

The contents and pharmacology of *Crotalaria juncea*- A review

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Abstract: The preliminary phytochemical screening of the *Crotalaria juncea* leaves revealed the presence of carbohydrates, steroids, triterpenes, phenolics, flavonoids, alkaloids, aminoacids, saponins, glycosides, tannins and volatile oils. The plant possessed hypolipidemic, reproductive, antioxidant, antibacterial, antifungal, anti-diarrhoeal, anti-inflammatory, hepatoprotective, and many other pharmacological effects. This review was designed to highlight the chemical constituents and pharmacological effects of *Crotalaria juncea*.

Keywords: constituents, pharmacology *Crotalaria juncea*.

I. INTRODUCTION

Herbal medicine is the oldest form of medicine known to mankind. It was the mainstay of many early civilizations and still the most widely practiced form of medicine in the world today⁽¹⁾. Plants generally produce many secondary metabolites which are bio-synthetically derived from primary metabolites and constitute an important source of many pharmaceutical drugs⁽²⁻⁶⁰⁾. The preliminary phytochemical screening of the *Crotalaria juncea* leaves revealed the presence of carbohydrates, steroids, triterpenes, phenolics, flavonoids, alkaloids, aminoacids, saponins, glycosides, tannins and volatile oils. The plant possessed hypolipidemic, reproductive, antioxidant, antibacterial, antifungal, anti-diarrhoeal, anti-inflammatory, hepatoprotective, and many other pharmacological effects. This review will highlight the chemical constituents and pharmacological effects of *Crotalaria juncea*.

II. PLANT PROFILE:

Synonymas:

Crotalaria benghalensis Lam., *Crotalaria cannabinus* Royle, *Crotalaria fenestrata* Sims, *Crotalaria ferestrata* Sims, *Crotalaria porrecta* Wall., *Crotalaria sericea* Willd., *Crotalaria tenuifolia* Roxb. and *Crotalaria viminea* Wall⁽⁶¹⁻⁶²⁾.

Taxonomic classification:

Kingdom: Plantae; **Subkingdom:** Tracheobionta; **Superdivision:** Spermatophyta; **Division:** Magnoliophyta; **Class:** Magnoliopsida; **Subclass:** Rosidae; **Order:** Fabales; **Family:** Fabaceae / Leguminosae; **Genus:** *Crotalaria*; **Species:** *Crotalaria juncea*⁽⁶³⁾.

Common names:

Arabic: Kinab, *Crotalaria*, **Bengali:** Ghore Sun, Shon, Shonpat; **Chinese:** Shu ma, Yin tu ma; **English:** Benares Hemp, Bengal hemp, Bombay hemp, Brown Hemp, Cascavelle, Cocosnut, Grand Sonnette, Grand Tcha-Tcha, Indian Hemp, Jubbalpore Hemp, Madras hemp, San Hemp, Sann Hemp, Sonnette, Sun Hemp, Sunn *Crotalaria*, Sunn Hemp, Tcha-Tcha. **French:** Chanvre du Bengale, *Crotolaire jonciforme*; **India:** San, Sunn; **Indonesia:** Orok-orok lembut; **Persian:** San; **Portuguese:** Cãhãmo-da-Índia, *Crotalaria*; **Russian:** Krotalyariya, Sitnikovaya; **Spanish:** Cãñamo De La India, Cãñamo San; **Swedish:** Sunnhampa; **Vietnamese:** Cãy Mung, Suc sat, Luc lac⁽⁶⁴⁻⁶⁵⁾.

Distribution:

The plant is native in Asia especially Asia tropical (Bangladesh; Bhutan; India). It is now widely cultivated in the drier areas of the tropics and subtropics and in many temperate areas with a hot summer. It often escaped from cultivation, naturalizes easily and grows in many areas as a ruderal plant. *Crotalaria juncea* is recorded in many countries across the African continent from the Atlantic coast to the Red Sea, from Tunisia to South Africa and in the Indian Ocean islands⁽⁶⁴⁻⁶⁵⁾.

III. DESCRIPTION:

Sunn hemp is a short-day, erect shrubby annual, generally 1 to 4 m in height. Stems up to 2 cm in diameter, cylindrical and ribbed with short appressed hairs. Leaves alternate, simple; stipules 1–2 mm long, slender; petiole 3–5 cm long; leaflets oblong-lanceolate, 5–13(–18) cm × 0.5–3 cm, finely appressed pubescent. Inflorescence a leaf-opposed raceme 10–50 cm long, laxly 6–20-flowered; bracts elliptical, 3–5 mm long. Flowers bisexual, zygomorphic, 5-merous; calyx 1.5–2 cm long, covered in short brown hairs with some longer ones interspersed, lobes 3–4 times as long as the tube; corolla bright yellow, with elliptical standard faintly reddish marked or tinged, wings a little shorter than keel, keel 17–22 mm long, with a long, slightly incurved

twisted beak; stamens 10, all joined in a sheath open at base; ovary superior, 1-celled, style curved, stigma small. Fruit a cylindrical pod 30–55 mm × 12–17 mm, short, velvety hairy, 6–12-seeded. Seeds oblique-cordiform, 6–7 mm long, dark brown to black⁽⁶⁶⁻⁶⁸⁾.

IV. TRADITIONAL AND ECONOMIC USES:

The major significance of *Crotalaria juncea* lies in its valuable bast fibre, which makes up about 8% of the dry stem weight. The fibre of commerce consists of greyish to pale yellow strands 75–150 cm long. Fibres are entangled in a mesh structure, and single filaments are obtained by combing and splitting the mesh structure. The ultimate fibre cells are (0.5–)6–8(–20) mm long and (10–)25–30(–50) µm wide, with a cell wall thickness of 3–11 µm. The fibre has a tensile strength of c. 73 kg/mm² and an elongation at break of 5.5%. The fibre is stronger when wet than when dry, and is fairly resistant to mildew, moisture and microorganisms in salt water, making it particularly suitable for fishing nets and marine cordage. The fibre contains 10% moisture, 67.8% cellulose, 16.6% hemi-celluloses, 3.5% lignin, 0.3% pectin, 1.4% water solubles and 0.4% fat and wax. The fibre possesses properties that make it an excellent choice for papermaking. Prepared pulps are suitable for a wide range of end use. The fibres are particularly suitable for cigarette paper because of the high cellulose and low ash content. The dried stalks and hay are used as forage⁽⁶⁴⁾.

It is used for medicinal, edible and culinary purposes by many tribal communities. It is traditionally used as blood purifier, abortifacient, astringent, demulcent, emetic, purgative, in the treatment of anaemia, impetigo, menorrhagia and psoriasis⁽⁶⁹⁻⁷²⁾.

Parts used: Roots, seeds and leaves⁽⁷⁰⁻⁷²⁾.

Physicochemical properties:

The physicochemical properties of *Crotalaria juncea* leaves (% w/w) were: total ash value 5.9, acid insoluble ash 2.7, water soluble ash 3.9, sulphated ash 5.1, moisture content 11, foreign matter 0.04, alcohol soluble extract value 5.84, water soluble extract value 20.4 and crude fiber content 52.6⁽⁷³⁻⁷⁴⁾.

Physicochemical properties of *Crotalaria juncea* oil: refractive index(Abbes 21C°): 1.47, specific gravity 21C°): 0.8832, color (lovibond): Y=10.1, R=0.3, acid value: 2.7, saponification value: 217, iodine value: 119.71 and maleic anhydride value: 8.04⁽⁷⁵⁾.

Chemical constituents:

The preliminary phytochemical screening of the *Crotalaria juncea* leaves revealed the presence of carbohydrates, steroids, triterpenes, phenolics, flavonoids, alkaloids, aminoacids, saponins, glycosides, tannins and volatile oils^(73,76-78). Riddelline, seneciphylline, senecionine, trichodesmine, chodesmine alkaloids, galactose-specific lectin and cardiogenin 3-O- [β]-d-xylopyranoside were also isolated from *Crotalaria juncea*⁽⁷⁹⁻⁸¹⁾.

Seeds of *Crotalaria juncea* contained 0.074% of toxic dehydropyrrolizidine alkaloids (DHPAs) (isohemijuncines 0.05%, trichodesmine 0.016%, and junceine 0.008%)⁽²²⁾. *Crotalaria juncea* seeds contained 45.19% carbohydrates, 36.43% protein, 4.22% oil, 10.85% moisture and 3.31% ash. Seed lipids and proteins of *Crotalaria juncea* were analyzed for fatty acids and amino acids composition. Gas chromatographic analysis of the oil gave palmitic acid (16.01-18.09%), stearic acid (7.29-10.15%), oleic acid (6.69-14.41%), linoleic acid (54.44-62.36%), linolenic acid (0.7-7.86%), myristic acid (0.197%), arachidic acid (1.199%) and behenic acid (1.369%)^(75, 83).

The defatted seed cake contained all the essential amino acids except methionine and six non-essential amino acids. The percent composition of amino acids (g/100g): essential amino acids (isoleucine 1.17, leucine 2.10, lysine 1.67, phenylalanine 0.92, threonine 0.88, tryptophan 0.53, tyrosine 0.78 and valine 0.96); and non-essential amino acids (alanine 2.12, arginine 2.72, glutamic acid 9.45, glycine 1.53 and proline 1.10)⁽¹⁵⁾. Unusual amino acid, 2-amino-5-hydroxyhexanoic acid was also isolated from the seeds of *Crotalaria juncea*⁽⁸⁴⁾.

The plant fibre contained 10% moisture, 67.8% cellulose, 16.6% hemi-celluloses, 3.5% lignin, 0.3% pectin, 1.4% water solubles and 0.4% fat and wax. The stems contained: cellulose 78.3%; pentosan 3.6%; urinic anhydrite 1.7%; acetyl content 1.5% and lignin 4.0%. Minor constituents included: fat and wax 0.5%; nitrogenous matter 1.4% and ash 0.3%. Monosaccharide constituents of the plant included: glucose 80.3%; xylose 5.2%; mannose 11.7%; galactose 2.1%; arabinose 1.7% and rhamnose 0.4%. Dried stalks of the plant, for cattle feed contained 14.4% moisture, 1.1% ether extract, 11.3% albuminoids, 35.8% carbohydrate, 27.4% woody fibre, and 6.4% soluble mineral matter. Seeds contained 8.6% moisture, 34.6% crude protein, 4.3% fat, 41.1% starch, 8.1% fibre, and 3.3% ash. Seeds were reported to contain trypsin inhibitors, and were said to be poisonous to cattle. Seeds oil contained 46.8% linoleic acid, 4.6% linolenic acid, and 28.3% oleic acid, and 20.3% saturated acids. The seeds also contained the toxic pyrrolizidine alkaloids trichodesmine, junceine, senecionine and seneciphylline and 25.6% of the polysaccharide galactomannan^(64, 85).

A galactose-specific lectin from seeds of *Crotalaria juncea* has been purified by fractional precipitation with ammonium sulfate followed by biospecific affinity chromatography and preparative

isoelectric focusing. The adsorbent was prepared by coupling galactose to Sepharose 6B activated with divinyl sulfone. The lectin was homogeneous as judged by ultracentrifugation and by electrophoresis in cellulose acetate strips and in polyacrylamide gradient gel. Its isoelectric point is pH 8.8 and the molecular weight is about 120 000. It was a glycoprotein containing 9.8% carbohydrate (mannose, N-acetyl-D-glucosamine, fucose, and xylose). The lectin contained 3.2 mol Ca²⁺, 2.2 mol Mg²⁺ and 0.2 mol Mn²⁺ per 120 000 g. No sulphur-containing amino acids were detected⁽⁸¹⁾.

V. PHARMACOLOGICAL EFFECTS:

Hypolipidemic effect:

The antihypercholesterolemic effects of 50 and 100 mg/kg bw per day of an ethanolic extract of *Crotalaria juncea* Linn (whole plant) were investigated in rats fed high-fat diet by evaluating food consumption, weight gain, fecal fat excretion, serum and liver lipids, and biochemical profiles as well as by histopathological studies. The results were compared to animals fed with the standard diet and animals fed with a high-fat diet and atorvastatin (10 mg/kg bw). The animal group administered with the ethanolic extract for 35 days showed decreased levels of TC, LDL, VLDL, TG, HDL+VLDL, VLDL+LDL, LDL/TC, AI, SGOT, SGPT, and elevated levels of HDL, HDL/TC, significantly ($p < 0.01$ and $p < 0.05$) in a dose-dependent manner. The evaluation of liver tissues of the animal groups treated with the herbal extract and standard, showed increased levels of SOD, GSH, and catalase, whereas levels of SGOT, SGPT, total glucose, HMG-CoA, lipase, amylase, and the percentage of malon-dialdehyde were decreased when compared with the high-fat-diet fed rats. Body weight and food intake in the treated groups were significantly lower than that in the control⁽⁸⁶⁾.

The antihyperlipidemic activity of alcoholic and methanol extract of leaves of *Crotalaria juncea* (CJ) was investigated against Triton induced hyperlipidemia in mice. CJ was administered at a dose of 100 and 200mg/kg (po) to Triton induced hyperlipidemic mice. Atorvastatin was used as reference standard. CJ showed a significant decrease in the levels of serum total cholesterol, triglyceride, LDL, VLDL and significant increase in the level of serum HDL at the dose of 100 and 200mg/kg (po) against Triton induced hyperlipidemia in mice⁽⁸⁷⁾.

The unusual amino acid, 2-amino-5-hydroxyhexanoic acid isolated from the seeds of *Crotalaria juncea*, showed dose dependent lipid lowering activity in the *in vivo* experiments and also showed good *in vitro* antioxidant activity. The cyclized compound, 3-amino-6-methyltetrahydro-2H-pyran-2-one showed better lipid lowering and antioxidant profile than the parent compound⁽⁸⁴⁾.

Anti-obesity effect of *Crotalaria juncea* leaves extract was documented in high fat induced obesity in rats⁽⁸⁸⁾.

VI. EFFECTS ON REPRODUCTIVE SYSTEMS:

The antifertility activity of various extracts of *Crotalaria juncea* seeds was studied in male mice. Adult male mice were gavaged the petroleum ether, benzene and ethanol extracts of *Crotalaria juncea* seeds, 25 mg/100mg/day for 30 days. On day 31 the animals were sacrificed by cervical dislocation and the testes, epididymis, vas deferens, seminal vesicles, prostate gland, bulbourethral gland and levator ani were dissected out and weighed. The organs were processed for biochemical and histological examination. In petroleum ether, benzene and ethanol extracts treated rats, there was a decrease in the weights of testis and accessory reproductive organs.

The diameters of the testis and seminiferous tubules were decreased. Spermatogonia, spermatocytes and spermatids in the testis and the sperm count in cauda epididymis were also decreased. There was a significant reduction in the protein and glycogen contents and an increase in the cholesterol content in the testis, epididymis and vas deferens. Of the 3 extracts, the ethanol extract appeared to be the most potent antispermatogenic extract. When the ethanol extract was tested in immature male mice, it exerted antiandrogenic effect as the weights of accessory organs were reduced⁽⁸⁹⁾.

Petroleum ether, benzene and ethanolic extracts of *Crotalaria juncea* seeds were administered intraperitoneally at the dose level of 25 mg/100 g body weight to albino male mice for 30 days. The results showed decreased number of spermatogonia, spermatocytes and spermatids in testis along with reduced caudal spermatozoa. Biochemical observations indicated increased level of cholesterol and significant reduction in protein and glycogen content. The increased cholesterol content along with degeneration of Leydig cells indicated inhibition of steroidogenesis. The decrease in the weight of accessory reproductive organs further attributes lowered availability of androgens due likely to inhibition of steroidogenesis. Out of three extracts, ethanolic extract seems to be more potent in antispermatogenic and antisteroidogenic activities. When ethanolic extract was tested in immature mice for androgenic activity, it showed its antiandrogenic potency as the weight of accessory sex organs were reduced⁽⁹⁰⁾.

Petroleum ether, benzene and alcohol extracts of seeds of *Crotalaria juncea* administered orally at the dose level of 25mg/100g bw to adult female mice for 30 days, resulted in irregular estrous cycle with prolonged estrus and metaestrus and reduced diestrus and proestrus during the experimental period. Histological studies of

the ovary indicated increases in the number of atretic follicles but decreases in the number of developing follicles, Graafian follicles and corpora lutea. The total cholesterol content of the ovary was increased, whereas ascorbic acid content is decreased. The weight of the uterus and its micrometric measurement in all experimental mice were increased significantly. The alcoholic extracts showed estrogenic activity in immature mice by early opening of the vagina, premature cornification of the vaginal epithelium and increases in uterine weight. However, alcohol extract of seeds of *Crotalaria juncea* was more effective in causing these changes compared to other extracts⁽⁷⁸⁾.

The ethanol extract of *Crotalaria juncea* seeds which showed promising antiovarulatory activity in female albino rats was examined for the isolation of its active fractions. Two fractions were obtained using thin layer chromatography (TLC). Both fractions were subjected for testing their anti-ovulation activity and the effect on estrous cycle in rats. After preliminary trials, the fraction I (200mg/kg body weights) showed maximum antiovarulatory activity when administered orally to the rats for 30 days. Decreased number of healthy follicles (Class I – Class VI) and corpora lutea and increased number of regressing follicles (Stage IA, Stage IB, Stage IIA, Stage IIB) were observed in the ovary after 30 days treatment. The treatment caused an increase in the cholesterol level and acid/alkaline phosphatase activity and a decrease in protein and glycogen contents of the ovary. Estrous cycle was affected as a significant increase in estrus and metaestrus phases with a decrease in diestrus and proestrus phases in the treated groups during experimental period of 30 days⁽⁹¹⁾.

Petroleum ether, benzene and alcohol extracts of the seeds of *Crotalaria juncea* were tested for antiimplantation and pregnancy interruption activities in female albino rats. Of these three extracts, the alcohol extract was found to be the most effective in causing antiimplantation and pregnancy interruption activities. These adverse effects on fertility were reversible upon withdrawal of the extract treatments. The alcohol extract was found to possess estrogenic activity⁽⁷⁶⁾.

VII. ANTIOXIDANT EFFECTS:

Antioxidant activity of *Crotalaria juncea* extracts were studied in goat liver lipid peroxidation, linoleic acid emulsion, α -amylase and lipase inhibitory activity. All the extracts had shown antioxidant property, α -amylase, and lipase inhibitory properties. Aqueous extract was found to show maximum antioxidant activity on goat liver. Antilipid peroxidation and antioxidant activity were determined as $66.94 \pm 0.616\%$ ($p < 0.01$) and $59.54 \pm 0.2\%$ ($p < 0.01$), respectively. Maximum α -amylase and lipase inhibitory activities of $71.42 \pm 1.37\%$ ($p < 0.01$) and $57.14 \pm 2.74\%$ ($p < 0.01$), respectively, were exhibited by macerated methanol extract⁽⁹²⁾.

The antioxidant activity of *Crotalaria juncea* seed oil (CJSPE) was evaluated by *in vitro* assay methods (2,2-Diphenyl-1-picrylhydrazyl (DPPH), hydroxyl and superoxide radical scavenging activity) of CJSPE; its antioxidant activity was found to be concentration dependent and IC₅₀ values were 132.31, 286.409 and 31.254 g/ml by the tree tests, respectively. Moreover, CJSPE has displayed dose dependant, significant inhibition of NO production in the isolated rat peritoneal macrophages⁽⁸³⁾.

VIII. ANTIBACTERIAL AND ANTIFUNGAL EFFECTS:

The ethanol extract of flowers part (CJFEE) and seeds part (CJSEE) were evaluated for the antibacterial activity by the agar disc diffusion method against *C. freundii*, *E. coli*, *E. faecalis*, *K. pneumonia*, *P. aeruginosa*, *S. flexneri*, *S. aureus*, *S. dysenteriae* and *V. cholerae*. Results revealed that CJSEE possess significant antibacterial activity against the *E. coli*, *K. pneumonia*, *P. aeruginosa*, *S. aureus* and *V. cholerae*. However, the ethanol extract of seeds part had higher antibacterial than ethanol extract of flower parts of *Crotalaria juncea*⁽⁷⁷⁾.

The antibacterial activity of *Crotalaria juncea* seed oil (CJSPE) was evaluated by the disc diffusion method against *E. faecalis*, *S. aureus*, *E. coli*, *K. pneumonia*, *P. aeruginosa*, *S. flexneri*, *S. dysenteriae* and *V. cholerae*. Results showed that CJSPE have good antibacterial activity against the *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia* and *Shigella flexneri*. However, the zone of inhibition showed by CJSPE was found less than that of ciprofloxacin (5 μ g/disc) used as standard⁽⁸³⁾.

Antibacterial activity of crude extracts prepared in sodium phosphate buffer against *Xanthomonas* strain was studied. There has been found a highly strong activity of *Crotalaria juncea* extracted in sodium phosphate buffer against plant bacterial pathogen, *Xanthomonas oxanopodis* pv. *Punicae*⁽⁹³⁾.

Moderate antifungal activity has been reported in the methylene chloride and methanol extract of aerial parts of *Crotalaria juncea* of Indonesian origin⁽⁹⁴⁾.

IX. ANTI-DIARRHOEAL EFFECTS:

The anti-diarrhoeal effects of methanolic extract of leaves of *Crotalaria juncea* (MECJ) was studied against castor oil-induced diarrhoea model and small intestine transit model in rats. The number of droppings and the distance traveled by charcoal in intestine were measured. MECJ at the doses of 200 and 400 mg/kg

significantly inhibited ($P < 0.001$) the castor oil induced charcoal meal transit. The MECJ showed marked reduction in the frequency of bowel motion as well as a modest reduction in intestinal transit⁽⁹⁵⁾.

X. ANTI-INFLAMMATORY EFFECT:

Anti-inflammatory effect of the *Crotalaria juncea* seed oil (CJSPE) was assessed by its effect on NO radical production in isolated macrophages from rat peritoneal (*in vitro* method); and carragennan-induced paw edema rat model and cotton pellet-induced granuloma formation in rat model (*in vivo* method). The result showed a dose dependant reduction of carragennan-induced rat paw edema by the CJSPE. Moreover, significant ($p < 0.001$) anti-inflammatory activity was displayed by CJSPE (200 mg/kg) in the late phase of inflammation; and the effect was comparable to that of diclofenac sodium. CJSPE was also found to be effective in the reduction of size ($48.55 \pm 0.244\%$) of granuloma formation and effect was nearly equal to that of diclofenac sodium⁽⁸³⁾.

The antiarthritic activity of ethanolic extract of the leaves of *Crotalaria juncea* (CJE) in complete Freund's adjuvant (CFA) induced arthritis model in rats, and also the anti-ulcerogenic activity of CJE was evaluated. Treatment with CJE at 200 and 400 mg/kg and standard indomethacin (0.3 mg/kg) was started on the same day and continued up to day 12. The paw volume was measured on day 1, 5, 12 and 21 for both the paws and antiarthritic activity. The drug CJE produced reduction in the inflammation of the paw produced by CFA. The antiarthritic action started on the day 5 and continued till day 12 and the activity was comparable to that of the standard on both days. In indomethacin treated animals, gastric ulcer was observed, while, CJE was found to protect the animals from ulcer formation. The authors concluded that CJE significantly inhibited adjuvant induced arthritis and has significant anti-inflammatory effect ($p < 0.001$). It has anti-ulcerogenic property compared to indomethacin, which may be due to appetite suppressant activity⁽⁹⁶⁾.

XI. HEPATOPROTECTIVE EFFECT:

The petroleum ether extract of *Crotalaria juncea* seed at low and high dose (100 and 500mg/kg) were tested for its efficacy against thioacetamide induced acute hepatic damage in rats. The different groups of rats were administered with thioacetamide (100mg/kg, sc). Drug Silymarin (100 mg/kg,) was used as reference standard. The rats were monitored for biochemical changes of serum glutamate oxaloacetate transaminase, serum glutamate pyruvate transaminase, serum alkaline phosphatase, and bilirubin (total and direct). Activity of antioxidant enzymes such as superoxide dismutase and catalase in liver tissue homogenate and Histopathological changes were observed. According to the results, it was proved that the *crotalaria juncea* seed extract (CJSE) possessed hepatoprotective potency in a dose dependent manner by reducing the elevated levels of marker enzymes and by increasing the decreased antioxidant enzyme activity⁽⁹⁷⁾.

XII. OTHER EFFECTS:

Recently, transgenic plants expressing immunogenic proteins of foot-and-mouth disease virus (FMDV) have been used as an oral or parenteral vaccines against foot-and-mouth disease (FMD). They exhibit advantages like cost effectiveness, absence of processing, thermostability, and easy oral application. FMDV VP1 protein of single serotype has been mostly used as immunogen. A bivalent vaccine with tandem-linked VP1 proteins of two serotypes, A and O, present in transgenic forage crop *Crotalaria juncea*. The expression of the bivalent protein in the transgenic plants was confirmed by Western blot analysis. Guinea pig reacted to orally or parenterally applied vaccine by humoral as well as cell-mediated immune responses including serum antibodies and stimulated lymphocytes, respectively⁽⁹⁸⁾.

XIII. CONTRAINDICATION AND TOXICITY:

Acute toxicity of ethanolic extract of the leaves of *Crotalaria juncea* was carried out in rats, it was found to be safe up to 2000 mg/kg body weight⁽⁹⁶⁾. However, Barethia, found that LD₅₀ of seed extract of *Crotalaria juncea* in rats was 0.681 (0.400- 1.160) g/Kg (4.086 g/m^2)⁽⁹⁹⁾. However, as a result of sunn hemp seeds contents of several pyrrolizidine alkaloids, it was toxic to animals and birds when ingested in sufficient amount⁽¹⁰⁰⁾. Seeds of *Crotalaria juncea* contained 0.074% of toxic dehydropyrrolizidine alkaloids (DHPAs) (isohemijunceines 0.05%, trichodesmine 0.016%, and junceine 0.008%)⁽⁸²⁾.

The effect of the ethanolic extract of *Crotalaria juncea* seeds has been assessed on liver, kidney, spleen and adrenals of adult rats. Results revealed that its administration at a dose of 200 mg/kg caused significant alterations, wet weight of the organs was reduced, protein and glycogen contents in all the organs were decreased significantly, whereas, the activity of acid and alkaline phosphatase was increased. Histology revealed remarkable disintegration necrosis and degeneration in the liver. Renal tubular cells showed degeneration and exfoliation. Adrenals showed hypertrophy in the region of zona glomerulosa. In the spleen, the number of megakaryotic cells and lymphocytes were increased⁽¹⁰¹⁾.

Extracts of roots, leaves, stems and seeds were analyzed for the presence of toxic dehydropyrrolizidine alkaloids. The alkaloids occurred mainly as N-oxides in roots, stems and leaves, but mainly as free bases in seeds. Because of human toxicity, it was necessary to examine animal-derived products (meat and milk), bee products (honey and pollen) and seed contamination of grain products^(80,102).

The effects and susceptibility of donkeys to *Crotalaria juncea* poisoning were determined at high and low doses. Seeds of *Crotalaria juncea* containing 0.074% of dehydropyrrolizidine alkaloids (DHPAs) (isohemijunceines 0.05%, trichodesmine 0.016%, and junceine 0.008%) were administered to three donkeys at 0.3, 0.6 and 1 g/kg bw daily for 365 days. No clinical signs were observed, and on liver and lung biopsies, and showed only mild liver megalocytosis in the donkeys ingesting 0.6 and 1 g/kg/day. Two other donkeys that received daily doses of 3 and 5 g seed/kg showed initial respiratory signs 70 and 40 days after the start of the administration, respectively. The donkeys were euthanized following severe respiratory signs and the main lung lesions were proliferation of Clara cells and interstitial fibrosis⁽⁸²⁾.

Twenty horses were died 30 days after being fed a diet containing 40% of triturated *Crotalaria juncea* seeds. Before death, they had staggering, dyspnea and fever. At necropsy the most evident lesions were areas of lung parenchyma consolidation and enlarged and congested livers. Histopathological examination revealed diffuse fibrosing alveolitis with hyaline membranes, suggesting a blood-borne insult, and passive congestion in the liver with compression of the hepatocyte trabecules. To confirm the diagnosis, guinea pigs were given 60% of a commercial diet + 40% triturated *Crotalaria juncea* seeds. After 4 months of feeding the animals died with dyspnea. Their lungs had diffuse fibrosing alveolitis with discrete formation of hyaline membranes and the livers were congested. These results supported the diagnosis of *Crotalaria juncea* intoxication in the horses⁽¹⁰³⁾.

XIV. CONCLUSION

The paper reviewed *Crotalaria juncea* as promising medicinal plant with wide range of pharmacological activities which could be utilized in several medical applications because of its effectiveness and safety.

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