Review of Literature
2.1. Pesticides

Pesticides constitute a heterogeneous category of chemicals specifically designed for the pest controlling for weeds or plant diseases. Their application is still the most effective and accepted means for the protection of plants from pests, and has contributed significantly to enhance agricultural productivity and crop yields. Organophosphate insecticides are extensively used in both agricultural and landscape pest control. The Nazi government enlisted them in developing OP nerve gases like sarin, soman and tabun and they used in militarily service. After World War II, American companies gained access to information from German laboratories and began to synthesize OP pesticides in large quantities. (Source: Toxipedia). For more than 45 years, OPs have been used as insecticides in the United States. In 1997, 66 poison control centers were established in the United States. It’s indicated that the organophosphorous insecticides were involved in more poisonings than any other single class of pesticide (Litovitz et al., 1998). These compounds have various other compound-specific chronic effects, including delayed polyneuropathy, immunotoxicity, carcinogenesis, and endocrine, developmental, and reproductive toxicity (Astroff et al., 1998; Crammer et al., 1978; Reuber, 1985; Sultatos, 1994).

2.1.1. Chlorpyrifos (CPF)

Chlorpyrifos is one of the most widely used organophosphorus insecticides both in agricultural and non-agricultural arenas (Aspelin, 1997). Chlorpyrifos is a broad-spectrum organophosphorus pesticide, displaying insecticidal activity against a wide range of insect and arthropod pests. Methodologically chlorpyrifos has a minimum purity of between 940 and 990 g/kg. Manufacturing concentration contain a minimum of 180 to 190 g/kg chlorpyrifos. Chlorpyrifos is the active component in a wide array of pesticide formulations. According to the U.S. Environmental Protection Agency (EPA), at one time there were over 400 different commercial products that contained chlorpyrifos as an active ingredient.

Chlorpyrifos is selected for use in the vegetable industry due to its persistence in the soil. This aims to controlling the ground dwelling insect larvae which only return to the surface to feed every few days. In the fruit growing industries, chlorpyrifos is used for the control of scale insects in mangoes and stone fruit. It is also used in low-chill stone fruit, after harvested ‘crawler’ insects population are increased in stone fruit, CPF can damage this ‘crawler’ insects population.
2.1.2. Physiochemical properties of Chlorpyrifos

Chlorpyrifos, O,O-diethyl-O-(3, 5, 6-trichloro-2-pyridinyl)-phosphorothioate (CAS Registry Number 2921-88-2; Molecular Weight 350.6, chemical formula C9H11Cl3NO3PS), is a broad spectrum organophosphate (OP) pesticide. Technical chlorpyrifos (CPF) is a white crystalline solid with a melting point of 41.5–42.5ºC. Chlorpyrifos is stable in neutral and acidic aqueous solutions; however, stability decreases with increasing pH. Chlorpyrifos is practically insoluble in water, but is soluble in most organic solvents such as acetone, xylene and methylene chloride.

2.1.3. Effect of organophosphate on reproductive system

Pesticides such as organophosphate (OP) insecticides are one of the most important environmental contaminants; they remain inevitably present as residues in food from both vegetable and animal origins (Bolognesi and Morasso, 2000). Organophosphorus compounds are widely used in agriculture as insecticides, acricides and also in medicine industry (Uzunhisarcikli et al., 2006) and has been shown to have toxic effects in human and animals (De-Bleecker et al., 1993; National Research Council., 1993; WHO., 1997; Littovitz et al.,1997; Tsatsakis et al., 1998; Eskenazi et al., 1999).

Many researchers have produced evidence of toxicity due to organophosphate exposure. It was established that due to the toxicity of methyl parathion upon different day’s treatment, there was decrease in weight of seminal vesicle, epididymis and prostate gland in male Wister rats (Narayan et al., 2006). According to Choudhary et al., (2008), Malathion toxicity severely decreased the testis weight of male rats, and the seminal vesicle and epididymis weight of male rats also significantly decreases due to the treatment of methyl parathion in a long time exposure (Prashanthi et al., 2006). Due to exposure of Pirimiphos-methyl with the dose of 41.67, 62.5 or 125 mg/kg/day, (Ngoula et al., 2007) the testis and seminal vesicle weight of male Wister rates decreased significantly. Similarly, Afaf and Kashoury (2009) reported that the profenofos exposure at the dose 23.14 mg/kg body weight decreased the body weight of rats and also testis and epididymal weight. Due to endosulfan exposure, 1.0 mg/kg/bd body weight for 30 days treatment showed a significant reduction in rat’s body weight, testis, epididymis and seminal vesicle weight (Chitra et al., 1999). Ahammad Sahib et al., (1981) revels those 48 hours 2 ppm malathion exposure decreased the body weight of Tilapia mossambica. Similarly Uzunhisarcikli et al.,
(2007) held that, 24 h methyl parathion exposure @ dose of 0.28 mg/kg/bw showed no significant changes in male rats body weight, absolute and relative testis weight but in 4 and 7 weeks treatment significantly decreased the body weight, absolute and relative testis weight. Abd El-Aziz et al., (1994) demonstrated that after a prolonged exposure, carbofuran potentially damage the male reproductive system in rats.

Pesticides act as a toxicant in male reproductive system, especially on sperm related parameters. According to Narayan., et al (2006), one of the conspicuous finding was the reduction of sperm density in the lumen of epididymal tubule and the structural changes in the areas of the epididymis, besides effects on intra-epithelial nuclei and epithelial necrosis were seen in the epididymis due to the methyl parathion exposure with a dose of 3.5 and 7 mg/kg upon 14 days treatment. According to Hallegue et al., (2003), due to dieldrin toxicity significantly decreased the epididymal sperm mobility and number of epididymal spermatozoa in male Wister rat treated with a dose of 3 and 6 mg/kg/d respectively. 125 mg/kg/day Pirimiphos-methyl exposure for 90 days treatment significant decreased sperm motility and sperm density in adult male rats (Ngoula et al., 2007). In a similar experiment by Choudhary et al., (2008), malathion exposure for 60 days in male wister rats showed low caudal epididymal sperm density and sperm motility. Similarly in another experiment Prashanthi et al., (2006) found that when methyl parathion was administrated @3.5 and 7 mg/kg/day for 14 days, the sperm density moderately decrease. In long term exposure for 77 days @ 0.5, 0.75, 1 and 3.5 mg/kg/day, as compared to short term exposure of 3.5 mg/kg/day doses for17 days, the long term exposure decreased the sperm density significantly than short term exposure. According to Afaf and Kashoury, (2009), Sperm count, sperm density and spermatozoal motility also decreased in 23.14 mg/kg profenofos treated male rats during 60 days treatment. Uzunhisarcikli et al., (2007) reveled that, the 24 h exposure of male rats to methyl parathion at the dose of 0.28 mg/kg body weight significantly decreased the testicular sperm count, sperm morphology and sperm motility, similarly in 4 and 7 weeks treatment showed the testicular sperm count, normal sperm morphology and sperm motility. A significant decreased in sperm density in testis, cauda epididymis and sperm motility were found in mancozeb treated male rats for 30 days exposure (Joshi et al., 2005). According to Pant et al., 1995, reduction of the epididymal sperm count, sperm motility and increment in sperm morphological abnormalities like head, neck and tail regions of spermatozoa
were observed when the rats were exposed to 0.2, 0.4, and 0.8 mg carbofuran kg⁻¹ body weight for 60 days exposure.

Changes of histopathological structure are one of the most important features of toxic damage. Histopathological examination of pirimiphos-methyl treated rats showed a less amount of seminiferous tubules, immature cells in the lumen, enlargement of interstitial tissue (oedema) and damaged sertolian cells in testicular architecture during 90 days of exposure (Ngoula et al., 2007). The nuclear pyknosis was seen in the epithelium of epididymis and also nuclear degeneration, immature germ cells were found in lumen in Methyl parathion treated rats (Narayan et al., 2006). In a report of Hallegue et al., (2003), dieldrin had a pronounced effect in the alteration of the architecture of testis, leading to a dramatically reduction of spermatozoa production in lumen and 90% of somniferous tubules sections were significantly reduced their tubular diameters. During 60 days Profenofos exposure, rats showed a structural change in testicular architecture. Some degenerative changes were found in seminiferous tubules associated with low luminal spermatozoal concentration in testis (Afaf and Kashoury., 2009). According to Uzunhisarcikli et al., (2007/8), methyl parathion induced rat’s shows no histopathological changes after 24h in testicular tissue. However, upon in 4 weeks methyl parathion exposure showed lesser number of spermatogenic cells and in 7 weeks treatments showed some abnormalities in seminiferous tubules, especially necrosis in some seminiferous tubules and also edema in interstitial tissue was observed.

Marked histopathological changes were observed in the seminiferous tubules after 500 mg/ kg mancozeb treatment in male rats during 30 days exposure. These changes are indicate the damaged of seminiferous tubules, which showed complete spermatogenic arrest, lumen contained cellular debris and is devoid of sperms (Joshi et al., 2005). The same result was found by Pant et al., (1995), which, histologically identified toxicity of carbofuran on testis of experimental animal. These identifications are consisted as oedema, congestion, damage to sertoli cells and germ cells, along with the accumulation of cellular debris and presence of giant cells in the lumen of a few seminiferous tubules.

Organophosphates as an insecticides are toxic to any animals. Considerable work has been done for investigated the mode of action and mechanism of organophosphate compound for the identification of the intensity of toxic damage. Biochemical changes are positive indicator of pesticides poisoning particularly for
ACP and ALP are very important parameters. According to Narayana et al., (2006), Methyl parathion affects the ACP activity in the epididymis due to its toxicity. During 30 days treatment of endosulfan with the dose of 1.0 mg/kg body weight also the ACP and ALP activity were significantly lower in male rats (Chitra et al., 1999). According to Prashanthi et al., (2006), the acid phosphatase concentration also decreased in methyl parathion treated male rats. A significant reduction in the alkaline phosphatase activity and an rise in the acid phosphates activity showed in malathion treated male rats during 60 days of exposure (Choudhary et al., 2008). During 60 days profenofos exposure the alkaline phosphates and acid phosphates level were decreased (Afaf and Kashoury, 2009). According to Joshi et al., (2005), a significant decrease was found in testicular alkaline phosphate activity but also a significant increment was found in acid phosphate activity.

Another very essential toxicological tress related parameter is uric acid. According to Sexena and Sexena, (2010), a prominent elevation was found in serum uric acid level in both technical and formulated cypermethrin (synthetic pyrethroid used as an insecticide) exposure on rat for 7, 14 and 21 days. According to Mahmoud and Salem, (2011) some mancozeb (fungicide) treated (Anadol, Blanko,Deltathin, Nemic, Tazoloin) rats increased the serum uric acid level significantly for eight weeks treatment. According to Uzun and Kalender, (2011), shows a significant increment was found in serum uric acid level of 27 mg/kg malathion induced male rats for 28 days treatment.

Narayan et al., (2006) shows due to Methyl parathion exposure decreased the total protein level in the accessory male reproductive organ of rats. Simultaneously Afaf and Kashoury, (2009), showed an increment of total protein level of profenofos treated rats. In a report of Chitra et al., (1999), the protein content was significantly increased due to endosulfan exposure. The same things was located in (Choudhary et al., 2008), malathion exposure increased the total protein content of adult male rats at dose levels of 50,150 and 250 mg/kg body weight for 60 days. Similarly due to pirimiphos-methyl exposure (Ngoula et al., 2007), the total protein contents of the testis was increased in 6.2 mg /kg/day treatment in adult male rats. A significant increase in testicular protein was found in mancozeb treated male rat for 30 days treatment (Joshi et al., 2005).

Copper, zinc, iron – metals, those components or activators are playing an important role in animal growth, development and reproduction (Underwood, 1977).
Deficiency or excess of any of these elements in tissues may lead to various diseases and even death (Rogers et al., 1985; Stemmer et al., 1985; Prohaska, 1987). Cadmium concentrations in the tissues of the animals were elevated. Highest cadmium concentrations were found in gill and liver. Copper concentration in some tissues of Caranx crysos varied significantly among the others. Chromium, Nickel concentration in the tissues was also found to be elevated condition of Mugil cephalus, Mullus barbatus and Caranx crysos (Kalay et al., 1999). During 60 days Profenofos treatment produced significant increment in iron (Fe), copper (Cu), zinc (Zn), as well as selenium (Se) levels in testicular tissues (Afaf and Kashoury, 2009).

According to Choudhary et al., (2008), serum testosterone level was decrease in melathion induced rat. El-Kashoury and Ei-far, (2004), revel that the administration of profenofos at @23.14 and 46.30 mg/kg body weight for 28 days and 60 days, significant decrease in thyroid hormone levels of adult rats (Afaf and Kashoury, 2009). Dieldrin exposure (3 and 6 mg/kg doses) for 10 days the serum testosterone level was severely decreased on male rats (Hallegue et al., 2003). Also a marked decline present in testosterone level of mancozeb treated male rat for 30 days treatment (Joshi et al., 2005).

2.1.4. Reproductive toxicity of Chlorpyrifos

In a dietary study, Sprague-Dawley rats were fed chlorpyrifos at doses of 0, 0.03, 0.1 and 0.3 mg/kg/d for the first generation, and 0, 0.1, 0.3 and 1.0 mg/kg/d for the second and third generations. The F3B foetuses were used for teratological examination. Clinical signs of toxicity were not seen in any parents or offspring. Parental body weights were not significantly affected by treatment, and food consumption was variable but unaffected by treatment in either sex. The fertility, gestation and lactation indices were comparable between groups and generations. The viability index was decreased at 1.0 mg/kg/d. This effect on pup viability at 1.0 mg/kg/d was also seen when comparing mean litter size at day 21. In the dams producing the F3B pups, mean bodyweight gain showed a dose-related increase; at 0, 0.1, 0.3 and 1.0 mg/kg/d, body weight gains for gestation days 0-20 were 114, 120, 123 and 126 g respectively. In this generation, there were no treatment-related effects on fertility (% pregnant), mean number of corpora lutea or implantations, viable litter size or pup weight following caesarean section. Skeletal examination of pups revealed common minor variants such as incomplete ossification of sternabrae, and the occurrence of extra ribs. Based on the reduction in pup viability seen in each
generation at 0.3 and 1.0 mg/kg/d, the decreased plasma and RBC cholinesterase activity seen at 1.0 mg/kg/d in males and females, and the decreased RBC cholinesterase activity seen in 0.3 mg/kg/d females, the NOEL for this study was 0.1 mg/kg/d (Thompson et al., 1971).

Quellette et al., (1983) shows experimental groups containing 24–29 pregnant Fischer 344 rats per group were treated orally with 0, 0.1, 3, or 15 mg/kg-day (in corn oil [vehicle], by gavage) on days 6–15 of pregnancy. There was no evidence for increased embryo-/fetotoxicity (e.g., prenatal mortality or decreased fetal weight) up to the highest dose level tested, and the incidence of malformations was not increased at any dose. There was no evidence for impairment of the reproductive functions (fertility) within the F0/F1 or the F1/F2 generation, up to a daily dose of 5 mg chlorpyrifos/kg, which, according to previous studies, represents the threshold for inducing effects on both neonatal development (body weight and survival) and maternal toxicity. Animals mated with normal frequency and exhibited normal pregnancies, offspring and lactation. A few deviations from controls were reported for offspring of mothers exposed to 5 mg/kg but were not consistent in F1 pups and F2 pups (Breslin et al., 1996).

2.1.5. Male Reproductive toxicity of Chlorpyrifos

It has also been hypothesized that chlorpyrifos could inhibit testosterone metabolism (Rose and Hodgson, 2005). The concentration of chlorpyrifos necessary to produce such inhibition in vivo must be established in order to ascertain its possible physiological relevance. The reproductive effect of dermal exposure of chlorpyrifos was investigated in young bulls. The animals were treated with an undetermined amount of Dursban-44 and sperm parameters were analyzed. An unspecified increase in nonmotile sperm, and decreased sperm mobility and ejaculate volume, were observed at 6 months postexposure, whereas no effect was observed 7–12 months postexposure (Everett, 1982).

Organophosphate insecticides chlorpyrifos is highly used and the low concentration of chlorpyrifos pollution in the environment becomes a common phenomenon (Joshi et al., 2003). It is reported that the CPF created a potential hazards to human health, it also linked with human genital deformities (Whorton et al., 1997) and damage the reproductive system of male rats (Viswanath et al., 2010). According to (Linlin et al., 2013), chlorpyrifos has adverse effect on reproductive system of male rats due to 90 days of exposure at 0, 2.7, 5.4 and 12.8mg/kg body
weight. In this experiments the testicular histology testosterone level, testicular sperm counts and sperm morphology were significantly altered due to CPF toxicity. Due to CPF exposure @ 23.43, 21.40, and 17.43 for 28 days showed a significant decrease in testis weight. The activity of alkaline and acid phosphates and lactate dehydrogenase (LDH) and total protein level were significantly increasing (Afaf et al., 2010). In male reproduction a dose dependent decrease was found in 5 and 50 ppm chlorpyrifos methyl treatment. The weight of seminal vesicle and testis, sperm motility, sperm count, serum testosterone level, LH level and FSH level were significantly decreased and simultaneously abnormal sperm morphology was increase for 65 days of exposure (Zidan, 2009). According to Joshi et al., 2007 @ 7.5, 12.5 and 17.5 mg/kg/d of CPF exposure for 30 days, showed a marked reduction in epididymal and testicular sperm count, decrease in serum testosterone concentration and also histopathological examination shows many degenerative changes in male reproductive organs.
2.2. *Emblica officinalis* Geartn

Ayurveda, which is the oldest health system in the world, appreciates and uses of *Emblica* promote positive health. Amla (*Emblica officinalis*) is called Amalaki and Dhartiphala in Sanskrit. In Hindu religious mythology the tree is worshipped as the Earth Mother as its fruit is considered to be so nourishing as to be thy nurse of mankind (Onions, 1994). It’s extensively used as a rejuvenator in Ayurveda. It is also used widely in combination with other two herbal products (chebulic and belleric) as called Triphala. Amla is indeed, the key ingredient in the popular Ayurvedic recipe of Chyavanprash. More than anything, it may be called as "King of Rasayana" (rejuvenation), owing to its multiple health benefits. Amla possesses a vast ethnomedical history and represents a phytochemical reservoir of heuristic medicinal value. It is one of the oldest oriental medicines mentioned in Ayurveda as potential remedy for various ailments. Amla is a gift of nature to mankind. It is an indispensable part of the Ayurvedic and Unani system with amazing remedial qualities. In India, it is common to eat gooseberries steeped in salt, water and turmeric to make the sour fruits palatable. There are two varieties of Amla - cultivated gramya and vanya. The wild amla is small, while cultivated amla is big, smooth and juicy. Chemical composition of the amla fruit contains more than 80% of water. It also has protein, carbohydrate, fiber and mineral, gallic acid and also content steroid (Gupta et al., 2013) which are a potent polyphenol. Vitamin C is important for human beings. It is necessary for the synthesis of the inter-cellular cementing substance which is responsible for keeping the cells of the body together. The amla fruit is reported to contain nearly 20 times as much vitamin C as orange juice. The edible amla fruit tissue has 3 times the protein concentration and 160 times the ascorbic acid concentration of an apple. The fruit also contains higher concentration of most minerals and amino acids than apples (Singh et al., 2011). It possesses antioxidant and free radical scavenging activity.

2.2.1. Regional name of *Emblica officinalis*

**English**: Emblic myrobalan, Indian Goose berry  
**Sanskrit**: Aamalaki  
**Hindi**: Amla  
**Kannada**: Nelli Kayi  
**Marathi**: Amla
Gujarati: Ambla
Malayalam: Nelli Kayi
Tamil: Nelli
Telugu: Usirikaya
Kashmir: Aonla

Vernacular names: *Emblica officinalis*

Botanical Name: Phyllanthus emblica

2.2.2. Essential part of *Emblica officinalis*

Dried fruit, leaf extracts, roots (Asmawi et al, 1993).

2.2.3. Botanical description

A small to medium sized deciduous tree, 8-18 meters height with thin light grey bark exfoliating in small thin irregular flakes, leaves are simple, sub sessile, closely set along the branchlets, light green having the appearance of pinnate leaves; flowers are greenish yellow, in axillary fascicles, unisexual, males numerous on short slender pedicels, females few, subsessile, ovary 3-celled; fruits globose, fleshy, pale yellow with six obscure vertical furrows enclosing six trigonous seeds (Indian Medicinal Plants, 1997).

2.2.4 Geographical distribution

It’s found throughout India, the sea-coast districts and on hill slopes up to 200 meters, also a cultivated in plains (Indian Medicinal Plants, 1997).

2.2.5. Traditional use

The fruits are sour, astringent, bitter, acrid, sweet, cooling, anodyne, carminative, ophthalmic, laxative, digestive, stomachic, alterant, aphrodisiac, rejuvenative, diuretic, antipyretic and tonic. They are useful in vitiated conditions of tridosha, diabetes, cough, asthma, bronchitis, cephalalgia, ophthalmopathy, dyspepsia, colic, hyperacidity, flatulence, peptic ulcer, erysipelas, skin diseases, leprosy, haematogenesis, inflammations, anemia, emaciation, hepatopathy, jaundice, strangury, diarrhoea, dysentery, hemorrhages, leucorrhoea, menorrhagia, cardiac disorders, intermittent fevers and greyness of hair (Nadkarni, 1993; Indian Medicinal Plants, 1997).

2.2.6. Phytochemical constituents and active ingredients
The fruit is rich sources of quercetin, phyllaemblic compounds, gallic acid, tannins, flavonoids, pectin, vitamin C and also contains various polyphenolic compounds. A wide range of phytochemical components including terpenoids, alkaloids, flavonoids, and tannins have been present (Kim et al., 2005; Arora et al., 2003). The fruits, leaves and bark are rich sources of tannins. The root contains ellagic acid and lupeol and bark contains leucodelphinidin. The seeds yield a fixed oil (16%) which is brownish-yellow in colour. It has the following fatty acids: linolenic (8.8%), linoleic (44.0%), oleic (28.4%), stearic (2.15%), palmitic (3.0%) and myristic (1.0%) (Thakur et al., 1989). The phytochemicals of this plant include hydrolysable tannins (Emblicanin A, Emblicanin B, punigluconin, pedunculagin) (Ghosal et al., 1996), flavonoids (Kaempferol 3 O alpha L (6” methyl) rhamnopyranoside, Kaempferol 3 O alpha L (6” ethyl) amnopyranoside), alkaloids (Phyllantidine and phyllantine). Gallic acid, ellagic acid, chebulinic acid, quercetin, chebulagic acid, corilagin together with isostrictinnin, were isolated from the fruit of emblica (Ali et al., 2011). The seeds of P. emblica contain fixed oil, phosphatides and small quantity of essential oil. In addition, the leaves contain gallic acid, ellagic acid, chebulagic acid and chebulinic acid. Phyllaemblic acid, a novel highly oxygenated norbisabolane were isolated from the roots of P.emblica and its structure was fully characterized by spectroscopic andchemical means (Ali et al., 2011). Ellagic acid and lupeol are present in roots of P.emblica (Kapoor, 1990; Rastogi and Mehrotra, 1993).

Table (1). Chemical Constituents of Amla:

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<th>S.No</th>
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<td>Alkaloids</td>
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<td>Phenolic compounds</td>
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<td>4.</td>
<td>Amino acids</td>
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<td>Carbohydrates</td>
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<td>Vitamin C</td>
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<td>Flavonoid</td>
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<td>8.</td>
<td>Ellagic acid</td>
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<td>Chebulinic acid</td>
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<td>10.</td>
<td>Emblicanin-A</td>
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<td>11.</td>
<td>Emblicanin-B</td>
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<td>12.</td>
<td>Citric acid</td>
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<td>13.</td>
<td>Gallic acid</td>
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<td>14.</td>
<td>Punighuconin</td>
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<td>Ellagotannin</td>
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<td>16.</td>
<td>Quercetin</td>
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<td>Trigallarly glucose</td>
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<td>19.</td>
<td>Pedunculagin</td>
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<td>20.</td>
<td>Steroid</td>
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2.2.7. Pharmacology and clinical studies

Clinical studies were conducted to investigate the effect of crude amla in gastritis syndrome. The crude amla was given in 20 cases in a dose of 3gm, 3 times/day for 7 days. The drug was found effective in 85% of the cases. It was observed that the drug did not have any significant beneficial effect in cases of hypochlorhydria. Only cases of hyperchloridia with burning sensation in abdominal and cardiac regions and epigastric pain were benefited (Rajarama Rao and Siddiqui, 1964)

Alcoholic extract of a plant (1g/kg) has shown an increase in the cardiac glycogen and a decrease in serum GOT, GPT and LDH in isoprotenol pretreated rats, suggesting a cardioprotective action. It showed a reduction in serum cholesterol level (Roy et al., 1991).

The lipid lowering and anti atherosclerotic effects of amla fresh juice were evaluated in cholesterol fed rabbits (rendered hyperlipidemic by atherogenic diet and cholesterol feeding). When rabbits were feed fresh amla juice at a dose of 5 ml/kg/d body weight for sixty days. Serum cholesterol, Triglycerides, phospholipid and Low-density lipoprotein levels were lowered by 82%, 66%, 77% and 90% respectively. Similarly, the tissue lipid level showed a significant reduction following amla juice administration. Amla juice is an effective hypolipidemic agent and can be used as a pharmaceutical tool in hyperlipidemic subjects (Mathur et al., 1996).

It is reported to have anti-cancer properties (Rao and Siddiqui, 1964). The crude extract of Emblica officinalis was reported to counteract hepatotoxic and renotoxic effects of metals due to anti-oxidant activity (Roy et al., 1991).

Safety: The drug is not reported to have any side effects even after prolonged use (Natural Remedies)

2.2.8. Application of Emblica officinalis

Many toxic chemicals are released into the environment through mining, smelting, discharging industrial agricultural and domestic waste, burning fossil fuel and using pesticides in agricultural and domestic purpose. A large number of compounds have been investigated for protective action by different workers, but these protectors are highly toxic at their effective dose level. Emblica is found to be a good herbal protector and at the same time non-toxic, inexpensive and easily
available. During cadmium chloride treated rat, an earlier and faster recovery was noted in *Emblica* treated groups. Due to *Emblica* treatment the cholesterol value shows almost normal level. *Emblica* showed an earlier and faster recovery protection. An increasing value of acid phosphates activity was observed in the drug treated groups but the *Emblica* administered groups shows significant (p<0.001) decreased in acid phosphates level (Chakrawarti et al., 2010). It also protects the mice from the ionizing radiation (Sing et al., 2006)

*Emblica officinalis* commonly known as amla (synonym ñ Indian gooseberry), is one of the fruits which contains an array of bioactive components showing antioxidative property and is widely used in India as a traditional medicine (Ghosal et al., 1996 and Bhattacharya et al., 1999), Ayurvedic herbal formulation and also in Unani medicines. Amla is native to tropical southeastern Asia, particularly in central and southern India (Warrier et al., 1995). Fresh or dried whole fruit is usually used for its medicinal properties. *Emblica officinalis* fruits are used to treat some number of diseases (Nadkarni, 1952; Chopra et al., 1958) and constituent of many hepatoprotective formulations (Antarkar et al., 1980, De et al., 1993). In Unani medicine the dried amla fruits are used to treat hemorrhage, diarrhea, and dysentery (Parrotta, 2001). Amla is a very rich source of ascorbic acid (Singh et al., 2011). Apart from ascorbic acid, it contains fat, phyllemblin, and tannins. Amla fruits also contain minerals such as phosphorus, iron, and calcium (Sidhu et al., 2011). The fruits of *Emblica officinalis* have also been used in treatment of vomiting, hemorrhage, fever, cough, eye inflammation, ulceration, anorexia, emaciation, scurvy, diabetes, jaundice, leucorrhoea, common cold, heart diseases, cancer, hepatotoxicosis, renotoxicosis, heart trouble, ulcer and anemia, (Roy et al., 1991; Sivarajan, 1994; Mathur et al., 1996/82; Ahmad and Chand, 2009; Khan, 2009). Similarly, it has application as antioxidant (Bhattacharya et al., 1999), antibacterial (Godbole and Pendse, 1960) and anti-inflammatory (Asmawi et al., 1993) immunomodulatory, antipyretic, analgesic, cytoprotective, antitussive and gastroprotective (Ahmad and Chand, 2009) properties. Its antioxidant property has been evaluated in many pathological conditions (Bhattacharya et al., 2000). One preliminary study has also evaluated the effects of *E. officinalis* in acute pancreatitis. *E. officinalis* decreased serum amylase levels and improved histopathological structure (Thorat et al., 1995). The pulpy portion of fruit contains gallic acid (1.32%), tannin, sugar (36.10%), gum (13.75%), albumin (13.08%), crude cellulose (17.08%), minerals (4.12%) and
moisture (3.83%). The active components include emblicanin A & B, puniglucanin, 2-ketoglusonolactone, ellagic acid, hexahydroxy diphenic acid and its conjugates (Ahmad and Chand, 2009).

*E. officinalis* is a major component of herbal health tonic Chyavanprash. It is claimed to reduce aging and age-related ailments (Ojha, 1988/85). Preparation of Chyavanprash involves making of decoction from 35 herbs, of which many are known immunomodulatory and antioxidative herbs. Both *E. officinalis* and Chyavanprash are shown to be potent free radical scavenging agents (Jeena and Kuttan, 1995), thereby, its prevent the carcinogenesis and mutagenesis (Jeena et al., 1997, 1999). When animal treated with *E. officinalis* extract, showed an increasing life span. Administration of Chyavanprash significantly reduced the ascites tumour (Chakraborty and Verma, 2009).

Table (2). Cytoprotective activity:

<table>
<thead>
<tr>
<th>S.NO</th>
<th>Cytoprotective activity</th>
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<tbody>
<tr>
<td>1</td>
<td>Antioxidant activity</td>
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<tr>
<td>2</td>
<td>Immunomodulatory Activity</td>
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<tr>
<td>3</td>
<td>Antimicrobial activity</td>
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<td>4</td>
<td>Analgesic activity</td>
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<td>5</td>
<td>Hepatoprotective activity</td>
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<td>6</td>
<td>Medicinal Activity</td>
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<td>7</td>
<td>Antipyretic activity</td>
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<td>8</td>
<td>Gastoprotective activity</td>
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<td>9</td>
<td>Chemopreventive activity</td>
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<td>10</td>
<td>Antitussive activity</td>
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<td>11</td>
<td>Anti-inflamantory activity</td>
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<td>12</td>
<td>Radiopreventive activity</td>
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<td>13</td>
<td>Antimitogenic activity</td>
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<tr>
<td>14</td>
<td>Antiatherogenic activity</td>
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<td>15</td>
<td>Antitumor activity</td>
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<td>16</td>
<td>Apoptotic activity</td>
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<td>17</td>
<td>Hypocholesterolemic activity</td>
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<td>18</td>
<td>Hypolipidemic activity</td>
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<td>19</td>
<td>Adaptogenic property</td>
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<td>20</td>
<td>Ophthalmic activity</td>
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<tr>
<td>21</td>
<td>Antiulcer activity</td>
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Ochratoxins is one of the natural mycotoxins which are secondary toxic fungal metabolites produced mainly by *Aspergillus ochraceus* and *Penicillium verrucosum* (Pitt, 1987). Oral administration of ochratoxin brought about significant reduction in sperm count, sperm motility, sperm viability and fertility rate. When animals were treated with ochratoxin and aqueous extract of *Emblica officinalis*, a significant
amelioration observed as compared to only ochratoxin treated animals. Ochratoxin
also affects the sperm morphological features, but these features were normalized
when aqueous extract of Emblica administered. Emblica officinalis (EO) mitigate the
morphological alterations (Chakraborty and Verma, 2009).

2.2.8.1. Uses of Emblica Officinalis in Diabetes

Regarding high blood sugar, orally administrated Emblica extracts (100 mg/kg
body weight) can reduce the blood sugar level. Tannoids are active ingredient of
Emblica which delaying the development of diabetic cataract in rats (Suryanarayan
et al., 2007).

2.2.8.2. Activity of Emblica officinalis in Liver

EO fruits have been reported to use for hepatoprotection in Ayurveda
(Bhattacharya et al., 2000). Its have a protection against ethanol induced liver injury
in rats (Pramyothin et al., 2006). The extract of EO and Chyavanaprash shows a
protection against hepatoprotective activity in liver injury.

2.2.8.3. Emblica Officinalis and its Anti-ulcer Activities

A herbomineral formulation of the Ayurveda medicine named Pepticare,
composed of Emblica officinalis, Glycyrrhiza glabra and Tinospora cordifolia was
tested for its anti-ulcer and anti-oxidant activity in rats. Its attributed the anti-oxidant
activity of E.O (Bafna and Balaraman, 2005). Methanolic extract of EO also
applicable against ulcer (Sairam et al., 2002).

2.2.8.4. Roles of Emblica Officinalis in Reducing Cholesterol and Dyslipidemia

Cu (2+)-induced LDL oxidation and cholesterol-fed rats were used to
investigate the effects of Amla on low-density lipoprotein (LDL) oxidation and
cholesterol levels in vitro and invivo. It was concluded that Amla may be effective for
hypercholesterolemia and prevention of atherosclerosis (Kim et al., 2005). Emblica
officinalis and Mangifera indica contains flavonoids which reduce the level lipid in
serum. Both cause the degradation and elimination of cholesterol have been done
through the E.O. (Anila and Vijayalakshmi, 2002).

2.2.9. Antioxidant Activities of Emblica Officinalis

The origin of disease of multifactorial nature is being understood due to the
vitiation in basic heamostatis balance phenomenon in the body. It is being realized
that majorities of the disease are mainly due to the imbalance between activities of
pro-oxidant and anti-oxidant. Proxidant condition dominates either due to increased
generation of free radicals and/or their poor quenching/scavenging into the body. Free
radicals are the fundamental to any biochemical process and represent an essential part of the aerobic life and our metabolism. They are continuously produced by body’s normal use of oxygen such as respiration and some cell mediated immune functions. Naturally, there is a dynamic balance between the amount of free radicals generated in the body and anti-oxidant to quench and or /scavenge them and protect the body against their deleterious effects. It is obvious therefore that any additional burden of free radicals either from environment or produced within the body, can tip the free radical (pro- oxidant) and anti- free radical (anti-oxidant) balance leading to oxidative stress which may result in tissue injury and subsequent diseases. Thus, the oxidant status in human reflects the dynamic balance between the anti-oxidant defence and pro-oxidant conditions and has been suggested as a useful tool in estimating the risk of oxidative damage.

EO was studied against the cold stress-induced alterations in the behavioral and biochemical abnormalities. Triphala administered orally about 1g/kg/animal body weight for 48 days significantly prevented cold stress-induced behavioral and biochemical abnormalities in albino rats. Thus Triphala supplementation can be regarded as a protective drug against stress (Dhanalakshmi et al., 2007).

The administration of ethyl acetate (EtOAc) extract of Amla or Sun Amla (Taiyo Kagaku Co., Ltd., Japan) reduced the elevated levels of urea nitrogen and serum creatinine in the aged rats. Oral administration of this extract significantly reduced thiobarbituric acid-reactive substance levels of serum, renal homogenate and mitochondria in aged rats, suggesting that Amla would ameliorate oxidative stress under aging. The increase of inducible nitric oxide synthase (iNOS) and cyclooxygenase (COX)-2 expression in the aorta of aging rats were also significantly suppressed by EtOAc extract of Amla or Sun Amla extract (Anilakumar et al., 2007).

Vitamin C in EO accounts for approximately 45-70% of the antioxidant activity (Scartezzini et al., 2006). Rats were examined for the antioxidant properties of Amla extracts and its effect on the oxidative stress in streptozotocin-induced diabetes was also reported. The extracts showed free radical scavenging activity in all the tissue. Amla extracts orally administered to the diabetic rats slightly improved body weight and also significantly reduce various oxidative stress. Moreover the decreased levels of albumin in the diabetic rats were significantly improved with this drug. It also significantly improved the serum adiponectin levels. Thus amla can be
used for relieving the oxidative stress and improving glucose metabolism in diabetes (Rao et al., 2005).

The aqueous extract of the fruits of *Terminalia chebula*, EO and *Terminalia belerica* and their equiproportional mixture called Triphala were evaluated for their *invitro* antioxidant activity. Gamma-Radiation induced strand break formation in plasmid DNA (pBR322) was effectively inhibited by Triphala and its constituents. *Terminalia chebula* has greater radical scavenging activity while EO shows greater efficiency in lipid peroxidation and plasmid DNA assay. Their mixture, Triphala, is expected to be more efficient due to the combined activity of the individual components (Naik et al., 2005).

DHC-1, an herbal formulation was made from the important herbal plants like EO, *Bacopa monniera*, *Glycyrrhiza glabra*, *Mangifera indica* and *Syzygium aromaticum* was studied for its antioxidant activity. The protective effect of DHC-1 was studied in isoproterenol-induced myocardial infarction and cisplatin-induced renal damage. DHC-1 possesses a protective effect against both damaged kidneys and heart in rats. This protective effect may be attributed, at least in part, to its antioxidant activity (Bafna and Balaraman, 2005). The plant extract lowered hepatic lipid peroxidation (LPO) and increased the superoxide dismutase (SOD) and catalase (CAT) activities in hyperthyroid mice, exhibiting its hepatoprotective nature. It potentially ameliorates the hyperthyroidism with an additional hepatoprotective benefit (Panda and Kar, 2003).

EuMil is a polyherbal formulation composed of standardized extracts of *Ocimum sanctum*, *Withania somnifera*, *Asparagus racemosus* and *Emblica officinalis* was used as an anti-stress agent to attenuate the various aspects of stress related disorders. It has significant anti-stress and adaptogenic activities, qualitatively comparable to *Panex ginseng*, against a number of behavioral, biochemical and physiological perturbations, induced by unpredictable stress, which has been proposed to be a better indicator of clinical stress than acute stress. The contribution of the individual constituents of EuMil (polyherbal formulation) in the adaptogenic action has been reported (Muruganandam et al., 2002). EO is used to protect the skin from the devastating effects of free radicals, non-radicals and transition metal-induced oxidative stress. It is suitable for use in, anti-aging, general purpose skin care products and as sunscreen (Chaudhuri. 2002). The fruits of EO contain tannoid principles that have been reported to exhibit antioxidant activity *invitro* and *invivo*. Emblicanin-A (37%) and -B
(33%) enriched fraction of fresh juice of EO fruits was investigated for antioxidant activity against ischemia-reperfusion-induced oxidative stress in rat heart. The study confirms the antioxidant effect of EO and also indicated that the fruits of the plant may exhibit a cardioprotective effect (Bhattacharya et al., 2002). The antioxidant activity of EO extract is associated with the presence of hydrolyzable tannins having ascorbic acid-like action have been also reported (Pozharitskaya et al., 2007).

A number of medicinal plants, traditionally used for thousands of years, are present in a group of herbal preparations of the Indian traditional health care system (Ayurveda) named Rasayana identified for their interesting antioxidant activities. EO has been reported for its antioxidant activity (Scartezzini and Speroni, 2002).

2.2.10. Antipyretic and Analgesic Activities of Emblica Officinalis

Extracts of EO fruits possess potent anti-pyretic and analgesic activities. A single oral dose of ethanolic extract and aqueous extract of E.O (500 mg/kg, i.p.) showed significant reduction in hyperthermia in rats induced by brewer's yeast. Both of these extracts elicited pronounced inhibitory effect on acetic acid-induced writhing response in mice in the analgesic test (Perianayagam et al., 2004). This may be due to the presence of tannins, alkaloids, phenolic compounds, amino acids and carbohydrates.

2.2.11. Memory Enhancing Effects of Emblica Officinalis

Amla churna (dust) produced a dose-dependent improvement in memory of young and aged rats. It reversed the amnesia induced by scopolamine and diazepam. Amla churna may prove to be a useful remedy for the management of Alzheimer's disease due to its multifarious beneficial effects such as memory improvement and reversal of memory deficits (Vasudevan and Parle, 2007).

2.2.12. Emblica Officinalis in Reproduction

Pathak et al., 2011, shows in his conclusion of that E. officinalis enhances the sexual activities. This obviously increases longevity, fertility, fecundity, ovarioles number along with developmental time. Finally, it is inferred that there is a linear interrelationship between sexual activities and fitness parameters in experimental culture. “Adding Life to years is better than adding years to life.” So, along with longevity, other reproductive fitness characters of flies were considered for this study so as to explore the hidden principles of Rasayana therapy which improves the quality of the life. Emblica officinalis boosts the male reproductive response and mitigates the toxic effect of pesticides. The protective effect of P. emblica fruit extract against
Clastogenicity induced by lead nitrate on the occurrence of sperm head abnormalities in the germ cells of mice. The results clearly point toward that extract exhibited significant reduction in the frequency of sperm head abnormalities. According to (Madhavi et al., 2007) shows that P. emblica plays a key role in inhibition of heavy metal mutagenesis in mammals.