatitis B virus (DHBV)-infected duck fetal hepatocytes and the HBV-transfected cell line HepG2.2.15 cells, respectively was evaluated. The results showed that cichoric acid attenuated significantly D-GalN-induced HL-7702 hepatocyte injury at 10–100 µg/mL and produced a maximum protection rate of 56.26%. Moreover, cichoric acid at 1–100 µg/mL inhibited markedly DHBV DNA replication in infected duck fetal hepatocytes. Also, cichoric acid at 10–100 µg/mL reduced significantly the hepatitis B surface and envelope antigen levels in HepG2.2.15 cells and produced the maximum inhibition rates of 79.94% and 76.41%, respectively. This study verifies the anti-hepatitis B effect of cichoric acid from *Cichorium intybus* leaves[69].

The possible mechanisms of hepatic protective activity of *Cichorium intybus* L. (cichory) in acute liver injury was investigated. Pathological observation, reactive oxygen species (ROS) detection and measurements of biochemical indexes on mouse models proved hepatic protective effect of *Cichorium intybus* L. Identification of active compounds in *Cichorium intybus* L. was executed through several methods including ultra-performance liquid chromatography/time of flight mass spectrometry (UPLC-TOF-MS). Similarity ensemble approach (SEA) docking, molecular modeling, molecular docking, and molecular dynamics (MD) simulation were applied in this study to explore possible mechanisms of the hepato-protective potential of *Cichorium intybus* L. Further the chemical composition of *Cichorium intybus* L., and their key targets was found. Furthermore, *in vitro* cytological examination and western blot were used for validating the efficacy of the selected compounds[70].

REFERENCES

1. Guguloth S, Riyazullah MS, Rajesh B, Santhosh, Pharmacognostical profiles on *Cichorium intybus* Linn., Leaves, International Research Journal of Pharmacy, 2(11), 2011, 85-87.
2. Bais HP, Ravishankar GA, *Cichorium intybus* L, Cultivation, processing, utility, value addition and biotechnology with an emphasis on current status and future prospects, Journal of Science of Food and Agriculture, 81, 5, 2001, 467-484.
3. Kirtikar KR, Basu BD, Indian medicinal plants, Vol I,II,II, IV and VI,2nd reprint Edn, Periodical experts book agency Delhi, 649-651, 1048-1055, 1140-1146, 1199-1206, 1210-1213, 1228-1231, 1980-1982, 2133-2135, 2141-2142, 2149-2150, 2171-2172, 2462-2463, 1987.
4. Judzentiene A, Udiene JB, Volatile constituents from aerial parts and roots of *Cichorium intybus* L. (chicory) grown in Lithuania, Chemija, 19, 2008, 25–28.
5. Cadalen T, Morchen M, Blassiau C. Development of SSR markers and construction of a consensus genetic map for chicory *Cichorium intybus* L., Molecular Breeding, 25, 4, 2010, 699–722.
6. Anonymous. The Wealth of India, Raw Materials. Publication and Information Directorate, Council of Scientific and Industrial research, New Delhi India, 3(revised), 1992, 556-561.
7. Prajapati DS, Purohit SS, Sharma KA, Kumar T, A Handbook of Medicinal plants a Complete Source Book, Agrobios Publishers, Agro house behind Nasrani Jodhpur, India, section-ii, 2003, 139.
8. Ara D, Philadelonia P, The review of Natural Products, The most complete source of Natural Product information, published by Fact and Comparisons, USA, 2, 2002, 162-164.
9. Zafar R, Ali SM, Antihepatoxic effects of root callus extracts of *Cichorium intybus* L, J. Ethnopharmacol, 63, 3, 1998, 227-231.

10. Zafar R, Ali SM, *Cichorium intybus* Linn. A Review, Hamdard Medicus, 41, 4, 1998, 98-109.

11. Bhalla S, Patel JR, Bhalla NP, Ethanomedicinal observations of some Asteraceae of Bhundelkhand region,Madhya Pradesh, J.Econ Tax Bot, 23, 173-176.

12. Sezik E, Yes E, Ilada, Honda G, Takaishi Y, Takeda and Tanaka T, Traditional medicine in Turkey X. Folk medicine in Central Anatolia, Journal of Ethanopharmacology, 75, 2-3, 2001, 95-115.

13. Wight WA, Niekerk JV, Determination of reducing sugars, sucrose and inulin, J. Agri. and Food Chem, 31, 1983, 282-285.

14. Seto M, Miyase T, Umehara, K, Hirano, Y, Otani N, Sesquiterpene lactones from *Cichorium endivia* L and *C.intybus* L. and Cytotoxic activity, Chem. Pharm. Bull, 36, 7, 1988, 2423-2429.

15. Du H, Yuan S, Jiang, P, Studies on the chemical constituents of *Cichorium intybus* L. Zhongguo Zhongyao Yao Za Zhi, 23, 11, 1998, 682-683.

16. Ahmed B, Al-Howiriny TA, Siddiqui AB, Antihepatotoxic activity of seeds of *Cichorium intybus*, J. Ethnopharmacol, 87, 2003, 237-240.

17. Jamshidzadeh A, MJ Khoshnood, Z Dehghani, H Niknahad, Hepatoprotective Activity of *Cichorium intybus* L. Leaves Extract Against Carbon Tetrachloride Induced Toxicity, Iranian Journal of Pharmaceutical Research, 1, 2006, 41-46.

18. Upur H, Amat N, Blazekovic B, Talip A, Protective effect of *Cichorium glandulosum* root extract on carbon tetrachloride-induced and galactosamine-induced hepatotoxicity in mice, Food and Chemical Toxicology, 47, 2009, 2022-2030.

19. Madani H, Talebolhosseini M, Asgary S, Naderi GH, Hepatoprotective Activity of *Silybum marianum* and *Cichorium intybus* against Thioacetamide in Rat, Pakistan Journal of Nutrition, 7, 1, 2008, 172-176.

20. Heibatollah S, Nikbakht MR, Ghaitasi I, Sabzali S, Hepatoprotective effect of *Cichorium intybus* on CCl_4 induced liver damage in rats, African Journal of Biochemistry Research, 6, 2008, 141-144.

21. Aktay G, Didem D, Ender E, Fatma E, Hepatoprotective effects of Turkish folk remedies on experimental liver injury, Journal of Ethanopharmacology, 73, 2000, 121-129.

22. Atta H, Elkoly TA, Mouneir SM, Gehan K, Alwabel NA, Shaimaa Zaher, Hepatoprotective effects of Methanol extracts of *Zingiber officinale* and *Cichorium intybus*, Indian J Pharm Sci., 72, 5, 2010, 564-570.

23. Kim JH, Mun YJ, Woo WH, Jeon KS, An NH, Park JS, Effects of the ethanol extract of *Cichorium intybus* on the immunotoxicity by ethanol in mice, Int Immunopharmacol., 2, 2002, 733–744.

24. Kim TW, Yang KS. Antioxidative effects of *Cichorium intybus* root extract on LDL (low density lipoprotein) oxidation. Arch Pharm Res., 24, 2001, 431–436.

25. Gilani AH, Janbaz KH, Shah BH, Esculetin prevents liver damage induced by paracetamol and CCl_4, Pharmacol Res, 37, 1, 1998, 31-5.

26. Gilani AH, Janbaz KH. Evaluation of the liver protective potential of *Cichorium intybus* seed extract on Acetaminophen and CCl_4-induced damage, Phytomedicine, 1, 1994, 193–197.

27. Eman GE, Helal Samia M, Abd El-Wahab, Atef M.Moussa Sharaf, Ghada Zedan, Effect of *Cichorium intybus* L. on fatty liver induced by oxytetracycline in albino rats, The Egyptian Journal of Hospital Medicine, 45, 2011, 522–535.

28. Bardhan Piyush, Sharma SK, Garg NK, *In vitro* Effect of an Ayurvedic Liver Remedy on Hepatic Enzymes in Carbon Tetrachloride Treated Rats, Indian Journal of Medical Research, 82, 1985, 359.

29. Girish C, Koner BC, Jayanthi S, Rao KR, Rajesh B, Suresh Chandra Pradhan, Hepatoprotective property of six polyherbal formulations in CCl_4 induced liver toxicity in mice, Indian Journal of Experimental Biology, 47,2009, 257-263.

30. Saraswat B, Visen PKS, Dayal R, Agarwal DP, Patnaik GK, Protective action of ursolic acid against chemical induced hepatotoxicity in rats, Ind. J. Pharmacol, 28, 1996, 232-39.

31. Padma VV, Suja, V, Shyamala, DCS, Prema, Hepatoprotective effect of Liv-52 on antitubercular druginduced hepatotoxicity in rats. Fitoterapia, 69, 1998, 520-522.

32. Desai SK, Gavitre BB, Patil MD, Mathapati SS, Gaikwad DT, Kulkarni VS, Khade TS, Nagare SK, Jadhav SG, Bobe KR, Khade AB, Khade MA, Evaluation of hepatoprotective activity of a polyherbal formulation (PHF-A) by using isoniazid and rifampicin induced hepatotoxicity in rats, International Journal of Pharmacology And Therapeutics, 1, 2011, 32-41.

33. Darbar Soumendra, Matish Ranjan Chakraborty, Shyama prasad Chattarjee, Bhaskar Ghos, Protective Effect of Livina, A Polyherbal liquid formulation against ethanol induced liver damage in rats, Ancient Science of Life, 28, 2009, 14-17.

34. Saxena Rahul, Kunj Bihari Sulakhiya, Rathore Manoj, *Cichorium intybus* Linn.: A Review of Pharmacological profile, International journal of Current Pharmaceutical Research, 6, 4, 2014, 11-15.

35. Balbaa ST, Zaki AY, Wahab, A Preliminary Phytochemical and pharmacological investigations of the roots of different varieties of *Cichorium intybus*, Planta Medica, 24, 2, 1973, 133-44.

36. Jindal MN, Patel VK, Patel VB, Pharmacological actions of aqueous and alcoholic extracts of roots of *C.intybus* Linn, Ind. J. Pharmacol, 7, 3, 1975, 24-3.

37. Hladon H, Dronzda B, Holub M, Szafameck PK, Sesquiterpene lactones (SL) part XXIV. Further studies on cytotoxic activities of SL in tissue cultures of human cancer cells, Pol. J.Parmacol. Pharm. 30, 1978, 611-620.

38. Seto M, Miyase T, Umehara, K, Hirano Y, Otani N, Sesquiterpene lactones from *Cichorium endivia* L and *C.intybus* L. and Cytotoxic activity, Chem. Pharm. Bull, 36, 7, 1988, 2423-2429.

39. Patel VK, Bhatt HV, Toxicity and antiseptic effect of chicory root extract in pyorrhea, The Antiseptic, 1985, 904-906.

40. Sultana S, Perwaiz S, Iqbal M, Athar M, 1995, Crude extracts of hepatoprotective plants.

41. Cadot P, Kochuyt AM. Inhalative occupational and ingestive immediate-type allergy caused by Chicory (*Cichorium intybus*), Clin. Expt. Allergy, 26, 8, 1996, 940-944.

42. Gadgoli C and Mishra BH, Antihepatotoxic activity of *Cichorium intybus*, J.Ethnopharmacol, 58, 2, 1997, 131-4.

43. Gadgoli C and Mishra SH, Preliminary screening of *Achillea millefolium*, *Cichorium intybus* and *Capparis spinosa* for hepatotoxic activity, Fitoterapia, 66, 4, 1995, 319-323.

44. Ki CG, Yim DS, Lee SY, Biological activities of root of *Cichorium intybus,* Nat. Prod. Sciences, 5, 4, 1999, 155-158.

45. Bahar A, Khan S, Masood MH, Siddique AH, Anti-hepatotoxic activity of cichotyboside, a sesquiterpene glycoside from the seeds of *Cichorium intybus*, Journal of Asian Natural Products Research, 10, 2008, 218-223.

46. Keshri G, Lakshmi V, Singh MM, Postcoital contraceptive activity of some indigenous plants in rats, Contraception, 57, 5, 1998, 357-60.

47. Saroja S, Padma PR, Radha P, Thilagavati P, Enzymic and non-enzymic antioxidants in *Cichorium intybus*, J Med. and Arom. Plant Sci, 22, 1, 2000, 36.

48. Santosh KM, Rangarajan P, Selvam R, Effect of *C.intybus* Linn, treatment on experimental hyperoxaluric rats. Proceedings of

International Congress on "Ayurveda-2000", Chennai, TN, India, 201.

49. Gazzani G, Daglia M, Papetti A, Gregotti C, *In-vitro* and *ex-vivo* anti- and pro-oxidant components of *Cichorium intybus*, J.Pharm. Biomed, Anal, 23, 1, 2000, 127-133.

50. Amirghofran Z, Azadbakht M, Karimi MH, Evaluation of immunomodulatory effect of five herbal plants, J. Ethnopharmacol, 72, 1-2, 2000, 167-72.

51. Rungeler P, Castro V, Mora G, Goren N, Vichnewski W. Inhibition of transcription factor NF-alpha beta by sesquiterpene lactones: a proposed molecular mechanism of action, Bio.org. Med. Chem. 7, 1999, 2343-2352.

52. Han M Harries, Fletcher ST, Duggan CM, Baker VA, The use of genomics technology to investigate gene expression changes in cultured human liver cells, Toxicol *In Vitro*, 15, 2001, 399–405.

53. Hazra B, Sarkar R, Bhatt. S, Roy P, Tumour inhibitory activity of Chicory root extract against Ehrlich ascites carcinoma in mice, Fitoterapia, 73, 7-8, 2002, 730-3.

54. Zubair A, Rizwan K, Shahid NM, VU, Antioxidant, antimicrobial & Phytochemical analysis of *Cichorium intybus* seeds extract and various organic fractions, Iranian journal of Pharmaceutical Research, 11, 4, 2012, 1145-1151.

55. Molan A.L., Duncan A.J., Barry T.N., McNabb W.C, Effects of condensed tannins and crude sesquiterpene lactones extracted from chicory on the motility of larvae of deer lungworm and gastrointestinal nematodes. Parasitol. Int. 2(3), 2003, 209-218.

56. Sakurai N, Lizuka T, Nakayama S, Vasorelaxant activity of caffeic acid derivatives from *Cichorium intybus* and *Equisetum arvense*. Yakugaku Zasshi, 123(7), 2003, 593-8.

57. Ghaderi R, Zardast M, Hosseini M, Delgir B, Hassanpour M, Comparison of antibacterial effect of *Cichorium intybus* Linn. With vancomycin, Ceftriaxone, Ciprofloxacin & Penicillin (*In vitro*), Clinical and Experimental Pharmacology, 2, 2, 1-3, 2012.

58. Sakurai N, Lizuka T, Nakayama S, Vasorelaxant activity of caffeic acid derivatives from *Cichorium intybus* and *Equisetum arvense*. Yakugaku Zasshi, 123, 7, 2003, 593-8.

59. Zimmerman MD, Hayman J, Function and integrity of the liver, In: Clinical diagnosis and management by laboratory methods, 17, 217-250.

60. Agarwal AK, Mehendale, Potentiation of carbon tetrachloride hepatotoxicity and lethality by chlordecone in female rats. Toxicology, 26, 1983, 231-42.

61. Handa SS, Sharma A, Hepatoprotective activity of Andrographolide from *Andrographis paniculata* against carbon tetrachloride, Indian Journal of Medical Research, 92, 2008, 276-292.

62. Keppler D, Lesch R, Reutter W, Decker K, Experimental hepatitis induced by D-galactosamine, Experimental and molecular pathology, 9, 1968, 279-290.

63. Paronetto F., Immunologic reactions in alcoholic liver disease, Semin Liver Dis, 13, 1993, 183-195.

64. Nevin KG, Vijayammal PL, Effect of *Aerva lanata* against hepatotoxicity of carbon tetrachloride in rats, Environmental Toxicology and Pharmacology, 20, 2005, 471-477.

65. Chen, CJ, Deng AJ, Liu C, Shi R, Qin HL, Wang AP, Hepatoprotective Activity of *Cichorium endivia* L. Extract and Its Chemical Constituents. *Molecules*, 16, 2011, 9049-9066.

66. Yeh SF, Gupta M, Sarma DNK, Mitra SK, Down regulation of hepatitis B surface antigen expression in human hepatocellular carcinoma cell lines by HD-03, a polyherbal formulation, Phytother. Res., 17, 2003, 89–91.

67. Vidyashankar A, Patki PS, Liv.52 attenuate copper induced toxicity by inhibiting glutathione depletion and increased antioxidant enzyme activity in HepG2 cells, Food and chemical toxicology, 48, 7, 2010, 1863-68.

68. Gosh N, Gosh R, Vivekananda M, Mandal SC, Recent advances in herbal medicine for treatment of liver diseases, Pharmaceutical Biology, 49, 9, 2011, 970-988.

69. Zhang Hong Li, Ling-Hao Dai, Yi-Ha Wu,X iaoPi Yu,Yong-Yong Zhang, RongFa Guan, Ta o Liu,and Jun Zhao, Evaluation of Hepatocytoprotective and Anti-hepatitis B Virus Properties of Cichoric Acid from *Cichorium intybus* leaves in Cell Culture, Biol. Pharm. Bull, 37, 7, 2014, 1214–1220.

70. Li GY, Gao HY, Huang J, Lu J, Gu JK, Wang JH. Hepatoprotective effect of *Cichorium intybus* L., a traditional Uighur medicine, against carbon tetrachloride-induced hepatic fibrosis in rats. *World Journal of Gastroenterology , WJG*. 2014, 2016, 4753-4760.

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