



Micropropagation, Phytochemistry and Pharmacology studies on *Pterocarpus marsupium* Roxb(Bijasal). An important medicinal forest tree. A Review

Naseer Ahmad¹, Aparna Alia²

1: Department of Botany , Baba Gulam Shah Badshah University, Rajouri J&K India.

2: Department of Botany Rajeev Gandhi PG College Bhopal, Madhya Pradesh. India.

Abstract: *Pterocarpus marsupium* Roxb belongs to the family fabaceaeis commly known as red Indian kino tree. This forest endangered medicinal tree has restricted distribution to southern parts of india. *Pterocarpus marsupium* is medium to large sized deciduous tree and can reach upto the height of 30m In nature *Pterocarpus marsupium* has a very restricted distribution and does not propagate well in nature due to which this tree species is declining in the wild and therefore it has been placed in the red data book. In order to conserve and sustainable use of this endangered species a lot of research efforts are been made on *Pterocarpus marsupium* in the field of micropropagation, phytochemistry and pharmacology.

Key words: *Pterocarpus marsupium*, Micro propagation, Phytochemistry and Pharmacology

Introduction:

Micropropagation: In a view of scientific interest it becomes necessary to examine what have already done and achieved in the said field. Since many efforts have been made on the medicinal plants for their conservation. *Ex-situ* and *In-situ* programmes are being implimented on various levels to protect plants from depletion. Plant tissue culture one such an important work in the field of plant conservation. . The science of plant tissue culture takes its roots from path breaking research in botany like discovery of cell followed by propounding of cell theory and other important contributions in this field which are mentioned below. Biotechnology is name given to the methods and techniques that involve the use of living organisms like bacteria, yeast, plant cells etc or their parts or products as tools (for example, genes and enzymes). They are used in a number of fields: food processing, agriculture, pharmaceuticals, and medicine, among others. Plant tissue culture can be defined as culture of plant seeds, organs, explants, tissues, cells, or protoplasts on nutrient media under sterile conditions.

The science of plant tissue culture takes its roots from path breaking research in botany like discovery of cell followed by propounding of cell theory. **Schleiden and Schwann (1839)** proposed that cell is the basic unit of organisms. They visualized that cell is capable of autonomy and therefore it should be possible for each cell if given an environment to regenerate into whole plant. Based on this premise, German physiologist, Gottlieb Haberlandt (1902) developed the concept of *in vitro* cell culture. He isolated single fully differentiated individual plant cells from different plant species like palisade cells from leaves of *Laminum purpureum*, glandular hair of *Pulmonaria* and pith cells from petioles of *Eicchornia crassiples* etc and was first to culture them in Knop's salt solution enriched with glucose. In his cultures, cells increased in size, accumulated starch but failed to divide. Therefore, Haberlandt's prediction failed that the

cultured plant cells could grow, divide and develop into embryo and then to whole plant. This potential of a cell is known as totipotency, a term coined by Steward in 1968. Despite lack of success, Haberlandt made several predictions about the requirements in media in experimental conditions which could possibly induce cell division, proliferation and embryo induction. G Haberlandt is thus regarded as father of tissue culture. Taking cue from Haberlandt's failure, **Hannig (1904)** chose embryogenic tissue to culture. He excised nearly mature embryos from seeds of several species of crucifers and successfully grew them to maturity on mineral salts and sugar solution. In 1908, Simon regenerated callus, buds and roots from *Poplar* stem segments and established the basis for callus culture. For about next 30 years (upto 1934), there was very little further progress in cell culture research. Within this period, an innovative approach to tissue culture using meristematic cells like root and stem tips was reported by **Kolte (1922)** and **Robbins (1922)** working independently. In 1926, Fritz Went discovered first plant growth regulator (PGR), indoleacetic acid (IAA). IAA is a naturally occurring member of a class of PGRs termed 'auxins'. All these research attempts involving culture of isolated cells, root tips or stem tips ended in development of calluses. There were two objectives to be achieved before putting Haberlandt's prediction to fruition. First, to make the callus obtained from the explants to proliferate endlessly and second to induce these regenerated calluses to undergo organogenesis and form whole plants. It was in 1930s, when progress in plant tissue culture accelerated rapidly owing to an important discovery that vitamin B and natural auxins were necessary for the growth of isolated tissues containing meristems. This breakthrough came from **White (1934)** who reported that not only could cultured tomato root tips but could be repeatedly sub cultured to fresh medium of inorganic salts supplemented with yeast extract and later (1937) replaced YE by vitamin B namely pyridoxine, thiamine and proved their growth promoting effect. **Roger J Gautheret (1934)** has reported the successful culture of cambium cells of several tree species to produce callus and addition of auxin enhanced the proliferation of his cambial cultures. Further research by **Nobecourt (1937)**, who could successful grow continuous callus cultures of carrot slices and White (1939) who obtained similar results from tumour tissues of hybrid *Nicotiana glauca x N langsdorffii*. Thus, the possibility of cultivating plant tissues for an unlimited period was independently endorsed by Gautheret, White and Nobecourt in 1939. Adding to the ongoing improvements in the culture media, **Johannes Van Overbeek (1941)** reported growth of seedlings from heart shaped embryos by enriching culture media with coconut milk besides the usual salts, vitamins and other nutrients. This provided tremendous impetus for further work in embryo culture. Stem tip cultures yielded success when Ernest Ball (1946) devised a method to identify the exact part of shoot meristem that gives rise to whole plant. After 1950, there was an immense advancement in knowledge of effect of PGRs on plant development. The fact that coconut milk (embryo sac fluid) is nutritional requirement for tobacco callus besides auxin, indicated the non auxinic nature of milk. This prompted further research and so other classes of PGRs were recognized. **(Skoog and Tsui 1957)** demonstrated induction of cell division and bud formation in tobacco by adenine. This led to further investigations by **(Skoog and Miller 1955)** who isolated 'kinetin'- a derivative of adenine (6-furyl aminopurine). Kinetin and many such other compounds which show bud promoting activities are collectively called cytokinins, a cell division promoter in cells of highly mature and differentiated tissues. **(Skoog and Miller 1957)** worked further to propose the concept of hormonal control of organ formation. Their experiment on tobacco pith cultures showed that high concentration of auxin promoted rooting and high kinetin induces bud formation or shooting. However, now the concept is altered to multiple factors like source of plant tissue, environmental factors, composition of media, polarity, growth substances being responsible for determination of organogenesis. Besides PGRs, scientists tried to improve culture media by differing essentially in mineral content. In this direction, **(Murashige and Skoog 1962)** prepared a medium by increasing the concentration of salts twenty-five times higher than Knops. This media enhanced the growth of tobacco tissues by five times. Even today MS medium has immense commercial application in tissue culture.

Having achieved success and expertise in growth of callus cultures from explants under *in vitro* conditions, focus now shifted to preparation of single cell cultures. (Muir 1953-54) demonstrated that when callus tissues were transferred to liquid medium and subjected to shaking, callus tissues broke into single cells. (Bergmann 1960) developed a technique for cloning of these single cells by filtering suspension cultures. This technique called Plating technique is widely used for cloning isolated single protoplasts.

Next step for realization of Haberlandt's objectives was development of whole plant from the proliferated tissue of these cells. Vasil and Hilderbrandt were first to regenerate plantlets from colonies of isolated cells of hybrid *Nicotiana glutinosa x N. tabacum*. (Steward 1966) the classical work on induction of somatic embryos from free cells in carrot suspension cultures brought an important breakthrough by finally demonstrating totipotency of somatic cells, thereby validating the ideas of Haberlandt. This ability of regenerating plants from single somatic cells through normal developmental process had great applications in both plant propagation and also genetic engineering. For e.g. micropropagation where small amounts of tissue can be used to continuously raise thousand more plants. Morel utilized this application for rapid propagation of orchids and Dahlias. He was also the first scientist to free the orchid and Dahlia plants from virus by cultivating shoot meristem of infected plants.

The role of tissue culture in plant genetic engineering was first exemplified by (Kanta and Maheshwari 1962). They developed a technique of test tube fertilization which involved growing of excised ovules and pollen grains in the same medium thus overcoming the incompatibility barriers at sexual level. (Guha and Maheshwari 1966) cultured anthers of *Datura* and raised embryos which developed into haploid plants initiating androgenesis. This discovery received significant attention since plants recovered from doubled haploid cells are homozygous and express all recessive genes thus making them ideal for pure breeding lines.

Next breakthrough in application of tissue culture came with isolation and regeneration of protoplasts first demonstrated by (C Cocking 1960). Plant protoplasts are naked cells from which cell wall has been removed. Cocking produced large quantities of protoplasts by using cell wall degrading enzymes. After success in regeneration of protoplasts, (Carlson 1972) isolated protoplasts from *Nicotiana glauca x N. langsdorfii* and fused them to produce first somatic hybrid. Since then many divergent somatic hybrids have been produced. With the advent of restriction enzymes tissue culture headed towards a new research area. The totipotent plant cells could now be altered by insertion of specific foreign genes giving rise to genetically modified crops.

Phytochemistry: Phytochemistry is a distinct discipline somewhere in between organic chemistry, plant biochemistry and closely related to natural products. It deals with a variety of organic substances accumulated in plants. The plant may be considered as a biosynthetic laboratory. Not only their chemical compounds such as carbohydrates, protein, and lipids that are used as food by man, but also a multitude of compounds like glycosides, alkaloids, flavonoids, etc. are used as medicines by him in various ways and means. The qualitative and quantitative estimation of the phytochemical constituents of a medicinal plant is considered to be an important step in medicinal plant research (Kokate, 1994). Phytochemical progress has been aided enormously by the development of rapid and accurate methods of screening plants for particular chemicals (Banso and Adeyemo, 2007). Medicinal plants contain physiologically active principles that over the years have been exploited in traditional medicine for the treatment of various ailments (Adebajo *et al.*, 1983). The drugs contained in medicinal plants are known as active principles. (Cowmann,1999, Banso and Olutimayin 2001) reported that plants contain a wide variety of active principles. There is a reasonable likelihood that medicinal plants with a long history of human use will ultimately yield novel

drug prototypes (Eshrat and Hussain, 2002). The most commonly encountered secondary metabolites of plants are saponins, tannins, flavonoids, alkaloids, anthraquinones, cardiac glycosides and cyanogenic glycosides. The pharmacological and other beneficial effects of antinutritional factors in plants have been reviewed by (Soetan 2008). The presence of these secondary metabolites in plants probably explains the various uses of plants for traditional medicine.

Pterocarpus marsupium

Morphological characteristics

Pterocarpus species can be recognized in field by its straight bole, longitudinally fissured bark, imparipinnate and elliptic leaves, fragrant flowers in large panicles, and winged, flat pods. The tree reaches up to 30 m in height and up to 2.5 m in girth with straight and clear bole. Bark is scaly, rough, and longitudinally fissured. Leaflets are generally five to seven in number, 8–13 cm long, oblong or elliptic, or rotund, with 15–20 pairs of lateral veins. Oleo-resin obtained from tree trunk is called kino-gum, which is fragrant, brittle, almost black in colour, angular and glistening, and occurs in small flakes.

Floral characteristics

Fragrant, yellow flowers occur in about 1–5 cm long large panicles. Pods are flat, orbicular, winged, and up to 5 cm in diameter. Seeds are one to three in number, bony and convex in shape. Flowering begins in November, while fruiting continues up to March.

Distribution

The tree is found in central and peninsular India, chiefly in dry mixed deciduous tropical forests of Gujarat, Madhya Pradesh, and sub-Himalayan tracts, at up to 1000 m altitude. Natural populations have greatly reduced and often no tender young saplings can be found in the forest. This is a threatened species on account of autogenic reproductive deficiency

Climate and soil

The tree occurs in tropical region and thrives well in open sun under moderate rainfall of 80–200 cm. It prefers fertile, deep clayey loam soil with good drainage. It can tolerate excessive temperatures in summer.

Taxonomical Classification:

Domain : Eukaryota
Kingdom : Plantae
Division : Magnoliophyta
Class : Magnoliopsida
Order : Fabales
Family : Fabaceae
Genus : *Pterocarpus*
Species : *marsupium*

Micropropagation studies of *Pterocarpus marsupium*:

Venkataramaiah et al., (1980). The native natural stands of *Pterocarpus marsupium* are fast disappearing. The conventional seed and vegetative propagation of the tree has not been very successful due to hard fruit coat, less germination ability together with poor viability of seeds. **Kalimuthu and Lakshaman** (1995) has reported that natural regeneration of *Pterocarpus marsupium* takes place by means of the seed but the germination percentage is only 30% and is very low.

Distabanjong and Geneve (1997) has inoculated the nodal explant on the MS medium showing differential response to all the three cytokinins (BA, Kin and 2iP) and noticed that nodal explant taken from the 18 day old axenic seedling showed better response for shoot induction as compared to 6, 12 and 24 days old seedlings. They also noticed that the age of the seedlings plays an important role in morphogenesis.

Rath et al (1998) has tested growth of different seedling explants on culture media containing various growth regulators and rhizogenesis occurred from callus but root development was not encouraging.

Anuradha and Pullaiah (1999) has reported *in vitro* seed culture and enhanced axillary branching in *P. santalinus* and *P. marsupium* but the number of shoots was less.

Choudhuri and Sarkar (2002) has reported that over exploitation of the *Pterocarpus marsupium* for its various useful applications coupled with low germination ability has included in the list of depleted plant species.

Sharad Tiwari, and Kanchan Singh (2003) inoculated nodal segments of *Pterocarpus marsupium* Roxb. on seven different media composition. Viz MS, B5 and White's with out growth hormones (MS₀₀, B5₀₀ and Wh₀₀) each supplemented with 3.0 mg l⁻¹ BAP and 0.5 mg l⁻¹ NAA (MSBN, B5BN, WHBN) and MS media supplemented with 0.2 mg l⁻¹ IBA (MSIB). They studied seed germination improved in all the media and found MS combination were the best (95-100%). They also reported that maximum number of shoot induction per explant was in MS₀₀ (3.25) followed by MSIB (2.26). According to them maximum nodes per shootlet were observed in medium MSIB (4.95), while shoot length was maximum in MSIB (2.92 cm) followed by MS₀₀ (2.41 cm).

Chand and Singh (2004) has raised this plant in *in vitro* by using cotyledonary nodal explants from 20 day old axenic seedling. They obtained the regeneration frequency 85% and 9.5 shoots per explant on MS medium supplemented with 4.44 μM BA and 0.26 μM NAA.

Anis et al., (2005) has reported that propagation and multiplication of *Pterocarpus marsupium* through tissue culture technique is urgently needed. Micropropagation offers a rapid means of producing a large number of clonal plants which can be used for afforestation and conservation of elite and rare germplasm.

Husain et al., (2007) have developed a protocol to regenerate the *Pterocarpus marsupium* using 'Thidiazuron'. MS medium supplemented with 0.1-10 μM thidiazuron successfully produced the multiple shoots from cotyledonary nodes and obtained highest regeneration frequency 90% and much maximum number of shoots 15.2±0.20 per explant on MS medium supplied with 0.4 μM thidiazuron.

Bharmukh and Nikam (2008) studied the seed germination of *Pterocarpus marsupium* and reported that 30 min. scarification treatment with concentrated H₂SO₄ is found fruitful to induce seed germination up to 85% with highest vigour index of 3.3.

Husain et al., (2008) propagated this plant through tissue culture technique using nodal explants from 18 day old

axenic seedling. They obtained highest regeneration frequency (85%) and maximum number of multiple shoots (8.6). The length of the shoot was increased on MS medium supplied with 4.0 μ M 6-benzyl-adenine (BA), 0.5 μ M indol three acetic acid (IAA), and 200 μ M adenine sulphate (ADS).

Husain et al., (2010) achieved Somatic embryogenesis (SE) from hypocotyl-derived callus cultures in *Pterocarpus marsupium*. Ninety percent of hypocotyl explants excised from 12-day old in vitro germinated axenic seedlings produced callus on Murashige and Skoog medium supplemented with 5 μ M 2,4-dichlorophenoxyacetic acid and 1 μ M 6-benzyladenine (BA). Induction of SE occurred after transfer of callus clumps (200 \pm 20 mg fresh mass) to MS medium supplemented with BA at 2.0 μ M, where a maximum of 23.0 \pm 0.88 globular stage embryos per callus clumps were observed after 4 weeks of culture. Subculturing of these embryos on MS medium supplemented with 0.5 μ M BA, 0.1 μ M α -naphthalene acetic acid and 10 μ M abscisic acid significantly enhanced the maturation of somatic embryos to early cotyledonary stage where 21.4 \pm 0.32 embryos per callus were recorded after 4 weeks of culture. Of 30 – well developed somatic embryos, 16.6 \pm 0.33 germinated and subsequently converted into plantlets on half-strength MS medium supplemented with 1.0 μ M BA. The morphologically normal plantlets with well – developed roots were first transferred to 1/4-liquid MS medium for 48 h and then to pots containing autoclaved soilrite and acclimatized in a culture room. Thereafter they were transferred to a green house, where 60% of them survived.

Phytochemistry studies of *Pterocarpus marsupium*:

In the Indian system of medicine many plants have been used for evaluations of various phytochemicals of which, *Pterocarpus marsupium* popularly known as Bijasar is one of the most potent species (**Rastogi and Mehrotra, 1989**), (**Sivarajan and Balchandran, 1994**) and (**Grover et al., 2002**). Many workers have studied and reported the medicinal potential of *Pterocarpus marsupium*.

Adinarayana and Syamsundar (1982) reported an eudesmane type sesquiterpene alcohol, selin-4(15)-en-1 β , 11-diol from the heart wood of the *P. marsupium*).

Mitra and Joshi (1982) isolated an isoflavon glycoside from the heart wood of the *Pterocarpus marsupium* and identified it as 5, 4'-dimethoxy-8-methylisoflavone. They also isolated three isoflavon glycosides namely retusin 7-glucoside, irisolidone 7-rhamnoside and 5, 7-dihydroxy-6-methoxyisoflavone-7-rhamnoside.

Subba Rao and Mathew (1982) characterized a naturally occurring hydrobenzoin, marsupol, 4,4'-dihydroxy- α -methyl hydrobenzoin and a novel 2-hydroxy-2-benzylcoumaranone, carpucin, characterized as 2-benzyl-2,4',6-trihydroxy-4-methoxybenzo(b) furan-3(2H) from the *P. marsupium* heart wood.

Maurya and Ray (1984) reported that the ethyl extract powered of *Pterocarpus marsupium* wood revealed the presence of following phytochemical constituents: pterostilbene, (2S)-7-hydroxyflavanone, isoliquiritigenin, 7,4-dihydroxyflavone, marsupsin, pterosupsin, p-hydroxybenzaldehyde, and 2R-3- p-hydroxyphenyl- lactic acid

Subba Rao et al., (1984) isolated propterol: A 1, 3-bis (4-hydroxyphenyl) propan-2-ol as one of the extractive of heart wood of *Pterocarpus marsupium*.

Bezuidenhoudt et al., (1987) reported two flavonoid analogue, 8-C- β -D-glucopyranosyl-3, 7, 4- trihydroxyflavone and 3, 7, 4'-tetrahydroxyflavone from the heart wood which are representatives of the first 5-deoxy- C-C-coupled flavonol glucosides, and rare 3'-C- β -D-glucopyranosyl- α - hydroxydihydrochalcone.

Tripathi and Joshi (1988) isolated three compounds from ethyl acetate fraction of *Pterocarpus marsupium*, retusin-8-O- α -L-arabinopyranoside, naringenin, lupeol. The resolution of ethyl acetate extract of the aqueous decoction of dried heartwood of *Pterocarpus marsupium* yielded pterocarpol among other compounds). They also isolated two new flavonoid glycosides from the roots of *Pterocarpus marsupium*, 7-Hydroxy-6,8-dimethyl flavanone-7-O- α -arabinopyranoside and 7,8,4-trihydroxy-3,5-dimethoxy flavanone-4-O- β -D-glucopyranoside.

Mohan and Joshi (1989) analyzed flower of *Pterocarpus marsupium* and reported two aurone glycosides, 4,6,4'-trihydroxyaurone-6-O-rhamnopyranoside and 4,6,4'-trihydroxy-7-methylauron rhamnopyranoside.

Jain et al., (1997) reported a novel 6,7,3',4-tetraoxygenated homoisoflavonoid, which has been characterized as 6-hydroxy-7-O-methyl-3-(3-hydroxy-4-O-methylbenzyl) chroman-4-one from ether soluble fractions of *Pterocarpus marsupium* heart wood and 6-hydroxy-3,5,7,4'-tetra methoxy flavone-6-O-rhamnopyranoside, a flavonol glycoside was characterised from the root (Yadav and Singh, 1998). An aqueous extract of heart wood yielded an isaurone C-glycoside named pterocarposide (Handa et al., 2000).

Suri et al (2003) isolated a novel C-glucoside, 1-(2,6-dihydroxyphenyl)- β -D-glucopyranoside from the aqueous decoction of powdered dried heartwood of *Pterocarpus marsupium*.

Grover et al., (2004) reported two interconvertible diastereomeric epimers 2 α 2 β -hydroxy-2-Phydroxybenzyl-3(2H)-benzofuranone-7-C-glucopyranoside from the heart wood.

Maurya et al., (2004) reported five new glycosides from the aqueous extract of *P. Marsupium*. These are (1) 6-hydroxy-2-(4-hydroxybenzyl)-benzofuran-7-C- β -D-glucopyranoside (2) 3-(α -methoxy-4-hydroxybenzylidene)-6-hydroxy-benzo-2(3H)-furanone-7-C- β -D-glucopyranoside. (3) 2-hydroxy-2-Phydroxybenzyl-3(2H)-6-hydroxybenzofuranone-7-C- β -D-glucopyranoside. (4) 8-(C- β -D-glucopyranosyl)-7,3',4'-trihydroxy flavones (5) 1,2-bis-(2,4-dihydroxy-3-C-glucopyranosyl)-ethane dione.

Maurya et al (2004). Prepared the aqueous extract of heart wood of *Pterocarpus marsupium* and isolated five new flavonoid C-glucosides: pteroside, pteroisaurone, marsuposide, flavon C-glucoside, vijayosin and two known compounds, C- β -D-glucopyranosyl-2,6-dihydroxy benzene and sesquiterpene. In another study, the bark of *Pterocarpus marsupium* was extracted with ethanol in a percolator and the phenolic constituent was identified as (-)-epicatechin. (Dimethyl ether of marsupin was synthesized by Srikrishna and Mathew 2009)

Pharmacology studies of *Pterocarpus marsupium*:-

Sheehan et al., (1983) studied antidiabetic potential of epicatechin in alloxan diabetic rats. In these rats no measurable effects have been noticed in control and (-) epicatechin treated rats and in already attained diabetic condition, the effect of (-) epicatechin was found to be nil.

Chakravarthy et al., (1985) studied that (-)-epicatechin extracted from the bark of *Pterocarpus marsupium* shows cardiac stimulant activity in perfused frog hearts producing increase in force along with increase in rate.

Ahamad et al., (1989) described the insulin like effects of (-) epicatechin. According to him (-) epicatechin increases the c-AMP content of the islets and insulin release. They observed that the conversion of proinsulin to insulin has been due to (-) epicatechin and the effect of (-) epicatechin was more in one month old rats than mature (12 month old rats). They also studied the hypoglycemic activity of the wood.

Jahromi and Ray (1993). Studied that ethanol extract decreased the serum triglyceride, total cholesterol and LDL and VLDL cholesterol levels without affecting the HDL cholesterol level. They found significant effect of liquiritigenin and pterosupin in lowering the serum cholesterol, LDL cholesterol and atherogenic index while; pterosupin was satisfactory in reducing the triglyceride level.

Rizvi et al., (1995) reported the insulin like activity of (-) epicatechin by studying the effect on erythrocyte osmotic fragility. Though the mechanism of action of both (-) epicatechin and insulin are different, they illicit their protective role on red cell osmotic fragility.

Manikam et al., (1997) reported that administration of three Phenolic compounds of *Pterocarpus marsupium* in hyperglycemic rats significantly minimized the blood sugar level. Marsupin and pterostilben were more effective than Pterospin and when compared with metoformin.

ICMR (1998) studied the antidiabetic potential of *P. marsupium* at multi-center level and hypothesized that, the plant significantly reduced blood glucose level without any side effects in non-insulin dependent diabetes mellitus or newly diagnosed mellitus.

Rizvi and Zaid (2001) studied the effect of insulin and (-) epicatechin on glutathione content in normal and Type-2 diabetic erythrocytes. They found that the glutathione content was lower in Type-2 diabetic erythrocytes than normal while, the insulin treatment both at 1mm and 10mm increased the glutathione level in normal and diabetic Type-2 patient. They also noticed that the (-) epicatechin treatment also increased the glutathione content at 1mm but did showed dose dependant effect like insulin and was ineffective below 1mm concentration. The effect of (-) epicatechin was remarkable at 1mm and 10mm when compared to insulin.

Grover et al., (2002). Studied the effect of aqueous extract of *P. marsupium* on glycogen content of tissue. According to them increase in glycogen content in renal and decrease in glycogen content in hepatic and skeletal muscle was partly prevented by aqueous extract of *Pterocarpus* treatment. Alterations in the activities of hexokinase, glucokinase and phosphofructokinase in diabetic and control were corrected by *Pterocarpus marsupium* extract. **Vats et al.**, (2002) reported the anti-cataract activity of the *P. marsupium* and *Trigonellla foenum* seed extract. They noticed that administration of aqueous extract of *P. marsupium* decreased the opacity index, indicating anticataract potential of the plant. Further, they also noticed that in cataract examined rats, it showed significant effect on body weight and blood glucose values. **Vats et al.**, (2002) found that absolute ethanol extract fraction dissolved in ethyl acetate was protective in lowering the blood sugar level and increased the insulin level in the blood sugar in alloxan diabetic rats while, aqueous extract of *P. marsupium* lowered blood sugar level from 72.32 ± 5.62 to 61.35 ± 1.2 mg in alloxan diabetic rats. The drug also lowered the blood glucose level from 202 ± 5.44 to 85.11 ± 11.28 mg when administrated daily.

Zaid et al., (2002) reported that lowered activities of erythrocytic membrane Ca^{++} -ATpase leads to cardiomyopathy indicated by reduction in contractibility, relaxation, cardiac work and diastolic complications in Type-2 diabetes mellitus. When the normal and diabetic type -2 patients treated with 1mm (-) epicatechin, the Ca^{++} -ATpase activity increased both in normal and diabetic type-2 patients.

Kar et al., (2003), also evaluated the hypoglycemic activity of vacuum dried 95% ethanolic extract, when administrated at a dose 250mg/ounce, twice or thrice daily found effective in lowering the glucose level in the blood to normal in alloxan diabetic rats.

Joshi et al., (2004) reported that *Pterocarpus marsupium* decreased the blood glucose levels both in normal and non-insulin dependent diabetic (NIDDM) rats. In NIDDM rats the propensity was increased to gastric ulcer which was induced by cold resistant stress, aspirin, and ethanol and pylorus ligation. They observed that the *Pterocarpus marsupium* did not show significant protection from the gastric ulcer in case of normal rats due to above inducers, but it protected the mucosa in NIDDM rats by affecting the mucosal offensive and defensive factors.

Vats et al (2004) demonstrated the anti- cataract activity of aqueous extract of *Pterocarpus marsupium* bark. This was evident from the decreased opacity index in the alloxan induced diabetic rats.

Hougee et al. (2005) performed a study in which a PGE₂ inhibitory effect of a commercially available extract of *Pterocarpus marsupium* characterized by pterostilbene, was demonstrated . *Pterocarpus marsupium* extract decreases PGE₂ production indicating COX-2 specific inhibition.

ICMR (2005) has evaluated the efficacy of Vijaysar (*P. marsupium*) in newly non-insulin dependent diabetes mellitus. They reported that blood glucose level and mean HbA1c levels were decreased significantly from 151-216mg/dl to 32-45mg/dl and 9.8 to 9.4 % respectively indicating the utility of Vijaysar in NIDDM patients.

Mankani et al., (2005) studied the hepato-protective activity of aqueous and methanolic extract of marsupium wood against carbon tetrachloride induced hepatotoxicity. They found marked increase in total bilirubin, serum transaminase and serum alkaline phosphatase activity caused due to carbon tetrachloride toxicity were restored by aqueous and methanolic extract and later it was more effective in restoring the altered levels of these parameters.

Gayathri and Kannabiran (2008) evaluated the ameliorative potential of aqueous extract of *P. marsupium* bark in streptozotocin (STZ) induced diabetic rats. Oral administration of aqueous extract normalized the glycosylated hemoglobin, total cholesterol, triglycerides and LDLcholesterol. Increased levels of various enzymes such as aspartate transaminase, alanine transaminase, alkaline phosphatase, glutamyle transferase and ceratine kinase were brought to normal level. They also indicated that the prominent effect of metabolic alterations in experimentally induced diabetes mellitus was due to restoration of the plasma insulin and liver glycogen levels.

Mohire et al., (2007) evaluated the cardiotoxic activities of aqueous extract of heart wood. The extract *Pterocarpus marsupium* protects cardiac muscle at 4mg/ml, as compared to standard drug Digoxin (0.5mg/ml). 5, 7, 2-4 terahydroxy isoflavone 6-6-glucosides which is protective in cardio vascular disease .

Pan et al., (2007) studied anti-cancerous potential of pterostilben. Further, stilbens isolated from berries and grapes posses anticancer properties and used to cure colon cancer in men and women (Rimando and Suh, 2008).

Rajalakshmi et al., (2008) studied the antioxidant activity of *P. marsupium* on isolated frog heart and found that the plant extract protected the cardiac muscles from oxidative stress induced by H₂O₂. While, the cardiac arrest time was prolonged by 14 minutes in the presence of plant extract than control, indicating the antioxidant activity of the methanolic extract of *Pterocarpus marsupium* bark.

Das et al (2011) performed a study in which they showed that *Acacia catechu* (L.) Willd. (Mimosaceae), *Pterocarpus marsupium* Roxb. (Papilionaceae), *Toddalia asiatica* (L.) Lamk. (Rutaceae) and *Ventilago denticulata* Willd. (Rhamnaceae), growing in West Midnapore district of West Bengal, an eastern state of India, yields dyes which showed strong antimicrobial activity against four bacterial strains (*Bacillus cereus*, *Escherichia coli*, *Klebsiella pneumoniae* and *Vibrio cholerae*). Among the dyes of the four plants tested, *Toddalia asiatica* showed the highest

antimicrobial activity with the inhibition zones ranging from 14 to 16 mm. Thin layer chromatography of the dyes, which were either substantive dyes or adjective type of dyes requiring mordents for fixing to fabrics, showed that most of these pigments were composed of xanthophylls and anthocyanin. The extracts of different plant parts are also used for various medicinal purposes by the local tribal people.

Udaysing Hari Patil et al (2011) Evaluated bactericidal potential of methanolic extract of stem bark (Apical bark, middle bark and Mature bark) of *Pterocarpus marsupium* with respect to pathogenic bacteria *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoneae*, *Salmonella typhi*, *Proteus mirabilis* and *Micrococcus sp*. They found that the methanolic extract of apical stem bark was effective than the middle bark and mature bark in inhibiting the growth of all bacteria. The bacterium *Staphylococcus aureus* was most sensitive among all the bacterial species studied.

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