2. REVIEW OF LITERATURE

Immense works have been, so far, carried out by many workers on various aspects of *Senna obtusifolia*, as well as other congeneric species. The fields of studies encompass taxonomy, phenology, morphology, cytology, biochemical analyses, in general, and also dealing with the active principle, like rhein etc. and matters related to their medicinal efficacy. A comprehensive account thereof has been presented below, which is quite axiomatic in presenting the significance of the concerned species and some of its related members.

2.1 Taxonomic Treatments

The genus *Senna* Mill., is a widespread, heterogenous and diversified genera of the Fabaceae or Leguminosae family. Phylogenetic analysis reveals that Leguminosae is a monophyletic family (Bruneau et al., 2001). The subfamily Caesalpinioideae of family Leguminosae holds a 'basal' paraphyletic assemblage in phylogenetic trees and shows large diversity in floral form and ontogeny (Doyle et al., 2000; Bruneau et al., 2001). Leguminosae is also an economically important family and remains most poorly understood taxonomically and phylogenetically. The subfamily Caesalpinoideae comprises approximately 160 genera (Bruneau et al., 2001). Caesalpinoideae comprising of Cercideae, Macrolobieae, Caesalpineae, Detarieae, and Cassieae tribes included by Tucker (2003). The tribe Cassieae was considered as a polyphyletic one though several infratribal groupings supported its monophyletic origin by trnL analysis. Irwin and Barneby (1981) considered Cassieae as an artificial one consisting of five subtribes, namely, Ceratoniinae, Labicheinae, Dialiinae, Duparqueniiae, Cassiinae, which by trnL analysis supported the monophyly of the subtribe. The subtribe Cassiinae is distinguished by abaxial nature of the sepals. Phylogenetic analysis revealing the

sequence of nucleotides (trnL) promoted trifurcation of *Cassia, Senna, Chamaecrista* to subtribe Cassiinae (Bruneau et al., 2001).

In the improvised system of classification, *Cassia* L. genus was raised to Cassiinae subtribe level by Irwin and Barneby (1981; 1982), and the formation of the genus *Cassia* L. or *Cassia* sensu lato into *Senna* Mill., *Chamaecrista* Moench, and *Cassia* sensu stricto (Lewis et al., 2005) has been acknowledged by taxonomical studies based different information, such as seed protein (Guareeb et al., 1999), reproductive, morphological and vegetative characters (Owens and Lewis 1989; Boonkerd et al. 2005), ontogenetic characteristics (Tucker 1996), molecular systematics (Bruneau et al., 2001), and cytogenetics (Elaine et al., 2005). Separation of *Senna* was confirmed further by phenetic studies (Boonkerd et al., 2005), structural (Endress, 1996), and taxonomic (Randell, 1990; Singh 2001) studies. Several ventures had been undertaken to scrutinize the taxonomic status of the specified trifurcated genera. Inflorescence, floral organ initiation and development have been researched and correlated by Tucker (1996) with an outcome of trifurcation of genera. Ghareeb et al. (1999) supported the isolation based on chromosomal number, morphological, and seed protein characteristics. The genus has approximately 350 species (earlier 600 species in *Cassia* L.) with a high diversity habits including lianas, shrubs, and sub-shrubs or undershrubs, trees, treelets, and herbs successfully colonizing in extensive habitats comprising of various climates and latitudes (Marazzi et al., 2006). However, it is largely diverse in regions of varied topography which have distinct seasonal climates (Acharya et al., 2011).

Senna is a paraphyletic genus having six divisions: *Chamaefistula, Psilorhegma, Senna, Astroites, Paradyction*and *Peiranisia* (Irwin and Barneby, 1982). *Senna* Mill.,

considered paraphyletic, is the largest among Caesalpinioideae and a part of thetwenty-five largest genera of dicotyledonous plants (Acharya et al., 2011; Dave and Ledwani, 2012; Resende et al., 2013).

The genus is characterised on the basis of distinct floral morphology along with extrafloral nectaries (Marazzi et al., 2006). This shifting of *Senna* marks the history of traditional systematics and taxonomy. These shifts are explained by morphological variation of *Senna* with an immense level of specialization in buzz pollinated *Senna* flowers which can unconditionally be used for taxonomic purposes. Flowers of the genus are bright yellow and nectarless which offers the pollens as a prize to their pollinators (Gottsberger and Silberbauer-Gottsberger, 1988). Ten stamens are usually present in the *Senna* flowers which are heterantherous; among which seven anthers are fertile and the rest are staminodial. All the fertile anthers showing poricidal opening are separated into two sets. The first set containing four stamens in the middle presented between the abaxial stamens and the adaxial staminodes on which buzing bees extract food as pollen. The next one containing 2-3 abaxial stamens of longer size. During buzzing, their pollens are placed on body surface of bees which finally elated to another flower's stigma (Carvalho and Oliveira, 2003). Asymmetric flowers are also present in the species of *Senna* (Gottsberger and Gottsberger-Silberbauer, 1988).

Senna of Fabaceae, distributed primarily in the Australian, African and American continents, is considered as a diverse genus within the family (Marazzi et al. 2006). *S. obtusifolia* and *S. tora* of the genus *Senna* is clearly distinguished by Linnaeus (1753). As both the species are congeneric and very much phenotypically similar, some confusions prevail regarding the differentiation of *S. obtusifolia* and *S. tora*. Both the species are mostly alike with morphologically overlapping characters (Tripathi et al.,

2009) and differentiated by cautious observations and can be called as congeneric species. Brenan (1958) stated that both are distinct and clearly separated and can be distinguished regarding the shape of anthers, areoles, and seed testa. The seed areoles observed in *S. tora* on the side of the seed is around 1.5-2.0 mm while in *S. obtusifolia* it is around 0.3- 0.5mm (Brenan, 1958). Singh (1978) supporting Brenan's view added that seed testa in *S. obtusifolia* is not distinctly veined and slightly muricated while in *S. tora* the seed testa not muricated, but distinctly veined. According to Retzinger (1984), the presence of a single extrafloral nectaries or gland or stipel present between the lowest leaflet pair in *S. obtusifolia* which distinguishes *S. tora* by the presence of biglandular or two extrafloral nectaries or two stipels between the lower pair of the leaflets. Waterhouse and Norris (1987) pointed out that the presence of foetid smell arising from the crushed foliage of *S. obtusifolia* which differs it from *S. tora*.

According to Randell (1988), *S. obtusifolia* possess petioles with length 1.5-2 cms, pedicels of around 2-3 cms in length, short beaked anthers and non-longitudinal narrow seed areole. On the contrary, *S. tora* possess petioles with length of 2-4.5 cms, pedicels of 1.5 cms in length, anthers truncate and seed aeroles longitudinal and broad. Literature reported the similarity of *S. obtusifolia* with a Vietnamese *Cassia tora* (Poethke et al., 1968). However, literature also reported their differences in chemical makeup (Koshioka et al., 1978). It is reported that *S. tora* can be derived from *S. obtusifolia* as various biochemical compounds present in the latter (Upadhyaya and Singh, 1986). Thus, in the evolutionary process, *S. obtusifolia* may be responsible for the origin of *S. tora* (Cock and Evans, 1984; Randell, 1995).

Koshioka et al., (1978) reported variations within a species pertaining to the amount of anthraquinones (Crawford et al.,1990). According to Dave and Ledwani (2012) and

Upadhyaya and Singh (1986), anthraquinones obtusin and obtusifolin are specific to *S. obtusifolia* only. Both the legumes do not show any nodulation (Parson and Cuthbertson, 1992). According to Irwin and Barneby (1982), there exists two forms of *S. obtusifolia* in America. One form having extra-floral nectary of uniglandular nature between the lowest leaflet pair on the upper surface of the rachis with chromosome number 2n = 28, originating from the Caribbean, and the other having biglandular extrafloral nectaries between the lower leaflet pairs, originating from the British Guiana, Surinam and Venezuela showing 2n = 26, with relatively narrower pods. Both the species are karyologically distinct with some forms of *S. obtusifolia* having haploid chromosome number 14, 13, 12, while *S. tora* of Indian origin having chromosome count 14, 13 suggesting the origin of the later through aneuploid loss from the former (Tandon and Bhatt, 1971; Upadhayaya and Singh, 1986).

2.2 Phenological Analyses

2.2.1 Germination and Growth

The species is weed and are quite prevalent on roadsides, wastelands, open woody areas, cultivated areas and pastures like some other congeneric herbs (Parsons and Cuthbertson, 1992). *S. obtusifolia* grows in an optimum pH of 5.5 to 6.0 but can tolerate pH ranging from 4.7 to 6.3 (Creel et al., 1968); however, stunted growth at pH 4.7 to 5.2 has also been reported (Buchanan et al., 1975). Sicklepod showed high optimum temperature for growth with warm night temperature (Patterson et al., 1993). Seed germination occurs at a wide range of temperature between 18℃ to 36℃ with the maximum rate of germination between 24℃ to 36℃.

The plant species is an erect shrub with a height of upto 2.5 meters (James and Fossett, 1983), but usually ranges around 1.5-1.8 meters (Patterson 1993) and depends also on

the ecotypes. Germination and growth of sicklepod occur at high optimum temperature. The maximum plant height occurs at a higher summer rainfall and temperature (Patterson, 1993). The roots of *S. obtusifolia* showed deep seated tap root but showed stunted root growth in prevailing drought conditions.

2.3 Morphological Analyses

2.3.1 Floral Biology

Appearance of first flower in *S. obtusifolia* occurs, generally, after 43 to 84 days of vegetative phase depending upon the ecotype and climate (Retzinger, 1984). The flowers in *S. obtusifolia* are self-pollinated and the gynoecium is recurved over the anther pore (Gottsberger and Gottsberger-Silberbauer, 1988) which is quite unusual in Cassiinae. This suggests the self-fertile nature of the species (Retzinger, 1984). Indeed, *S. obtusifolia* probably showed self-fertilization as the flower fertilizes in the late-bud even before its opening. This sicklepod plant has been identified as a short-day species, in which the reproductive phase is enhanced by 12 hours day light or short photoperiods, though, it depends on the ecotypes of the plant, too (Patterson, 1992).

2.3.2 Fruit and Seed

Earlier report on the average number of seeds of the species has shown to vary greatly from 5280 to 8520 and the number of pods in the plant varies from 63 to 342, while the average number of seeds vary from 24 to 28 per pod (Retzinger, 1984). Fruits of this species dehisce and disperse seeds having hard seed coat giving 5-10 years (Anning et al., 1989). The seeds are covered by a wax like covering of about 0.1 millimeters in thickness (Creel et al., 1968). Seeds require scarification for germination either scrubbing manually or with acid treatment for maximizing as well as to get uniform germination (Cock and Evans, 1984). Fire scarification of the seeds has also shown in

Australia a fast mass germination after a good rain (Anning et al., 1989). Sicklepod seeds can withstand drought conditions and can germinate in low moisture soils which supports its colonization in bare barren lands and sandy soils (Hoveland and Buchanan, 1973). Germination of *S. obtusifolia* can occur uo to 12.7 cm depth underneath soil surface (Teem et al., 1980). Viability of the sicklepod seeds decreases to 50% irrespective of other germinating conditions. Seed exudates released from sicklepod seeds inhibits the germination of other seeds (Creel et al., 1968). *S. obtusifolia* having dehiscent pod disperse seeds up to a distance of five meters from the source. Long distance seed dispersal occurs by water movement over soil surface, stream flow, seed with the mud attached on the machinery and vehicles. Cattle, horses, goats etc., after nibbling the seeds, the undigested part passes through gut and dispersed in their dung.

2.4 Cytological Studies

Senna exhibits mainly X = 14 as the the basic number while X = 13 was also observed. The occurrence of autopolyploidization is also observed which is quite important in respect of the evolutionary status (Resende et al., 2013). Diversification of the various species including *S. obtusifolia* of the genus *Senna* regarding intraspecific and, or interspecific karyotypic variations making it an important taxon throughout various regions of the world (Cordeiro and Felix, 2018). According to Cordeiro and Felix (2018), around twenty percent of *Senna sp.* exhibits a chromosome count of 2n = 28. Variation of chromosome count recorded in some species including *S. obtusifolia* is 2n = 22, 24, and 26 (Rice et al., 2015). Intraspecific diversity in the *Senna rugosa* (n = 14, and n = 28) was stuidied by Resende et al. (2013). Polyploidy is also quite prevalent in these genera with chromosome counts of 112, 56, and 42 in *S. rugosa*, and 52 in *S. gardneri* (Resende et al., 2014). *S. obtusifolia* showed intraspecific karyotypic variations showing 2n = 24, 26 or 28 (Rice et al., 2015; Cordeiro and Felix, 2018).

These types of intraspecific variations occurred in *S. obtusifolia* as a result of diverse karyological phenomenons such as disploidy, neopolyploidy (Guerra, 2008), aneusomy and polysomy (Rodrigues et al., 2009) causing chromosome number variability. Study of karyotype using fluorochromes for studying heterochromatin pattern can be a means for understanding the taxonomic relationships and evolutionary trends. For analyzing heterochromatin patterns, Souza and Benko-Iseppon, (2004) observed two terminal CMA+/DAPI- bands in *S. obtusifolia,* while Cordeiro and Felix (2018) found six terminal CMA+/DAPI- bands. Within the same species in *S. obtusifolia*, differences may arise in heterochromatin patterns (Souza and Benko-Iseeppon, 2004).

2.5 Genetical Studies

Various molecular tools are employed to determine the polymorphism at intraspecific levels. They include RFLP, RAPD, AFLP, microsatellites or SSR, ISSR, SCoT, SNP, ITS and SCAR (Williams et al., 1993; Zabeau and Vos, 1993; Meudt and Clarke, 2007). AFLP method is the most efficient one producing larger percentage of independent polymorphic loci (Sathyanarayana et al., 2011; Kim et al., 2013). These different DNA fingerprinting techniques used by Mao et al., (2017) employed 100 ISSR and 85 SCoT in *S. obtusifolia*. RAPD, ISSR and SSR techniques were used in 28 species of *Cassia* to reveal the phylogenetic relationship and genetic diversity (Mohanty et al., 2010). Dendrograms prepared from these analyses helped unveiling the nature and extent of relationships among the individuals.

2.6 Biochemical Studies

Numerous secondary metabolites or natural bioactive compounds produced by *S. obtusifolia*. This makes the weed species a subject of continuing and subsequent interest aiming medicinal fields, dye and cosmetic industries, wastewater treatment plants and

papermill, and effluent industries with revenue turnover in billion dollars. Various types of anthraquinones are present in this species in along with sennosides, glycosides, aglycones, naphthopyrones, flavonoids, triterpenoids, anthrones, xanthones, polyketides (Yang et al., 2015; Zhang et al., 2009). Different analytical tools like HPLC (Xu et al., 2012), Capillary electrophoresis (Koyama et al., 2003), UHPLC coupled with mass spectrometry (Zhang et al., 2012), and Ultraviolet spectrophotometry (Zhang et al., 2007) were used to identify and quantify various bioactive compounds. A detailed description of the bioactive compounds isolated from *S. obtusifolia* has been mentioned earlier. The presence of various anthraquinones with uses have also been elaborated later in this chapter. Anthraquinones produced by this species are mainly produced by various biosynthetic pathways among which the polyketide pathway and shikimic, isochorismic and o-succinylbenzoic acid pathway are mostly confirmed (Han et al., 2001; Shukla et al., 2017). Recently, the medicinal components separated from seeds of the species by a combined process of HPLC and HPLC-MS-CID gave the components recoverable from the plant (Harry-O-kuru et al., 2012).

2.7 Medicinal Properties

2.7.1 Traditional, Herbal, Ethnomedicinal and Folklore Properties

Many species of *Senna* are known to have great potential for medicinal uses (Burkill, 2002; Tona et al., 2004). Plant derived substances as therapeutic agents or phytopharmaceuticals are gaining interest in recent years for its biosafety nature and is used frequently for preventing various diseases (Newman et al., 2003). WHO reported that about 80% of total world populations rely and depend on herbal medicines. According to Essiett and Bassey (2013), *S. obtusifolia* is nutritionally rich and contains all the essential elements for good human and animal health. Various medicinal plants produce numerous plants derived therapeutic agents. *S. obtusifolia* is the most valuable

and traditional, herbal, ethnomedicinal plant and treated in several cases of ethnomedicinal and folklore practices in many countries of East Asia with extensive medical uses (Seo et al., 2017). *S. obtusifolia* root and bark decoctions are quite safe which can be used for children and pregnant women as a mild laxative in and consumed as healthy drinks for alleviating constipation, contributing in ethnopharmacological and folklore practices (Ajayi et al., 2014; Yang et al., 2015). Zhu (1998) reported that seeds of *S. obtusifolia* have been traditionally used in Korea, Japan, and China for treating inflammation in eyes, photophobia, and lacrimation. The dried seeds are usefull for brewing tea (Jung et al., 2016), sometimes drunk as roasted tea (Yang et al., 2015). They are also used to treat headaches, dizziness, hypertension, and liver problems (Cong et al., 2014; Hao et al., 2001). *S. obtusifolia* is also reported to exhibit antidiarrheal, diuretic, and antiseptic properties (Tang et al., 2008). The species helps to enhance liver function and helps in proper bowel movement (Hao et al., 2001), and used as an antioxidant, antimutagenic, and tonic (Jain and Patil, 2010; Tang et al., 2008). Leaf decoctions are applied in gastrointestinal treatments. Leaves of the plant are chewed for the remedy of cough pneumonia, fever, ulcer and skin-infections (Ajayi et al., 2014). Roots of the plant is used as purgative and anthelminthic (Toruan-Purba, 1999). Both the fresh leaves and its fermented products “Kawal” is consumed by tribals of Sudan. According to Dirar et al., (1985), “Kawal” is considered as the meat substitute due to its high protein content. “Kawal” is used for the treatment of jaundice. The herbal preparation of the plant containing chrysophanol and aloe-emodin have antimicrobacterial activity (Smolarz et al., 2013). The leaves, roots, seeds, and flowers of *S. obtusifolia* are used as folk medicine in Laos and Thailand (Doughari et al., 2008). In snakebite treatment, *S. obtusifolia* is used as an ethnomedicinal practice (Upasani et al., 2017).

2.7.2 Modern Medicinal Properties

The ethanol extract from the seeds of *S. obtusifolia* exhibits hepatoprotective effect in HepG2 cells. From active ethyl acetate fraction, ten phenolic glycosides have been isolated. Out of which, toralactone 9-O-gentiobioside showed maximum efficacy (Seo et al., 2017).

The active compounds- emodin, questin, toralactone, gentiobioside, etc. obtained from *S. obtusifolia* seed extract was used in the treatment of Alzheimer's disease (Kim et al., 2007; Jung et al., 2016). *S. obtusifolia* seed extracts ameliorates amyloid β-induced synaptic dysfunction (Yi et al., 2016). *S. obtusifolia* is found to be effective against Parkinson disease (Kim et al., 2007; Ittiyavirah and Hameed, 2014). *S. obtusifolia* leaf ethanolic and seed methanolic extracts have larvicidal effects against *Anopheles stephensi* mosquito (Jang et al., 2002; Kamraj et el., 2011). Larvicidal activity of this species is observed by applying the extracts against three mosquito vectors (Bagavan and Rahuman, 2011; Diarra et al., 2015).

The anticancer potential of *S. obtusifolia* is quite impressive, which shows that the preparations containing rhein, aloe-emodin, and emodin possess antitumor, antiviral, and anti-inflammatory activities (Weber, 2013). Controlling the breast cancer cell proliferation is triggered by emodin and aloe-emodin (Huang et al., 2013). Emodin and obtusifolin exhibits inhibitory activity on end product formation on glycation (Jang et al., 2007), while aurantio-obtusin along with emodin showed inhibitory effects on rat aldolase reductase. Physcion shows anti-cervical cancer activity in human (Wijesekara et al., 2014).

The literature reported antioxidant property of *S. obtusifolia* (Doughari et al., 2008; Zhang et al., 2011; 2012; Lin et al., 2015). The Semen Cassiae is useful in acute inflammatory disease attributable to its anti-inflammatory actions (Jung et al., 2016).

Antimicrobial activity of *S. obtusifolia* was studied by Kitanaka and Takido (1986). Though the actions of 1,2-, 1,4- and 1,8-dihydroxyanthraquinones inhibits the growth against *C. perfringens* but strongly promotes the growth and activity of *Bifidobacterium bifidum* (Sung et al., 2004).

2.7.3 Other Important Uses of *S. obtusifolia*

Anthraquinones from *S. obtusifolia* imparts shades of different colours and is used in dye industries (Dave and Ledwani, 2012). Organic compounds used as insecticides and pesticides are extracted from different parts of *S. obtusifolia* (Dave and Ledwani, 2012). The natural seed gum of *S. obtusifolia* acts as a coagulant aid in caogulation-flocculation treatments, high strength agro-industrial waste, and waste water with natural pH (Shak and Wu, 2014; 2015; Oladoja, 2015). For treating paper mill effluent and raw pulp in the paper mill industry, *S. obtusifolia* seed gum is used (Subramonian et al., 2014).

2.8 Review of Literature on Active Compounds

Several active compounds were isolated from *S. obtusifolia* which are medicinally important such as flavonoids, triterpenoides, anthrones, sennosides and anthraquinones. The aglycones that has been identified and characterised from *S. obtusifolia* are obtusifolin, obtusin, physcion, chrysophanic acid, aurantioobtusin, chrysoobtusin,and emodin (Takido, 1960). Numerous other anthraquinones include torachrysone (Shibata et al., 1969) and toralactone (Takahashi, 1973). Wu et al., (2011) reported6-di-O-acetylglucopyranoside, 6-di-O-acetylglucopyranoside, obtusifoline-2-O-b-D-

glucopyranoside,6-di-O-acetylglucopyranoside, obtusifoline-2-O-b-D-3, obtusifoline-2-O-b-D-2, and obtusifoline-2-O-b-D-4 from the seeds of *S. obtusifolia*. Air dried twigs and leaves of *S. obtusifolia* when extracted gave friedelin (Klass and Tinto, 1992), lupeol (Kuiate et al., 2007); stigmasterol (Forgo and Kover, 2004). Air dried leaves of *S. obtusifolia* yielded euxanthone (Locksley et al., 1969);1,8-dihydroxy-3-methoxy-6-methylxanthone (Sob et al., 2008); chrysophanol, physcion, 1,2,8-trihydroxy-6,7-dimethoxyanthraquinone (Guo et al.,1998), obtusifolin (Takido, 1958), 1,5-dihydroxy-3-methoxy-7-methylanthraquinone (Kazmi et al., 1994), 1,7-dihydroxy-3-methoxyxanthone (Kijjoa et al., 2000), 1-hydroxy-7-methoxy-3-methyl-anthraquinone (Guo et al., 1998), 1-O-methyl chrysophanol (Guo et al., 1998), 8-O-methyl chrysophanol (Guo et al., 1998), 1,3,6-trihydroxy- 8-methylxanthone (Tanahashi et al., 1999), stigmasterol (Alam et al., 1996), friedelin (Candy et al., 1968), lupeol (Solichin et al., 1980), (4R*,5S*,6E,8Z)8Z)-ethyl-4-((E)-but-1-enyl)-5-hydroxypentdeca-6,8-dienoate (Sob et al., 2008), (24S)-24-ethylcholesta-5,22(E),25-trien-3b-ol (Gaspar et al., 1996), (-)-acetoxy-9,10-dimethyl-1,5-octacosanolide (Rizvi et al., 1972), and (E)-eicos-14-enoic acid (Rod'kina, 2005).

S. obtusifolia seeds contain various other compounds. An exhaustive list of such compounds is reported by Kitanaka et al., (1981); Guo et al., (1998), Zhou et al., (2006); Sob et al., (2008); Zhang et al., (2009), and Wu et al., (2011).

2.8.1 Anthraquinones

S. obtusifolia extracts has some compounds contained therein have shown that the plant has been used traditionally. These compounds are used to produce a variety of modern drugs. The literature reported isolation of different constituents from this plant which include anthrones, anthraquinones, naphthopyrones, sennosides, lactones, and their

glycosides, triterpenoids and flavonoids (Kitanaka et al.,1985; Yun-Choi et al., 1990, Hatano et al., 1999; Zhang et al., 2009; Yang et al., 2015).

Natural products of remarkable varieties were isolated from the number of plant species. They are also used as repellents of herbivores, insecticides, fungicides, etc. As these resources are renewable and biodegradable in nature, they are environment friendly. Sustainable development of these products opens a new area in research (Dave and Ledwani, 2012).

Anthraquinones are such compounds that occurs naturally in some plant species, fungi, lichens and insects. There are wide range of applications of natural anthraquinones. Anthraquinones are anthracene group of chemicals with parent structure 9,10-dioxoanthracene. Anthraquinones present in *Senna* are majorly found as glycosides (anthrones dianthrones or oxanthrones) (Crawford et al., 1990).

2.8.2 Rhein

Rhein is a monomer of anthraquinone derivatives useful in Chinese traditional medicine to treat various clinical ailments for over 1000 years (Zhou et al., 2015). The chemical composition of rhein is 4,5-dihydroxyanthraquinone-2-carboxylic acid. It is the main source and initial raw material or precursor of Diacerin, a drug used for hypertension. Rhein is found in medicinal herbs like *Aloe barbadensis Miler, Polygonum multiflorum* Thunb*., Rheum palmatum* L., and *S. tora* L. etc. Rhein shows linear pharmacokinetics in the range of 50 and 200 mg (Layek et al., 2008) and its pharmacological benefits regarding human health have been explored actively and intensively. The use of rhein as a medicinal agent are briefly mentioned next.

2.8.2.1 Hepatoprotective Activity of Rhein

Rhein is effective in modulating cytochrome P450 (CYP) enzymes in rat liver microsomes where it significantly inhibited CYP2E1, CYP2C9, and CYP3A and mild inhibitory effects on CYP1A2 and CYP2D6 (Tang et al., 2009). Rhein when administered with rich fat meal in hepatitis B virus-transgenic mice decreases fasting plasma glucose, total cholesterol and triglyceride and increases glucose and lipid metabolism in serum levels (Bian et al., 2013). It targets the regulation of lipogenic enzyme sterol and several other genes in liver which accelerate energy expenditure. On the other hand, decreases liver triglyceride and cholesterol. Rhein was also found to be effective in improving hepatic steatosis and insulin resistance and regulating T helpers in T-cells with the increase in the expression of GATA binding protein-3 (Sheng et al., 2011). Rhein acts as an antifibrotic agent in hepatic disorders to check liver fibrosis (Guo. et al., 2002; Wynn, 2007; Zhou et al., 2015).

2.8.2.2 Antioxidant Activity of Rhein

In many pathophysiological disorders like degenerative rheumatic and neurodegenerative diseases, cancer etc., rhein inhibits reactive oxygen species (ROS) production by activating N-formyl-methionyl-leucyl-phenylalanine in human. According to Zhao et al., (2011), rhein decreases urea nitrogen content, creatinine, glutamate-oxaloacetic transaminase and GSH concentration levels in kidney and liver.

2.8.2.3 Anti-Inflammatory Activity of Rhein

Inflammation induced vascular complications can be reduced by the decrease of the expression of vascular cell adhesion molecule-1 and endothelial cell adhesion molecules. The transcription of these molecules is reduced by rhein treatment in the endothelial cells of umbilical vein (Hu et al., 2013).

2.8.2.4 Antidiabetic Activity of Rhein

In vivo study, administration of rhein for 8 weeks increased secretion of insulin level. Additionally, protection of beta cell mass and inhibition of beta cell apoptosis is observed under rhein treatment (Du et al., 2010; 2012). Oral administration of Rhein improves glucose tolerance and reduction of fasting βblood glucose level in 8-12 weeks. Rhein can protect ultrastructure of mitochondria by inhibiting beta cell apoptosis on localization on the same (Liu et al., 2013). Rhein can also be used for treating Type II diabetes where intragastric treatment of rhein significantly lowered blood glucose concentrations after a specific time span of glucose load, suppressing beta- cell apoptosis in pancreas and elevation of insulin secretion in early phase in mice (Du H. et al., 2011).

2.8.2.5 Nephroprotective Activity of Rhein

Nephroprotective property of Rhein has been studied by several researchers both in vitro and in vivo. Rhein helps in protein expression of intestinal epithelial tissues and repairs damaged tight junctions, and protect intestinal barrier (He et al., 2012). Rhein also helps to reduce interstitial fibrosis of renal tissues, in vitro when an oral dose of 150mg/K/d is applied to mice with unilateral uretal obstruction (Peng et al., 2013).

Rhein considerably increases mesangial expansion and glomerular hypertrophy with renal capsule dilation in rats and decreases the urinary red blood cells volume, and IgA deposition in glomerulus (Peng et al., 2013).In chronic nephropathy Rhein reduces interstitial inflammation and renal fibrosis along with fibronectin and collagen IV reduction, whereas increases hepatic growth factor levels (Su et al., 2013).Rhein helps in the protection against renal injury progression (Ji et al., 2005) and inhibition of hypertrophy of renal proximal tubular epithelial cells (Yu et al., 2010).

2.8.2.6 Chondroprotective Activity of Rhein

Rhein's effect on Osteoarthritis chondrocytes by reducing the secretion of Interleukin-1beta (chondrocytes, synovial cells and macrophages) having important role in cartilage destruction (Moldovan et al., 2000). Rhein enhanced 46.5% aggrecan, 50% prostaglandin E2, while reduced interleukin-6, matrix metalloproteinase-3, and Interleukin 1beta (Sanchez et al., 2003).

2.8.2.7 Anticancer Activity of Rhein

Various anticarcinogenic activities of Rhein include metastasis and proliferation of the cells in vitro, such as inhibition of the proliferation of hypertrophic scar fibroblasts (Wang et al., 2012). Rhein inhibits nasopharyngeal carcinoma of humans by suppressing the expression of growth factor of vascular endothelium, recetor bound protein 2 and Ras, inhibition of p38 MAPK and phoshorylation of ERK, while activating NF-kappa B (Lin et al., 2009). Rhein restricts the viability of hormone dependence and cell cycle in independent breast cancer cells (MCF-7), (MDA- MB-435s) (Fernand et al., 2011). Rhein suppresses the inclusion and migration of tongue cancer cells in humans - SCC-4 cells and prevents the mRNA expression of MMP-9 causing metastasis and tumor invasion of human cancer of various types (Chen et al., 2010). Rhein induces apoptosis in cancer cells of humans such as BEL-7402 cells which induces hepatocellular carcinoma in humans. Rhein also induces the apoptosis of promyelocytic leukemia cells in humans (Lin et al., 2003a) and increases the generation of p38 kinase (Lin et al., 2003b).

Anticarcinogenic effects of Rhein includes in the inhibition of colon cells of human adenocarcinoma(Caco-2), and cell proliferation. Rhein reduces H_2O_2 responsible for DNA damage and restricting cancer invasion. Rhein is reported to cause apoptosis-a

physiological process of removing malignant cancer cells, in human hepatocellular carcinoma cells, increases caspase-3 gene expression and decreases c-Myc gene expression (Shi et al., 2008). Human cervical cancer can be abrogated by cleavage of bid protein while reducing the Bcl-2 level while increasing p21, p53, Fas, and cytoplasmic Ca2+, caspase-8,-9, caspase - 3 activation which ultimately helps in DNA fragmentation (Ip et al., 2007).

2.8.2.8 Antimicrobial Activity of Rhein

Rhein as a potential antibacterial agent can suppress the growth of *Helicobacter pylori* in peptic ulcer patients by inhibiting arylamine N-acetyltransferase (Chung et al., 1998). Rhein also inhibited the *Staphylococcus aureus* growth by preventing the transcription of the genes responsible for fermentation and anaerobic respiration (Yu et al., 2008). Rhein having a high affinity for CpG DNA - a bacterial DNA responsible for the pathogenesis of sepsis, prevents it from binding to its receptor thus reducing pathogenicity (Liu et al., 2009).

2.8.2.9 Purgative Activity of Rhein

Rhein shows purgative activity by stimulating submucosal neurons which activates chloride secretion on mucosal or serosal applications by releasing endogenous prostaglandins and acetylcholine (Frieling et al., 1993). Rhein also helps to induce secretion of ion in human CaCo-2 monolayer cells (Raimondi et al., 2002). Rhein reduces water and sodium ion absorption in rat colon in-situ thus increases purgative activity (Leng-Peschlow, 1993).

2.8.2.10 Lipid-Lowering Activity of Rhein

Rhein helps in the suppression of LXR target genes responsible for cholesterol and adipogenesis metabolism, the expression of which regulates homeostasis of cholesterol and lipid and plays a major role in energy metabolism. In brown adipose tissue, rhein activates uncoupling protein 1 expression suggesting that obesity and other related metabolic disorders can be protected by rhein (Sheng et al., 2012). Rhein reverses adiposity and downregulates the mRNA levels of adipogenesis specific transcription factors in mice (Liu et al., 2011).

2.8.2.11 Other Activities of Rhein

Rhein is found to be effective in having estrogenic activity (Kang et al., 2008). Rhein also induces apoptosis, helps in releasing cytochrome c in cytosol, reduction in Bcl-xL, Bcl-2 expression of TNF-alpha in the smooth muscle cells of human aorta thus inhibiting cell proliferation (Heo et al., 2009). Rhein is found to have inhibition in degranulation of mast cells and suress lipoxygenase enzyme activity proving its anti-allergic activity (Singh et al., 2012). Rhein also have protective effect on endothelial dysfunction, treatment of vascular diseases and prevention of intervertebral disc degeneration (Zhu et al., 2003; Li et al., 2011).