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Nature versus nurture – plant resources in management of male infertility

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1. ABSTRACT

Male infertility, apart from being a multi-factorial disorder, has no defined etiology in almost half of the infertile men. The complex etiology demands a complex remedy which can heal several ailments together. Currently available specific treatments are largely inefficient in infertility treatment. Medicinal plants present a repertoire capable of providing varied constituents which could be helpful in infertility management. However, the literature on the same is scanty and we have not explored even 1% of the available plant resources. Herein, we present a systematic review of clinical and experimental data on the use of Indian medicinal herbs in the treatment of male infertility. Literature suggests that most of the medicinal herbs exhibit a three dimensional effect of reducing oxidative/psychological stress, fatigue and promoting libido. This review is oriented to identify and highlight aphrodisiac, adaptogenic, anti-oxidant and nutritional properties of these plants and aims at promoting exploration of these valuable medicinal resources.

2. INTRODUCTION

Traditional medicine is defined by the World Health Organization (WHO) as the sum total of all knowledge and practices, whether explicable or not, used in diagnosing, preventing and eliminating physical, mental or societal imbalances. The traditional medicine mainly depends on prevention of illnesses and development of natural resistance to diseases, and thus believes in promotion of general well-being (1). It is documented that 80% of the world's population has faith in traditional medicine, particularly plant drugs for primary healthcare (1). Traditional medicine was in existence long before Western medicine came into practice and is based mainly on the practical experience and observation handed down generation to generation, verbally or in writing (2). India has over 6000 plants used in traditional systems of medicine with 3000 of these recognized officially for their properties. Medicinal herbs have been in use in one form or the other, under indigenous systems of medicine like *Ayurveda*, *Sidha* and *Unani*. The research into safety and efficacy of medicinal plants is mainly done *in vitro* or on

animal models. The inability to identify the active principle and its isolation in most of these plants has hampered clinical trials even after activity was reported in animal models. Despite lack of sufficient data from animal studies, few plants have been systematically tested for management of infertility in human subjects. The promising results obtained in these trials should also promote evaluation of other herbs in animal models followed by human subjects.

Approximately, 15% of human couples are infertile, and almost 50% of these are because of male factors (3). Although overall human fertility does not appear to have declined, there is evidence for a decline in sperm quality (4) and a simultaneous increase in the number of infertile couples for the last few decades. Infertility is defined as the inability to conceive after 1 year of unprotected intercourse (5). The malignancy may be caused by low sperm production (oligozoospermia), poor sperm motility (asthenozoospermia) or abnormal sperm morphology (teratozoospermia); however, generally a combination of these is considered to be the most common cause of male sub-fertility (6). A multitude of factors, such as physical obstruction to sperm release, reduced sperm count or motility, altered sperm morphology, infections and hormonal imbalances have been identified as contributors to male infertility (7). Anatomic defects, endocrinopathies, immunologic problems, ejaculatory failures and environmental exposures are significant causes of infertility (7). Extra-testicular causes of male infertility are less common (5) and specific therapies are readily available for such cases.

Direct testicular injury to male germ cells, Sertoli cells, and Leydig cells is one of the major causes of infertility (5). It is assumed that in about 30% of the cases, male infertility is caused by chromosome aberrations or mutations in genes functioning in male germ line (8). In most of the infertile patients, levels of LH, FSH, T and prolactin hormones are imbalanced (9). Despite identification of the above factors, the etiology of infertility remains unexplained in almost 50% of individuals. Because of etiological heterogeneity, the treatment of male infertility is not straight-forward. The variety of causes discussed above makes the diagnosis difficult and so is the case with treatment. We have earlier reviewed that a little percentage of cases receives specific therapy upon presumptive diagnosis, a good percentage receives empirical therapy and a significant percentage receives trial therapy, adding up to a success rate of not more than 10% (5). Various possible causes and treatment modalities of male infertility are listed in our earlier review (5).

In addition to specific causes of infertility, general causes such as nutritional deficiency, in particular, the deficiency of antioxidants, vitamins, coenzymes and stimulants, lifestyle factors such as type of job, smoking, and exposure to pollutants may also impair male potency or fertility. One common element in most of these is oxidative stress. Recently, several reports have pointed out oxidative stress (OS) as a major cause for infertility in up to 30-80% of the unexplained cases (10). These claims are getting stronger both by the findings of elevated ROS levels in seminal plasma of infertile patients (11) and their good response to antioxidant therapies (12), (13), (14). Oxidative stress is a condition that occurs when the production of ROS overwhelms the antioxidant defense produced against them. In male reproductive pathological conditions, oxidative stress significantly impairs spermatogenesis and sperm function, which may lead to infertility (15). ROS are generated by sperm and seminal leukocytes within semen which may cause infertility by two key mechanisms; i) damaging the sperm membrane, decreasing sperm motility and its ability to fuse with the oocyte and ii) altering sperm DNA by inducing cross-links, resulting in passage of defective paternal DNA on to the conceptus (16).

Though specific antioxidants, stimulants and nutrients such as vitamins are available in the modern medicinal systems, none of them provides as varied constituents as may be required to combat the multi-factorial condition of male infertility. Plant products are the best known alternatives when general and varied effects are desired. A good number of plants are known to be adaptogens (relatively non-toxic substance that acts by increasing resistance of the organism to a broad spectrum of adverse biological, chemical and physical factors, and helps regulate or normalize organ and system function within the organism), are rich in essential nutrients such as vitamins and antioxidants, and may contain several other active ingredients unknown till today. Few recent studies, including ours (13, 14, 17), have shown tremendous potential of plant products in management of male infertility. Several other studies have focused only on selected plants with superficial exploration in most of the cases. We have taken up a review of literature to bring forward the known facts about certain plants with particular emphasis on their aphrodisiac, adaptogenic, anti-oxidant and nutritional properties, and highlight several others bearing promise for use in male infertility management.

3. MEDICINAL PLANTS

3.1. *Asparagus racemosus* (Family: Solanceae, Sanskrit Name: Satavari)

Asparagus racemosus Willd is an important medicinal plant of tropical and subtropical India. Its medicinal usage has been reported in the Indian and British Pharmacopoeias and in traditional systems of medicine such as *Ayurveda*, *Unani* and *Siddha*. The plant is mainly distributed throughout the tropical and subtropical parts of India up to an altitude of 1500 m. The genus *Asparagus* includes about 300 species around the world. The genus is considered to be medicinally important because of the presence of steroidal saponins and sapogenins in various parts of the plant. Out of 22 species of *Asparagus* recorded in India; *Asparagus racemosus* is the one most commonly used in traditional medicine. The root is the part that finds use in various medicinal preparations (18). *A. racemosus is* one of the most commonly used medicinal herb in *Ayurveda* (more than 60 formulations) since time immemorial (19).

Male rats fed with *A. racemosus* root powder (0.5 g/kg rat feed) for 21 consecutive days exhibited

significantly higher testes weights as compared to untreated controls. This, however, is an isolated report and can be investigated further to broaden our understanding regarding the effect of *A. racemosus* on the male reproductive system (20). A single study conducted on oligozoospermic men (n=100) administered a combination of *A. recemosus*, *W. somnifere* and *Tribulus terrestris* (4gm/day). In comparison to allopathic treatment (Testosterone undecanoate—40 mg t.i.d, Clomiphene citrate—25 mg per day, Tamoxifen Citrate) a significant improvement in sperm count, motility and morphology was reported (21).

A. racemosus has been reported to protect from oxidative stress induced by gamma-radiation in rat liver mitochondria *in vitro* (22) and also possesses aphrodisiac activity (23). The root extracts of *A. racemosus* were effective in providing protection against abdominal sepsis (24), intraperitoneal adhesions (25), ochratoxin A (26) and improving both humoral and cell mediated immunity (24), proving its adaptogenic properties. In *Ayurveda*, it has been described as absolutely safe for long term use, even during pregnancy and lactation. Both acute and chronic toxicity studies conducted in rat have established the safety of *A. racemosus* (27).

3.2. *Asteracantha longifolia* (Family: Acanthaceae, Sanskrit name: Kokilaksa)

Asteracantha longifolia Nees is a robust, erect, annual herb distributed throughout India. The roots, leaves and seeds have been used in Indian systems of medicine for management of various disorders. Seeds are used traditionally to treat sexual debility, premature ejaculation, erectile dysfunction and oligozoospermia (29).

Studies done in rat using ethanolic extract of seeds at 100, 150 and 200 mg/kg for a period of 28 days increased the rate of gain in body and reproductive organ weight with significant increase in sperm count as well as fructose level of seminal vesicle (28). The same study also reported a pronounced aphrodisiac effect. *A. longifolia* has hepatoprotective ability and potent *in vitro* antioxidant property (assessed by Ferric thiocynate [FTC] method and Thiobarbituric acid [TBA] method) (29). The latter may be the property helping improve fertility.

3.3. *Chlorophytum borivilianum* (Family: Liliaceae, Common name: Safed musli)

Chlorophytum borivilianum Santa Pau and Fernandes is a tiny annual herb that grows well in tropical and sub-tropical climates with altitudes up to 1500 meters and is distributed in northern and western India. This is a very popular herb in traditional Indian medicine and constitutes a group of herbs used as '*Rasayan*' or adaptogen. *C. borivilianum* has been described in ancient Indian literature as `*Vajikaran*' or aphrodisiac which is a special type of immunomodulator (30-32). Root tubers are used in Ayurvedic system as aphrodisiac and claimed to increase sperm count (33). The roots of the plant contain steroidal saponins, namely neotigogenin, neohecogenin, stigmasterol and tokorogenin, as secondary metabolites (33).

A study designed to evaluate aphrodisiac and spermatogenic potential of the aqueous extract of dried roots of this plant in rat at 125 and 250 mg/kg/day reported a marked increase in sperm count at day 60th in both the groups, in a dose dependent manner. The same dose also produced significant aphrodisiac action, increased libido, sexual vigor and sexual arousal after 14-28 days. Both the doses were equally effective with 250 mg/kg/day producing saturation effect after day 14 (34). A comparative study on aphrodisiac activity of this plant reported a significant variation in the sexual behavior of animals as reflected by reduction of mount latency, ejaculation latency, post ejaculatory latency, intromission latency, and an increase of mount frequency and penile erection in rat (35). Studies conducted in hyperlipidaemic rats, streptozotocin-induced diabetic animals and ulcer-models have confirmed potent antioxidant and adaptogenic properties of *C. borivilianum* (36-38).

3.4. *Clerodendrum serratum* (Family: Verbaneceae, Sanskrit Name: Bharangi)

Clerodendron serratum Linn known as '*Bharangi*' in *Ayurveda* and '*Sirutekku*' in *Siddha* system of medicine, is claimed to be useful in treating inflammatory disorders (39). It is a perennial shrub found throughout India up to 1500 meters height and abundantly in Eastern Himalayas, Nepal, Kumao, Bengal and Bihar. Root of this plant were claimed to be very effective in treatment of male infertility associated with chronic chest infection like pulmonary tuberculosis (29). However, there are no experimental studies supporting these claims.

3.5. *Dioscorea bulbifera* (Family: Dioscoreacea, Sanskrit Name: Varahikanda, Varahi)

Dioscorea bulbifera Linn is native to South Asia and it is cultivated to a limited extent in tropical and subtropical areas of East Africa to Polynesia. According to the *Ayurveda*, the tuber of *D. bulbifera* is considered bitter, pungent, tonic, alterative, aphrodisiac, stomachic and anthelmintic. Its activity has been associated with the presence of diosgenin, which is similar in structure to the adrenal hormone dehydroepiandrosterone (DHEA) (29). However there are no experimental studies to prove these claims. A single study designed to investigate the possible antioxidative effect of diosgenin on rat fed with a high-cholesterol diet showed that SOD in plasma and liver, glutathione peroxidase in erythrocytes, and catalase in erythrocytes and liver were significantly increased by diosgenin feeding (40).

3.6. *Morinda citrifolia* (Family: Ruiaceae, Sanskrit name: Ayushka)

Morinda citrifolia Linn is an evergreen tree or shrub that is distributed across India through Southeast Asia and Australia to Eastern Polynesia and Hawaii (41). The fruit juice is in high demand in alternative medicine for various ailments such as inflammatory conditions, infections, cancers and drug addictions; however, there are no scientific studies to prove these benefits

conclusively (41). Fruits, leaves and roots are used to increase the general and sexual strength in men i.e., to treat impotence, premature ejaculation and infertility (http://www.nutritionalwellness.com/nutrition/herbs/m/morinda_root.php, http://www.drshen.com/herbsforenergy.htm). The main areas of focus on research on *M. citrifolia* are in the field of cancer, imflammation and metabolic diseases (42).

An *in vivo* study conducted to investigate protective effects of *M. officinalis* (other species of the same genus) against oxidative injury in human sperm membrane reported that the extracts improved superoxide dismutase (SOD), vitality of sperm suspension, reduced the content of malondialdehyde (MDA), intervened in the injury of sperm membrane by ROS to some extent and protected some functions of sperm membrane. The extract also reduced lipid peroxidation in sperm membrane by guarding it against oxidation, thus protecting its structure and function (43).

Studies on human volunteers aimed at determining the effects of a poly-herbal formulation containing *M. citrifolia* fruit juice and seven other herbs on perceptions of stress showed it to be potential as an effective 'adaptogenic' aid in dealing with stress (44). Though the traditional literature suggests it to be useful for treatment of impotence, there are no scientific studies demonstrating its libido enhancing properties. *M. citrifolia* fruit juice has been approved as a safe food in many nations. *In vitro* hepatotoxicity tests conducted on human liver cells and a subchronic oral toxicity test performed in rat reported no histopathological changes or evidence of dose-responses in hematological and clinical chemistry measurements, including liver function tests. The no-observed-adverse-effect level (NOAEL) for freeze-dried noni fruit puree is greater than 6.86 g/kg body weight, equivalent to approximately 90 ml of fruit juice/kg. These findings establish *M. citrifolia* fruit juice to be absolutely safe for human consumption (45).

3.7. *Moringa oleifera* (Family: Moringaceae, Sanskrit name: Shobhanjana)

Moringa oleifera Lam (syn. *M. ptreygosperma Gaertn.*) is one of the best known and most widely distributed and naturalized species of a monogeneric family Moringaceae (39). *M. oleifera*, native of the western and sub-Himalayan tracts, India, Pakistan, Asia Minor, Africa and Arabia is now distributed in the Philippines, Cambodia, Central America, North and South America and the Caribbean Islands (46). The Moringa plant provides a rich and rare combination of zeatin, quercetin, β-sitosterol, caffeoylquinic acid and kaempferol. Leaves, root, seed, bark, fruit, flowers and immature pods are used extensively in management of different ailments in the indigenous system of medicine, particularly in South Asia (46). Flowers have been traditionally used to treat impotence; however there are no scientific studies to confirm these benefits (46). *M. oleifera* extracts have potent *in vitro* and *in vivo* antioxidant properties (47) (48).

3.8. *Mucuna pruriens* (Family: Fabaceae, Sanskrit name: Kapikachhu)

Mucuna pruriens Linn is a tropical legume cultivated in India, Sri Lanka, South East Asia and Malaysia (49). It is commonly used in the management of parkinsonism as it is rich in l-3,4 dihydroxyphenyl alanine (l-DOPA) (50). *M. pruriens* seed extract is also widely used to manage free radical-mediated diseases, such as rheumatoid arthritis, diabetes, atherosclerosis, neuronal disorders and male infertility (51) (52). *M. pruriens* contains wide range of medicinal and nutritional constituents like alkaloids, mucunine, mucunadine, mucunadinine, prurienidine and nicotine, besides *b*-sitosterol, glutathione, lecithin, vernolic acid, gallic acid and other bioactive substances like; tryptamine, alkylamines, steroids, flavonoids, coumarins, cardenolides and metals like magnesium, copper, zinc, manganese and iron (53) and oleic acid, linoleic acid and palmitic acid (54). These constituents make it an excellent combination of ingredients capable of healing several disorders.

Study conducted to evaluate protective efficacy of *M. pruriens* on reactive oxygen species (ROS)-induced pathophysiological alterations in structural and functional integrity of epididymal sperm in aged Wister albino rat (characterized by significant reduction in sperm count, viability and motility, increased morphological damage and an increase in the number of sperm with cytoplasmic remnant), reported reversal of age related changes in spermatozoa after administration of *M. pruriens* ethanolic seed extract (200mg/kg b.w). *M. pruriens* administration also significantly reduced ROS and LPO production and increased both enzymatic and non-enzymatic antioxidant levels. Spermatozoal DNA integrity, chromosomal integrity and mitochondrial membrane permeability were also restored after treatment. This study showed potent antioxidant property, free radical quenching ability and spermatogenic efficacy of *M. pruriens*. (55). Our previous studies conducted to investigate the impact of *Mucuna pruriens* seeds on infertile men have shown that treatment with *M. pruriens* significantly inhibited lipid peroxidation, elevated spermatogenesis, improved sperm motility, recovered the levels of total lipids, triglycerides, cholesterol, phospho-lipids, and vitaminA, C, and E, corrected fructose in seminal plasma of infertile men (13). The treatment with *M. pruriens* significantly improved T, LH, dopamine, adrenaline, and noradrenaline levels in infertile men and reduced levels of FSH and PRL in serum of infertile men (56).

Further investigation undertaken on infertile men with psychological stress showed treatment with *M. pruriens* significantly ameliorated psychological stress and reduced seminal plasma lipid peroxide levels along with improved sperm count and motility. This demonstrates that *M. pruriens* not only reactivates the anti-oxidant defense system of infertile men but it also helps in the management of stress and improves semen quality (14). Therefore both animal and human studies reported effects of *M. pruriens* on sperm count where as motility was only slightly improved. The fertility restoring ability of this plant was attributed to its potent anti-stress and adaptogenic

properties (14). Restorative and invigorating effects of its seeds were reported to increase semen volume and act as aphrodisiac in rat (57, 58). A study conducted in on rat showed that ethanolic extract of *M. pruriens* (200 mg/kg) was very effective in increasing mounting frequency, intromission frequency and ejaculation latency, and decreasing mounting latency, intromission latency, post-ejaculatory interval and inter-intromission interval. Further, the potency test significantly increased erections, quick flips, long flips and total reflex (58).

3.9. *Plumbago zeylanica* (Family: Plumbaginaceae, Sanskrit Name: Chitrak)

Plumbago zeylanica Linn is a useful Indian medicinal plant found wild in peninsular India and also cultivated as ornament. The root of the plant and its constituents are credited with potential therapeutic properties including anti-atherogenic, cardiotonic, hepatoprotective and neuroprotective. It is also used in traditional Chinese medicine as anti-inflammatory agent.

The plant has been claimed to improve fertility in men by regulating seminal fluid and prostatic secretions (29). However, there are no scientific studies supporting these claims. Traditional literature of Ayurveda has been prescribing *P. zeylanica* as aphrodisiac. Significant antioxidant properties have been reported for plumbagin (principle active ingredient) by ferric reducing/antioxidant power (FRAP), radical scavenging of 1,1-diphenyl-2-picryl hydrazyl (DPPH) and 2,2'-azobis-3-ethylbenzthiazoline-6-sulfonic acid (ABTS) tests in rat liver mitochondria induced by different agents (59). Seselin was demonstrated to be the principal immunomodulatory component of *P. zeylanica* (60).

3.10. *Semecarpus anacardium* (Family: Anacardiaceae, Sanskrit Name: Ballataka)

Semecarpus anacardium Linn is one of the best, versatile, most commonly used herbs as a household remedy in India since centuries. Varieties of medicinal properties have been confirmed by experiments on nuts of this herb (61). *S. anacardium* nut contains biflavanones including semecarpuflavanone, jeediflavanone, galluflavanone, nalluflavanone, semecarpetin and anacardiflavanone (62).

It has been mentioned as a rejuvenative and aphrodisiac, especially beneficial in the erectile dysfunction and sexual debility (29). However, there are no scientific studies supporting these claims. Study designed to investigate antioxidant activity of the aqueous extract of *S. anacardium* nut in AKR mouse liver during the development of lymphoma, reported an increase in the activities of antioxidant enzymes (63).

3.11. *Solanum surratense & Solanum Xanthocarpum* (Family: Solanceae, Sanskrit Name: Kantakari)

Solanum surratense and *Solanum Xanthocarpum* are found in Southeast Asia, Malaya and tropical Australia. The plant is very commonly found throughout India in plains from seashore to hills up to 1000 m high. Stem, flowers and fruits have been used for management of various ailments. These herbs are claimed to be useful in patients with idiopathic infertility (29); however, there are no scientific studies validating the fertility enhancing effects.

3.12. *Tribulus terrestris* (Family: Zygophilliceae, Sanskrit name: Gokshru)

Tribulus terrestris Linn is a flowering plant, native to warm temperate and tropical regions of the Old World in southern Europe, southern Asia, throughout Africa, and in northern Australia. This plant or its products have been extensively used both in the Chinese and Indian traditional medicine as a health tonic for management of several disorders (64, 65). *T. terrestris* is beneficial alone and in combination with Satavari and Ashwagandha in idiopathic male infertility (29).

Studies in rat reported higher weight gain and improvement in sexual behavior parameters upon administration of *T. terrestris* extract (2.5, 5 and 10 mg/kg body weight) (66). Similar study designed to investigate the influence of *T. terrestris* extract on androgen metabolism in young males reported that *T. terrestris* steroid saponins possess neither direct nor indirect androgen-increasing properties (67).The protodioscin gets converted to dehydroepiandrosterone (DHEA) inside the body, which enhances the sexual function in males (68). Study on diet-induced hyperlipidemic mice model has reported increase in the activities of superoxide-dismutase (SOD) in liver on administration of saponin from *T. terrestris* (69).

3.13. *Trichopus zeylanicus* (Family: Trichopodaceae, Common name: Ginseng of Kerala, Arogyapacha)

Trichopus zeylanicus Gaerten is an endemic herb found in the Southwestern mountain ranges of India, Sri Lanka and Malaysia. Fruits were consumed by South Indian tribes to rejuvenate from fatigue. The plant possesses many pharmacological activities like antihepatotoxic, antiulcer and antifatigue (70). *T. zeylanicus* contains NADH, polyphenols and sulfhydryl compounds, which have the ability to scavenge ROS, suggesting that the antioxidant activity may be an important mechanism of action of *T. zeylanicus* to combat fatigue (71). Leaf (ethanol extract) extract has proven aphrodisiac properties as evidenced by an increase in the number of mounts and mating performance in mice (72). A glyco-peptido lipid fraction ("AF") from the alcoholic extract of *T. zeylanicus* exhibited significant antistress activity in a dose dependent manner to induce non-specific stress (73) (74).

3.14. *Withania somnifera* (Family: Solanceae, Sanskrit name: Ashwagandha)

Withania somnifera Dunal is popularly known as Ashwagandha or Winter Cherry. It is a green shrub (75) found throughout the drier parts of India, Baluchistan, Pakistan, Afghanistan, Sri Lanka, Congo, South Africa, Egypt, Morocco and Jordan. In India, it is widely grown in the provinces of Madhya Pradesh, Uttar Pradesh, plains of Punjab and northwestern parts of India like Gujarat and Rajasthan (76). The practitioners of the traditional system of medicine in India regard *W. somnifera* as the “Indian

Ginseng" (77). Various parts of the plant have been used for centuries to treat variety of ailments (78).

Many pharmacological studies have reported anti-inflammatory (79), anticancer (80, 81), antistress, immunomodulatory (82-84), adaptogenic (85), central nervous system rejuvenating (86), endocrine (87) and cardiovascular (88) activities of this plant. Its effects on central nervous system mainly operate through the modulation of GABAergic [(gamma-amino-butyric acid (GABA)] (89) and cholinergic (90) neurotransmission. The major biochemical constituents of *W. somnifera* are steroidal alkaloids and lactones, a class of constituents together known as withanolides (steroidal lactones with ergostane skeleton) (91). So far 12 alkaloids, 35 withanoloids and several sitoindosides have been isolated and their structures elucidated (88). Various alkaloids include withanine, somniferine, somnine, somniferinine, withananine, psuedo-withanine, tropine, psuedotropine, 3-α-gloyloxytropane, choline, cuscohygrine, isopelletierine, anaferine and anahydrine. Two acyl steryl glucoside viz. sitoindoside VII and sitoindoside VIII, two glycowithanoloids viz. sitoindoside IX and sitoindoside X have been isolated from the root. Withanolides structurally resemble ginsenosides found in *Panax ginseng*; hence *W. somnifera* is regarded as the Indian Ginseng (92).

Study on 20 days old immature male Wistar rat administered *W. somnifera* root extract reported increase in testicular weight, diameter of seminiferous tubules (ST), seminiferous epithelial cell layers (CL) and serum levels of Interstitial Cell Stimulating Hormone (ICSH) with concurrent reduction in serum Testosterone and FSH levels (93). Human trials were conducted in India to asses the benefits of *W. somnifera* root in the management of idiopathic male infertility (17). In this study 75 men undergoing infertility screening were prescribed *W. somnifera* root powder. The treatment reduced lipid peroxidation and protein carbonyl content and improved sperm count and motility. Treatment also restored the seminal plasma levels of antioxidant enzymes and vitamins A, C, and E and corrected fructose. Moreover, treatment also significantly improved serum T and LH and reduced the levels of FSH and PRL.

W. somnifera has been described in folk medicine as an aphrodisiac and geriatric tonic (94). *W. somnifera* root extract has shown beneficial effects in reducing lipid peroxidation (95) and increasing the superoxide dismutase (SOD) and catalase activity (96) in rabbit and mice models. Withania has strong adaptogenic activity as proven by various animal model studies (85, 92, 95, 84, 97, 98, 74 and 99). Both acute and chronic toxicity profile studies have reported no adverse effects of *W. somnifera* (76). Hence the traditional name "Indian Ginseng" cannot be an exaggeration.

3.15. Herbal Formulations

Each herb discussed above has some unique medicinal property or active ingredient. Based on these facts, combinations of several herbs are being used traditionally in Ayurvedic system as well as newer formulations are available in market designed based on scientific evidence. The Ayurvedic system of medicine has prescribed a formulation (Rasayana) consisting of 8 herbs in specific concentrations for treatment of male infertility. The recipe consists of *Withania somnifera* (roots) - 15%, *Mucuna pruriens* (seeds) - 25%, *Tribulus terrestris* (fruits) - 20%, *Glycyrrhiza glabra* (roots) - 10%, *Terminalia arjuna* (bark) - 10%, *Phyllanthus emblica* (fruits) - 10%, *Zingiber officinale* (roots) - 5% and *Piper longum* (fruits) - 5% (100).

Another commonly used herbal formulation (Speman®) for male infertility treatment is manufactured by Himalaya herbal healthcare, Bangalore, India. Speman® consists of *Orchis mascula* - 25%, *Asteracantha longifolia* – 12%, *Lactuca scariola* - 6%, *Mucuna pruriens* - 6%, Mosaic gold - 6%, *Argyreia speciosa* - 12%, *Tribulus terrestris* - 12%, *Leptadenia reticulate* - 12%, *Parmelia perlata* - 6%. (http://www.himalayahealthcare.com/products/speman.htm). Speman® has been tested widely in human patients and has been found to be very effective in the managemanet of idiopathic infertility (101, 102) (find list of all the publications at http://www.himalayahealthcare.com/researchpaper/speman.htm).

These two formulations are essentially mixtures of antioxidants, anti-stress, adaptogenic and immunomodulatory principles which act together to reinvigorate the reproductive system and stimulate the spermatogenesis and glandular secretions (100).

4. DISCUSSION

The floral diversity in India is very high due to availability of different soil types, varying altitudes and climatic conditions. The country notes very high rainfall in the North-East region, endures dry conditions in the West, exhibits severe cold to severe hot seasons in upper north and north region and engulfs south with temperate weather with high rainfall and humidity. The varied climatic conditions are the reason behind the high number of medicinal plant varieties (approximately 20,000 species) found in India. Apart from highly manipulated Gangatic plains, there is naturally preserved large 'Himalayan' stretch. The long term exploration of the natural resources for their medicinal properties has given India an edge in natural medicine over several other countries. The same is evident by the fact that traditional Indian systems of herbal medicines are one of the oldest in the world. Major medicinal systems such as *'Ayurveda'* are based on the philosophy of rational enquiry into the nature of the truth. The principle basis for these traditional medicines dates back from 1000 BC (Charak Samhita) to 100 AD (Sushrut Samhita) and the Materia medica give detailed description of over 1500 herbs and 10,000 formulations. Further, till very recent, modern medicinal practices were uncommon in India and people used to believe and practice natural therapies. The common man in rural areas still practice natural therapies using traditional knowledge. The best example of the same are the tribal populations spread all

Table 1. Promising medicinal plants and their properties in management of male infertility

Plant	Part(s) used	Activity			Experimental species	Reference
		Antioxidant	Aphrodisiac	Adaptogenic		
Asparagus recemosus	Root	✓	✓	✓	Human, rat	19, 20-26
Asteracantha longifolia	Seeds	✓	✓	✓	Rat	27, 28
Chlorophytum borivilianum	Roots	✓	✓	✓	Rat	33-37
Clerodendrum serratum	Roots	NA	NA	NA	NA	-
Dioscorea bulbifera	Tubers	✓	NA	NA	Rat	39
Morinda Citrifolia	Fruits	✓	NA	✓	Human	42, 43
Moringa oleifera	Flowers	✓	NA	NA	Rat	46, 47
Mucuna pruriens	Seeds	✓	✓	✓	Human, Rat	13, 14, 54-57
Plumbago zeylanica	Roots	✓	NA	NA	Rat	58, 59
Semecarpus anacardium	Nuts	✓	NA	NA	Mouse	62
Solanum surratense & *Solanum Xanthocarpum*	Stem, flowers and fruits	NA	NA	NA	NA	-
Tribulus terrestris	Whole plant	✓	✓	NA	Human, Rat, Mouse	29, 68
Trichopus zeylanicus	Leaves	NA	✓	✓	Mice	70-73
Withania somnifera	Roots	✓	✓	✓	Human, Rat	16, 93, 95, 91, 94, 96, 97

across the country who do not believe in modern medicinal system and use home-made herbal formulations to treat all their ailments, many of which are even in clinical practice by the name of '*Ayurvedic*' or '*desi*' medicines.

The 14 Indian medicinal herbs mentioned above are in use for management of male infertility since time immemorial in various parts of India, based on their distribution and availability (100). The reported success of herbal medicines is largely based on opinions and anectodes. As mentioned above, a significant number of these plants have been tested on animal models (Table 1). Since there is no good experimental infertility animal model to study fertility enhancing effects, *in vivo* tests are conducted either on normal or fertility compromised animals (using endocrine disruptors such as estrogen). However, nothing mimics the condition of infertility as in human patients. Therefore, human trials are necessary before we can accept the claimed benefits of these herbs. Only few of these have been tested in human subjects (13, 17, 103), of which, two (*Mucuna pruriens* and *Withania somnifera*) were tested by our group and collaborators (13, 17). There is a long list of herbs awaiting evaluation in human trials.

Almost half of the infertile males presenting with no detectable underlying pathology may have oxidative stress as the underlying cause (16). ROS is largely produced by macrophages and neutrophils, but spermatozoa can also generate large amounts in pathological conditions (104). Oxidative stress results from an imbalance between production of ROS and its efficient removal by the antioxidant system. Oxidative stress can damage all membrane lipids, cellular proteins and macromolecules including DNA and RNA (105). Testis, due to its very high metabolic rate, stands prone to damage by oxidative stress. The major antioxidant enzyme system in testis is superoxide dismutase, catalase and glutathione peroxidase whereas non-enzymatic antioxidant defense is constituted by Vitamin C, Vitamin E and melatonin (106). It has already been proven that these antioxidants and resveratrol (a botanical antioxidant) are very effective in reducing testicular oxidative stress (107-110).

Environmental toxicants like pesticides, industrial pollutants, xenobiotics (estrogenic) and high concentration of certain metals (iron, lead, cadmium etc) severely compromise male fertility by increasing oxidative stress (111). Lifestyle factors such as excessive alcohol consumption or cigarette smoking increases free radical production in testis and compromise fertility (112, 113). Most of the common causes of male infertility like vericocele, cryptorchidism, ageing and testicular torsion are associated with elevated ROS levels (111). Chemotherapy, ionizing radiations and localized/systemic inflammations lead to transient or permanent damage to testis due to high ROS generation (114, 115). Research focused on change of sperm quality in men exposed to stress like war (116) and earthquake (117) has reported significant reduction in sperm count, which may impact the marital life of the couple. Amongst several factors affecting male fertility in emotional stress conditions, oxidative stress was also reported to be important (118). All the above emphasizes the role of oxidative stress in male infertility.

Most of the plants discussed in this review are good source of antioxidants (Table 1). It is possible that pro-fertility activities of these are due to their ability to alleviate oxidative stress. However, only few studies have monitored the antioxidant/oxidant levels with treatment. We have previously shown that infertility in a large number of patients could be attributed to oxidative stress and that *M. pruriens* and *W. somnifera* are very effective in improving male fertility due to their ability to combat oxidative stress (13, 14 and 16). Most of the animal studies till date have focused on the effects of plants on sperm count and motility and have not explored the concept of oxidative stress as a cause and antioxidant therapy (of plant material) as the treatment. Based on our observations and well known anti-oxidant properties of several plants, we recommend the analysis of oxidative stress parameters in all the studies evaluating plant products in infertility management.

Another treatable impairment which may lead to infertility is erectile dysfunction (ED). ED is the inability to achieve or maintain erection sufficient enough for sexual intercourse (119). Normal erectile function depends on

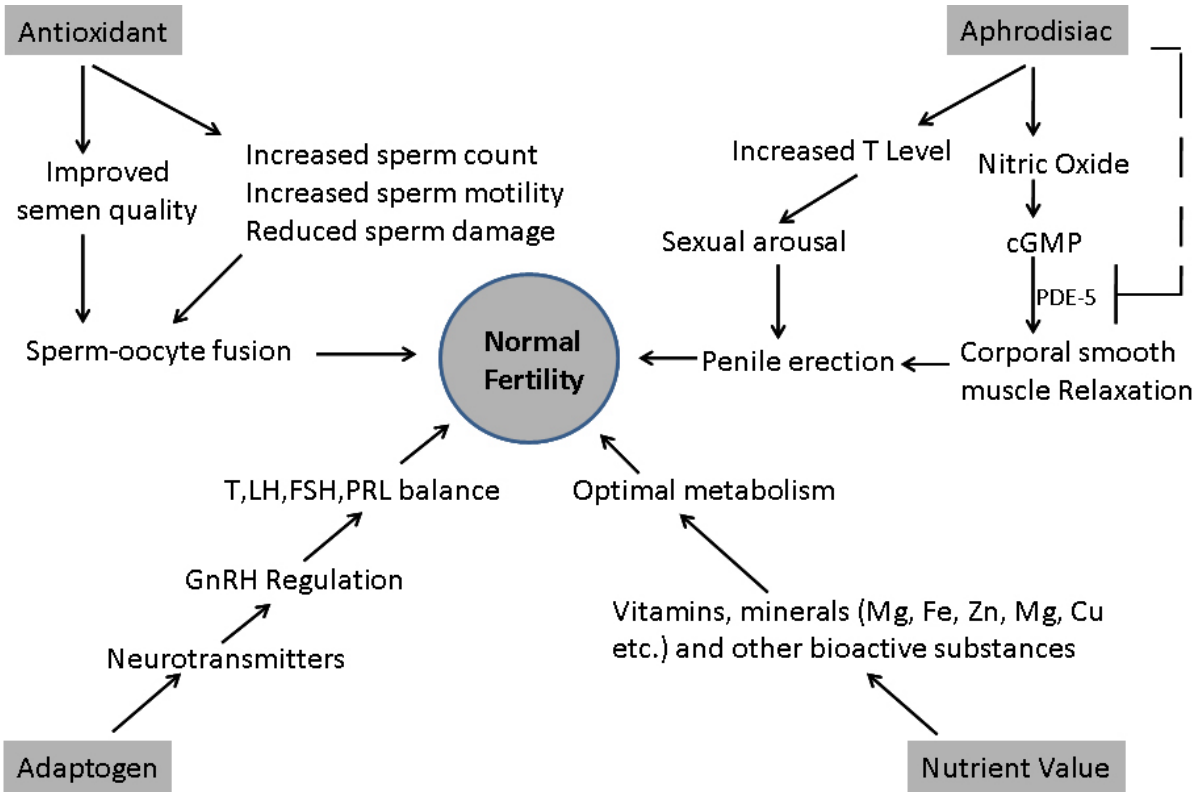

Figure 1. Possible mechanism of action of promising medicinal plants for male infertility treatment

psychological, hormonal, neurological, vascular and cavernosal factors and alteration of their precise balance may lead to ED (120). Recently, oxidative stress has been repeatedly implicated as one of underlying factors for ED (120). Though there is no concrete evidence of this association; reports from *in vitro* and *in vivo* experiments have shown ROS to be one of the major causes of ED (120). Population studies have shown diabetes, hypertension and cigarette smoking, which grossly elevate the ROS levels, as risk factors for ED (121, 122). Since most of the studies till date have been carried out on rat/mice models, the effect of herbal treatments on ED is largely unclear. However, aphrodisiac activity in many studies has been reported, which may be extrapolated to state that herbal treatments can be effective in the treatment of ED. The later effect may be the mechanism of action in several cases or may supplement antioxidant effects to help recover from infertility. Our studies on human subjects also did not investigate ED parameters, since individual criterion to define normal erection varies and the information from patients was thought to be unreliable. Nevertheless, the improvement in testosterone levels in most of the patients could be thought to cure ED, if any. Studies in future should investigate if reduction in ED is one of the important mediators of pro-fertility activity of these herbs.

The use of hormonal therapies, which is presently the most researched and commonly practiced treatment option for management of male infertility, has very low success rate as suggested by our analysis of the published literature (5). The use of complimentary medicine (both single and multiple preparations) with multiple benefits is definitely a better option compared to targeted therapies in the treatment of idiopathic male infertility. There are several reasons why we should think of and explore herbal medicines against hormonal therapies or other modern methods in male infertility treatment. First, most of the hormone based/other modern drugs have one or few targets and the outcome of treatment would depend if these targets were the underlying cause. Herbal medicines, on the other hand, act on multiple targets, thereby increasing the likelihood of success. Second, the herbal medications also contain many important trace nutrients in addition to known active compounds, which make them excellent combination in single form. Third, herbal treatments are very less likely to cause side effects, if any however, modern medicines including hormonal therapies are well known for their adverse side effects. Hormonal therapies often lead to severe consequences like endocrine cancers and tissue over-growth.

Several medicinal plants, traditionally used for thousands of years, are present in a group of herbal preparations of the Indian traditional health care system named Rasayana. These plants are interesting because of their antioxidant activities, and some of these namely *Emblica officinalis*, *Curcuma longa*, *Mangifera indica*, *Momordica charantia*, *Santalum album*, *Swertia chirata*, and *Withania somnifera* have been thoroughly investigated for their antioxidant properties. These plants contain

medicinal properties like resistance building, immunomodulation, cardioprotective, neuroprotective, hepatoprotective and are helpful in management of geriatric and chronic disorders. These effects are mediated by reduction of oxidative stress (123). Given significant correlation of oxidative stress with male infertility, many more plants than listed in this review deserve screening for their fertility enhancing properties. We have established cause and effect relationship between oxidative stress and male subfertility in our previous studies done by measuring oxidative stress biomarker levels in normozoospermic (n=40), oligozoospermic (n=40) and asthenozoospermic (n=40) individuals (13, 17). The relationship was further confirmed by alleviation of oxidative stress and improvement of fertility on prescription of natural antioxidants, *Withania somnifera* and *Mucuna pruriens*. The time is now ripe to investigate the importance of each constituent of these Rasayanas and work out to design superior formulations based on experimental evidence.

Similarly, the adaptogenic properties of *Panax ginseng, Eleutherococcus senticosis, Glycyrrhiza glabra, Withania somnifera, Ocimum sanctum, Rhodiola rosea, Emblica officinalis, Tinospora cordifolia* etc, have been established as the basis for their antifatigue, antiinflammatory, antioxidant, anxiolytic, antidepressant, nervine, and amphoteric effects, which are utilized in the treatment of numerous disorders (87, 99) (Table 1). These medicinal herbs need to be evaluated extensively in animal models and if found promising, can be tested in human trials. The traditional medicinal preparations are complex mix of several active ingredients; hence the effects may be synergistic results of these principles ultimately leading to overall desired response (Figure 1, Table 1). The identification of effects of individual components in these preparations is arduos task. To enter the mainstream western medicinal practice, the active ingredients of these preparations need to be identified, but that will be equal to impossible. Unbiased evaluation in the natural form to establish safety and dose of these preparations, rather than spending time and capital in identifying active ingredients should be the way forward to exploit the tremendous promise these plants bear. However, identification of active ingredients may be warranted to avoid pressure on natural resources for medcinal purposes.

5. CONCLUSION

Research in past years has established the claimed benefits of some of these herbs in management of male infertility. Since most of these plants have been part of human diet in some or other form since ages, side effects of these, if any, might have lead to general disapproval and discontinuation of their use. However, studies investigating safety aspects should be encouraged to expose unknown short or long-term effects. Once, these parameters are established along with suitable dose, commercial use can be achieved even if the active ingredients are unknown. As mentioned in our studies above, some of the herbs like *Mucuna pruriens* and *Withania somnifera* have been very effective in treatment of idiopathic male infertility in human patients. Since these trials were conducted mainly on the basis of traditional knowledge; even without extensive toxicological testing, other plant preparations can be evaluated similarly. As seen in this review, not very extensive literature exists on medicinal values of plants especially for management of reproductive disorders. With this compilation we want to encourage the exploration of several wonder drug plants for their medicinal uses, which could either be used as such or could lay foundation to the identification of many important drug molecules.

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7. REFERENCES

1. B. Patwardhan, D. Warude, P. Pushpangadan and N. Bhatt: Ayurveda and traditional Chinese medicine: a comparative overview. *Evid Based Complement Alternat Med*, 2(4), 465-73 (2005)

2. The promotion and development of traditional medicine: report of a WHO meeting. *World Health Organ Tech Rep Ser*(622), 1-41 (1978)

3. S. Bhasin, D. M. de Kretser and H. W. Baker: Clinical review 64: Pathophysiology and natural history of male infertility. *J Clin Endocrinol Metab*, 79(6), 1525-9 (1994)

4. J. Auger, J. M. Kunstmann, F. Czyglik and P. Jouannet: Decline in semen quality among fertile men in Paris during the past 20 years. *N Engl J Med*, 332(5), 281-5 (1995)

5. D. Madhukar and S. Rajender: Hormonal treatment of male infertility: promises and pitfalls. *J Androl*, 30(2), 95-112 (2009)

6. D. S. Guzick, J. W. Overstreet, P. Factor-Litvak, C. K. Brazil, S. T. Nakajima, C. Coutifaris, S. A. Carson, P. Cisneros, M. P. Steinkampf, J. A. Hill, D. Xu and D. L. Vogel: Sperm morphology, motility, and concentration in fertile and infertile men. *N Engl J Med*, 345(19), 1388-93 (2001)

7. A. Ferlin, F. Raicu, V. Gatta, D. Zuccarello, G. Palka and C. Foresta: Male infertility: role of genetic background. *Reprod Biomed Online*, 14(6), 734-45 (2007)

8. P. H. Vogt: Molecular genetics of human male infertility: from genes to new therapeutic perspectives. *Curr Pharm Des*, 10(5), 471-500 (2004)

9. J. D. Meeker, L. Godfrey-Bailey and R. Hauser: Relationships between serum hormone levels and semen quality among men from an infertility clinic. *J Androl*, 28(3), 397-406 (2007)

10. A. Agarwal and R. A. Saleh: Role of oxidants in male infertility: rationale, significance, and treatment. *Urol Clin North Am*, 29(4), 817-27 (2002)

11. F. F. Pasqualotto, R. K. Sharma, D. R. Nelson, A. J. Thomas and A. Agarwal: Relationship between oxidative stress, semen characteristics, and clinical diagnosis in men undergoing infertility investigation. *Fertil Steril*, 73(3), 459-64 (2000)

12. A. Agarwal, K. P. Nallella, S. S. Allamaneni and T. M. Said: Role of antioxidants in treatment of male infertility: an overview of the literature. *Reprod Biomed Online*, 8(6), 616-27 (2004)

13. M. K. Ahmad, A. A. Mahdi, K. K. Shukla, N. Islam, S. P. Jaiswar and S. Ahmad: Effect of Mucuna pruriens on semen profile and biochemical parameters in seminal plasma of infertile men. *Fertil Steril*, 90(3), 627-35 (2008)

14. K. K. Shukla, A. A. Mahdi, M. K. Ahmad, S. P. Jaiswar, S. N. Shankwar and S. C. Tiwari: Mucuna pruriens Reduces Stress and Improves the Quality of Semen in Infertile Men. *Evid Based Complement Alternat Med* (2007)

15. K. Makker, A. Agarwal and R. Sharma: Oxidative stress & male infertility. *Indian J Med Res*, 129(4), 357-67 (2009)

16. K. Tremellen: Oxidative stress and male infertility--a clinical perspective. *Hum Reprod Update*, 14(3), 243-58 (2008)

17. M. K. Ahmad, A. A. Mahdi, K. K. Shukla, N. Islam, S. Rajender, D. Madhukar, S. N. Shankhwar and S. Ahmad: Withania somnifera improves semen quality by regulating reproductive hormone levels and oxidative stress in seminal plasma of infertile males. *Fertil Steril* (2009)

18. Anonymous, 1987 Anonymous, The Wealth of India, Raw materials, Publication and Information Directorate, CSIR, N. Delhi (1987) pp. 468–472.

19. N. Bopana and S. Saxena: Asparagus racemosus--ethnopharmacological evaluation and conservation needs. *J Ethnopharmacol*, 110(1), 1-15 (2007)

20. Ghumare, V.P. Vadlamudi and S.R. Rajurkar, Effect of Asparagus racemosus on growth and development of testes in wistar rats, *Aryavaidyan* 18, 45–48 (2004)

21. R. D. Papolu, Vijaya Laxmi, C. Chatterjee, and R. Adusumilli: ‘‘Alternative medicine’’—a right choice for male infertility management. *International Congress Series* 1271, 67– 70 (2004)

22. J. P. Kamat, K. K. Boloor, T. P. Devasagayam and S. R. Venkatachalam: Antioxidant properties of Asparagus racemosus against damage induced by gamma-radiation in rat liver mitochondria. *J Ethnopharmacol*, 71(3), 425-35 (2000)

23. P. C. Sharma, M. B. Yelne, T. J. Dennis: Database on Medicinal Plants Used in Ayurveda, Volume I. *Yugantar Prakashan,* New Delhi (2000)

24. R. K. Goyal, J. Singh and H. Lal: Asparagus racemosus--an update. *Indian J Med Sci*, 57(9), 408-14 (2003)

25. N. N. Rege, H. M. Nazareth, A. Isaac, S. M. Karandikar and S. A. Dahanukar: Immunotherapeutic modulation of intraperitoneal adhesions by Asparagus racemosus. *J Postgrad Med*, 35(4), 199-203 (1989)

26. J. N. Dhuley: Effect of some Indian herbs on macrophage functions in ochratoxin A treated mice. *J Ethnopharmacol*, 58(1), 15-20 (1997)

27. A. V. Muruganandam, V. Kumar and S. K. Bhattacharya: Effect of poly herbal formulation, EuMil, on chronic stress-induced homeostatic perturbations in rats. *Indian J Exp Biol*, 40(10), 1151-60 (2002)

28. N. S. Chauhan, V. Sharma and V. K. Dixit: Effect of Asteracantha longifolia seeds on the sexual behaviour of male rats. *Nat Prod Res*, 1-9 (2009)

29. P. Shanmugasundaram and S. Venkataraman: Hepatoprotective and antioxidant effects of Hygrophila auriculata (K. Schum) Heine Acanthaceae root extract. *J Ethnopharmacol*, 104(1-2), 124-8 (2006)

30. A. Triveni: Rasendrasarasangrah: Vajikaranadhikar. *Nutan Press*, Rajkot, India (1976)

31. K. R. Kirtikar, B. D. Basu: Indian Medicinal Plants. *International Book Distributors*, Dehradun India (1995)

32. M. Thakur, S. Bhargava and V. K. Dixit: Immunomodulatory Activity of Chlorophytum borivilianum Sant. F. *Evid Based Complement Alternat Med*, 4(4), 419-23 (2007)

33. G. S. Thakur, M. Bag, B. S. Sanodiya, M. Debnath, A. Zacharia, P. Bhadauriya, G. B. Prasad and P. S. Bisen: Chlorophytum borivilianum: A White Gold for Biopharmaceuticals and Neutraceuticals. *Curr Pharm Biotechnol*, 10(7), 650-666 (2009)

34. R. Kenjale, R. Shah and S. Sathaye: Effects of Chlorophytum borivilianum on sexual behaviour and sperm count in male rats. *Phytother Res*, 22(6), 796-801 (2008)

35. M. Thakur, N. S. Chauhan, S. Bhargava and V. K. Dixit: A Comparative Study on Aphrodisiac Activity of Some Ayurvedic Herbs in Male Albino Rats. *Arch Sex Behav* (2009)

36. N. P. Visavadiya and A. V. Narasimhacharya: Ameliorative effect of Chlorophytum borivilianum root on lipid metabolism in hyperlipaemic rats. *Clin Exp Pharmacol Physiol*, 34(3), 244-9 (2007)

37. S. Narasimhan, R. Govindarajan, V. Madhavan, M. Thakur, V. K. Dixit, S. Mehrotra and K. P. Madhusudanan: Action of (2-->1)Fructo-oligopolysaccharide fraction of Chlorophytum borivilianum against Streptozotocin-Induced oxidative stress. *Planta Med*, 72(15), 1421-4 (2006)

38. R. D. Kenjale, R. K. Shah and S. S. Sathaye: Anti-stress and anti-oxidant effects of roots of Chlorophytum borivilianum (Santa Pau & Fernandes). *Indian J Exp Biol*, 45(11), 974-9 (2007)

39. K. M. Nadkarni: Indian Materia Medica. *Popular Prakashan*, Bombay (1954)

40. I. S. Son, J. H. Kim, H. Y. Sohn, K. H. Son, J. S. Kim and C. S. Kwon: Antioxidative and hypolipidemic effects of diosgenin, a steroidal saponin of yam (Dioscorea spp.), on high-cholesterol fed rats. *Biosci Biotechnol Biochem*, 71(12), 3063-71 (2007)

41. M. Y. Wang, B. J. West, C. J. Jensen, D. Nowicki, C. Su, A. K. Palu and G. Anderson: Morinda citrifolia (Noni): a literature review and recent advances in Noni research. *Acta Pharmacol Sin*, 23(12), 1127-41 (2002)

42. O. Potterat and M. Hamburger: Morinda citrifolia (Noni) fruit--phytochemistry, pharmacology, safety. *Planta Med*, 73(3), 191-9 (2007)

43. X. Yang, Y. H. Zhang, C. F. Ding, Z. Z. Yan and J. Du: [Extract from Morindae officinalis against oxidative injury of function to human sperm membrane]. *Zhongguo Zhong Yao Za Zhi*, 31(19), 1614-7 (2006)

44. D. Seely and R. Singh: Adaptogenic potential of a polyherbal natural health product: report on a longitudinal clinical trial. *Evid Based Complement Alternat Med*, 4(3), 375-80 (2007)

45. J. W. Brett, X. S. Chen and C. J. Jensen: Hepatotoxicity and subchronic toxicity tests of Morinda citrifolia (noni) fruit. *J Toxicol Sci*, 34(5), 581-5 (2009)

46. F. Anwar, S. Latif, M. Ashraf and A. H. Gilani: Moringa oleifera: a food plant with multiple medicinal uses. *Phytother Res*, 21(1), 17-25 (2007)

47. M. Nandave, S. K. Ojha, S. Joshi, S. Kumari and D. S. Arya: Moringa oleifera leaf extract prevents isoproterenol-induced myocardial damage in rats: evidence for an antioxidant, antiperoxidative, and cardioprotective intervention. *J Med Food*, 12(1), 47-55 (2009)

48. B. N. Singh, B. R. Singh, R. L. Singh, D. Prakash, R. Dhakarey, G. Upadhyay and H. B. Singh: Oxidative DNA damage protective activity, antioxidant and anti-quorum sensing potentials of Moringa oleifera. *Food Chem Toxicol*, 47(6), 1109-16 (2009)

49. C. P. Kharelep: Encyclopedia of Indian Medicinal Plants (Regional Western Therapy Ayurvedic and Other Traditional Usage, Botany). *Springer-verlag, Berlin, Heidelberg, New York* (2004)

50. S. A. Molloy, E. N. Rowan, J. T. O'Brien, I. G. McKeith, K. Wesnes and D. J. Burn: Effect of levodopa on cognitive function in Parkinson's disease with and without dementia and dementia with Lewy bodies. *J Neurol Neurosurg Psychiatry*, 77(12), 1323-8 (2006)

51. Y. Rajeshwar, G. P. S. Kumar, M. Gupta and U. K. Mazumder: Studies on *in vitro* antioxidant activities of methanol extract of Mucuna pruriens (Fabaceae) seeds. *European Bulletin of Drug Research,* 13, 31–39 (2005)

52. M. Dhanasekaran, B. Tharakan and B. V. Manyam: Antiparkinson drug--Mucuna pruriens shows antioxidant and metal chelating activity. *Phytother Res*, 22(1), 6-11 (2008)

53. L. Misra and H. Wagner: Extraction of bioactive principles from Mucuna pruriens seeds. *Indian J Biochem Biophys*, 44(1), 56-60 (2007)

54. Y. A. Adebowale, A. Adeyemi and A. A. Oshodi: Variability in the physicochemical, nutritional and antinutritional attributes of six Mucuna species. *Journal of Food Chemistry,* 89, 37–48 (2005)

55. S. Suresh, E. Prithiviraj and S. Prakash: Effect of Mucuna pruriens on oxidative stress mediated damage in aged rat sperm. *Int J Androl* (2009)

56. K. K. Shukla, A. A. Mahdi, M. K. Ahmad, S. N. Shankhwar, S. Rajender and S. P. Jaiswar: Mucuna pruriens improves male fertility by its action on the hypothalamus-pituitary-gonadal axis. *Fertil Steril* (2008)

57. K. V. A. Kumar, K. K. Srinivasan, T. Shanbhag and S. G. Rao: Aphrodisiac activity of the seeds of Mucuna pruriens. *Indian Drug,* 31, 321–327 (1994)

58. S. Suresh, E. Prithiviraj and S. Prakash: Dose- and time-dependent effects of ethanolic extract of Mucuna pruriens Linn. seed on sexual behaviour of normal male rats. *J Ethnopharmacol*, 122(3), 497-501 (2009)

59. J. C. Tilak, S. Adhikari and T. P. Devasagayam: Antioxidant properties of Plumbago zeylanica, an Indian medicinal plant and its active ingredient, plumbagin. *Redox Rep*, 9(4), 219-27 (2004)

60. W. J. Tsai, Y. C. Chen, M. H. Wu, L. C. Lin, K. A. Chuang, S. C. Chang and Y. C. Kuo: Seselin from Plumbago zeylanica inhibits phytohemagglutinin (PHA)-stimulated cell proliferation in human peripheral blood mononuclear cells. *J Ethnopharmacol*, 119(1), 67-73 (2008)

61. R. Mythilypriya, P. S. Sachdanandam and P. Sachdanandam: Ameliorating effect of Kalpaamruthaa, a Siddha preparation in adjuvant induced arthritis in rats with reference to changes in proinflammatory cytokines and

acute phase proteins. *Chem Biol Interact*, 179(2-3), 335-43 (2009)

62. B. Premalatha: Semecarpus anacardium Linn. nuts--a boon in alternative medicine. *Indian J Exp Biol*, 38(12), 1177-82 (2000)

63. N. Verma and M. Vinayak: Semecarpus anacardium nut extract promotes antioxidant defense system and inhibits anaerobic metabolism during development of lymphoma. *Biosci Rep* (2008)

64. R. Anand, G. K. Patnaik, D. K. Kulshreshtha and B. N. Dhawan: Activity of certain fractions of Tribulus terrestris fruits against experimentally induced urolithiasis in rats. *Indian J Exp Biol*, 32(8), 548-52 (1994)

65. B. Wang, L. Ma and T. Liu: [406 cases of angina pectoris in coronary heart disease treated with saponin of Tribulus terrestris]. *Zhong Xi Yi Jie He Za Zhi*, 10(2), 85-7, 68 (1990)

66. K. Gauthaman, A. P. Ganesan and R. N. Prasad: Sexual effects of puncturevine (Tribulus terrestris) extract (protodioscin): an evaluation using a rat model. *J Altern Complement Med*, 9(2), 257-65 (2003)

67. V. K. Neychev and V. I. Mitev: The aphrodisiac herb Tribulus terrestris does not influence the androgen production in young men. *J Ethnopharmacol*, 101(1-3), 319-23 (2005)

68. A. Adimoelja: Phytochemicals and the breakthrough of traditional herbs in the management of sexual dysfunctions. *Int J Androl*, 23 Suppl 2, 82-4 (2000)

69. S. Chu, W. Qu, X. Pang, B. Sun and X. Huang: [Effect of saponin from Tribulus terrestris on hyperlipidemia]. *Zhong Yao Cai*, 26(5), 341-4 (2003)

70. B. Tharakan and B. V. Manyam: Botanical therapies in sexual dysfunction. *Phytother Res*, 19(6), 457-63 (2005)

71. B. Tharakan, M. Dhanasekaran and B. V. Manyam: Antioxidant and DNA protecting properties of anti-fatigue herb Trichopus zeylanicus. *Phytother Res*, 19(8), 669-73 (2005)

72. A. Subramoniam, V. Madhavachandran, S. Rajasekharan and P. Pushpangadan: Aphrodisiac property of Trichopus zeylanicus extract in male mice. *J Ethnopharmacol*, 57(1), 21-7 (1997)

73. B. Singh, D. K. Gupta and B. K. Chandan: Adaptogenic activity of a glyco-peptido-lipid fraction from the alcoholic extract of Trichopus zeylanicus Gaertn. *Phytomedicine*, 8(4), 283-91 (2001)

74. A. Singh, E. Saxena and K. K. Bhutani: Adrenocorticosterone alterations in male, albino mice treated with Trichopus zeylanicus, Withania somnifera and Panax ginseng preparations. *Phytother Res*, 14(2), 122-5 (2000)

75. A. Dafni and Z. Yaniv: Solanaceae as medicinal plants in Israel. *J Ethnopharmacol*, 44(1), 11-8 (1994)

76. S. K. Kulkarni and A. Dhir: Withania somnifera: an Indian ginseng. *Prog Neuropsychopharmacol Biol Psychiatry*, 32(5), 1093-105 (2008)

77. B. Singh, A. K. Saxena, B. K. Chandan, D. K. Gupta, K. K. Bhutani and K. K. Anand: Adaptogenic activity of a novel, withanolide-free aqueous fraction from the roots of Withania somnifera Dun. *Phytother Res*, 15(4), 311-8 (2001)

78. A. Bhattacharya, S. Ghosal and S. K. Bhattacharya: Anti-oxidant effect of Withania somnifera glycowithanolides in chronic footshock stress-induced perturbations of oxidative free radical scavenging enzymes and lipid peroxidation in rat frontal cortex and striatum. *J Ethnopharmacol*, 74(1), 1-6 (2001)

79. S. K. Bhattacharya, K. S. Satyan and A. Chakrabarti: Effect of Trasina, an Ayurvedic herbal formulation, on pancreatic islet superoxide dismutase activity in hyperglycaemic rats. *Indian J Exp Biol*, 35(3), 297-9 (1997)

80. P. U. Devi, A. C. Sharada, F. E. Solomon and M. S. Kamath: *In vivo* growth inhibitory effect of Withania somnifera (Ashwagandha) on a transplantable mouse tumor, Sarcoma 180. *Indian J Exp Biol*, 30(3), 169-72 (1992)

81. R. Mohan, H. J. Hammers, P. Bargagna-Mohan, X. H. Zhan, C. J. Herbstritt, A. Ruiz, L. Zhang, A. D. Hanson, B. P. Conner, J. Rougas and V. S. Pribluda: Withaferin A is a potent inhibitor of angiogenesis. *Angiogenesis*, 7(2), 115-22 (2004)

82. J. N. Dhuley: Nootropic-like effect of ashwagandha (Withania somnifera L.) in mice. *Phytother Res*, 15(6), 524-8 (2001)

83. D. Rai, G. Bhatia, T. Sen and G. Palit: Anti-stress effects of Ginkgo biloba and Panax ginseng: a comparative study. *J Pharmacol Sci*, 93(4), 458-64 (2003)

84. R. Archana and A. Namasivayam: Antistressor effect of Withania somnifera. *J Ethnopharmacol*, 64(1), 91-3 (1999)

85. S. K. Bhattacharya and A. V. Muruganandam: Adaptogenic activity of Withania somnifera: an experimental study using a rat model of chronic stress. *Pharmacol Biochem Behav*, 75(3), 547-55 (2003)

86. P. S. Naidu, A. Singh and S. K. Kulkarni: Effect of Withania somnifera root extract on haloperidol-induced orofacial dyskinesia: possible mechanisms of action. *J Med Food*, 6(2), 107-14 (2003)

87. S. Panda and A. Kar: Changes in thyroid hormone concentrations after administration of ashwagandha root extract to adult male mice. *J Pharm Pharmacol*, 50(9), 1065-8 (1998)

88. L. C. Mishra, B. B. Singh and S. Dagenais: Scientific basis for the therapeutic use of Withania somnifera (ashwagandha): a review. *Altern Med Rev*, 5(4), 334-46 (2000)

89. A. K. Mehta, P. Binkley, S. S. Gandhi and M. K. Ticku: Pharmacological effects of Withania somnifera root extract on GABAA receptor complex. *Indian J Med Res*, 94, 312-5 (1991)

90. R. Schliebs, A. Liebmann, S. K. Bhattacharya, A. Kumar, S. Ghosal and V. Bigl: Systemic administration of defined extracts from Withania somnifera (Indian Ginseng) and Shilajit differentially affects cholinergic but not glutamatergic and GABAergic markers in rat brain. *Neurochem Int*, 30(2), 181-90 (1997)

91. M. Elsakka, E. Grigorescu, U. Stanescu, U. Stanescu and V. Dorneanu: New data referring to chemistry of Withania somnifera species. *Rev Med Chir Soc Med Nat Iasi*, 94(2), 385-7 (1990)

92. A. Grandhi, A. M. Mujumdar and B. Patwardhan: A comparative pharmacological investigation of Ashwagandha and Ginseng. *J Ethnopharmacol*, 44(3), 131-5 (1994)

93. E. M. Abdel-Magied, H. A. Abdel-Rahman and F. M. Harraz: The effect of aqueous extracts of Cynomorium coccineum and Withania somnifera on testicular development in immature Wistar rats. *J Ethnopharmacol*, 75(1), 1-4 (2001)

94. L. Arambewela and R. Silva: Withania somnifera. *Ceylon Institute of Scientific and Industrial Research press,* Colombo (1999)

95. J. N. Dhuley: Effect of ashwagandha on lipid peroxidation in stress-induced animals. *J Ethnopharmacol*, 60(2), 173-8 (1998)

96. S. Panda and A. Kar: Evidence for free radical scavenging activity of Ashwagandha root powder in mice. *Indian J Physiol Pharmacol*, 41(4), 424-6 (1997)

97. J. N. Dhuley: Adaptogenic and cardioprotective action of ashwagandha in rats and frogs. *J Ethnopharmacol*, 70(1), 57-63 (2000)

98. S. Panda and A. Kar: Withania somnifera and Bauhinia purpurea in the regulation of circulating thyroid hormone concentrations in female mice. *J Ethnopharmacol*, 67(2), 233-9 (1999)

99. N. N. Rege, U. M. Thatte and S. A. Dahanukar: Adaptogenic properties of six rasayana herbs used in Ayurvedic medicine. *Phytother Res*, 13(4), 275-91 (1999)

100. R. P. Samy, P. N. Pushparaj and P. Gopalakrishnakone: A compilation of Bioactive Compounds from Ayurveda. *Bioinformation*, 3(3), 100-10 (2008)

101. L. F. Nikolaeva, Dedov, II and V. A. Kurbanov: [Sexual functions of men with a history of myocardial infarct during rehabilitation]. *Kardiologiia*, 26(7), 82-5 (1986)

102. P. G. Jayatilak, D. S. Pardanani, B. Dattatreya and A. R. Sheth: Effect of an indigenous drug (Speman) on accessory reproductive functions of mice. *Indian J Exp Biol*, 14(2), 170-3 (1976)

103. N. N. Rege, J. Date, V. Kulkarni, A. R. Prem, S. V. Punekar and S. A. Dahanukar: Effect of Y virilin on male infertility. *J Postgrad Med*, 43(3), 64-7 (1997)

104. R. J. Aitken and J. S. Clarkson: Cellular basis of defective sperm function and its association with the genesis of reactive oxygen species by human spermatozoa. *J Reprod Fertil*, 81(2), 459-69 (1987)

105. W. A. Pryor, K. N. Houk, C. S. Foote, J. M. Fukuto, L. J. Ignarro, G. L. Squadrito and K. J. Davies: Free radical biology and medicine: it's a gas, man! *Am J Physiol Regul Integr Comp Physiol*, 291(3), R491-511 (2006)

106. J. C. Kefer, A. Agarwal and E. Sabanegh: Role of antioxidants in the treatment of male infertility. *Int J Urol*, 16(5), 449-57 (2009)

107. V. R. Narra, R. W. Howell, K. S. Sastry and D. V. Rao: Vitamin C as a radioprotector against iodine-131 *in vivo*. *J Nucl Med*, 34(4), 637-40 (1993)

108. M. Gavazza and A. Catala: Melatonin preserves arachidonic and docosapentaenoic acids during ascorbate-Fe2+ peroxidation of rat testis microsomes and mitochondria. *Int J Biochem Cell Biol*, 35(3), 359-66 (2003)

109. R. Kutlubay, E. O. Oguz, B. Can, M. C. Guven, Z. Sinik and O. L. Tuncay: Vitamin E protection from testicular damage caused by intraperitoneal aluminium. *Int J Toxicol*, 26(4), 297-306 (2007)

110. S. Uguralp, U. Usta and B. Mizrak: Resveratrol may reduce apoptosis of rat testicular germ cells after experimental testicular torsion. *Eur J Pediatr Surg*, 15(5), 333-6 (2005)

111. T. T. Turner and J. J. Lysiak: Oxidative stress: a common factor in testicular dysfunction. *J Androl*, 29(5), 488-98 (2008)

112. D. R. Mattison: The effects of smoking on fertility from gametogenesis to implantation. *Environ Res*, 28(2), 410-33 (1982)

113. D. Wu and A. I. Cederbaum: Alcohol, oxidative stress, and free radical damage. *Alcohol Res Health*, 27(4), 277-84 (2003)

114. K. Manda, M. Ueno, T. Moritake and K. Anzai: Alpha-Lipoic acid attenuates x-irradiation-induced

oxidative stress in mice. *Cell Biol Toxicol*, 23(2), 129-37 (2007)

115. A. N. Spiess, C. Feig, W. Schulze, F. Chalmel, H. Cappallo-Obermann, M. Primig and C. Kirchhoff: Cross-platform gene expression signature of human spermatogenic failure reveals inflammatory-like response. *Hum Reprod*, 22(11), 2936-46 (2007)

116. A. A. Abu-Musa, A. H. Nassar, A. B. Hannoun and I. M. Usta: Effect of the Lebanese civil war on sperm parameters. *Fertil Steril*, 88(6), 1579-82 (2007)

117. M. Fukuda, K. Fukuda, T. Shimizu, W. Yomura and S. Shimizu: Kobe earthquake and reduced sperm motility. *Hum Reprod*, 11(6), 1244-6 (1996)

118. S. Eskiocak, A. S. Gozen, A. S. Kilic and S. Molla: Association between mental stress & some antioxidant enzymes of seminal plasma. *Indian J Med Res*, 122(6), 491-6 (2005)

119. NIH Consensus Conference. Impotence. NIH Consensus Development Panel on Impotence. *Jama*, 270(1), 83-90 (1993)

120. A. Agarwal, K. C. Nandipati, R. K. Sharma, C. D. Zippe and R. Raina: Role of oxidative stress in the pathophysiological mechanism of erectile dysfunction. *J Androl*, 27(3), 335-47 (2006)

121. D. M. Mannino, R. M. Klevens and W. D. Flanders: Cigarette smoking: an independent risk factor for impotence? *Am J Epidemiol*, 140(11), 1003-8 (1994)

122. M. Burchardt, T. Burchardt, L. Baer, A. J. Kiss, R. V. Pawar, A. Shabsigh, A. de la Taille, O. R. Hayek and R. Shabsigh: Hypertension is associated with severe erectile dysfunction. *J Urol*, 164(4), 1188-91 (2000)

123. P. Scartezzini and E. Speroni: Review on some plants of Indian traditional medicine with antioxidant activity. *J Ethnopharmacol*, 71(1-2), 23-43 (2000)

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