Scientific Basis for the Therapeutic Use of *Withania somnifera* (Ashwagandha): A Review

Lakshmi-Chandra Mishra, MD (Ayur), PhD, Betsy B. Singh, PhD, Simon Dagenais, BA

Abstract
OBJECTIVE: The objective of this paper is to review the literature regarding *Withania somnifera* (ashwagandha, WS) a commonly used herb in Ayurvedic medicine. Specifically, the literature was reviewed for articles pertaining to chemical properties, therapeutic benefits, and toxicity. DESIGN: This review is in a narrative format and consists of all publications relevant to ashwagandha that were identified by the authors through a systematic search of major computerized medical databases; no statistical pooling of results or evaluation of the quality of the studies was performed due to the widely different methods employed by each study. RESULTS: Studies indicate ashwagandha possesses anti-inflammatory, antitumor, antistress, antioxidant, immunomodulatory, hemopoetic, and rejuvenating properties. It also appears to exert a positive influence on the endocrine, cardiopulmonary, and central nervous systems. The mechanisms of action for these properties are not fully understood. Toxicity studies reveal that ashwagandha appears to be a safe compound. CONCLUSION: Preliminary studies have found various constituents of ashwagandha exhibit a variety of therapeutic effects with little or no associated toxicity. These results are very encouraging and indicate this herb should be studied more extensively to confirm these results and reveal other potential therapeutic effects. Clinical trials using ashwagandha for a variety of conditions should also be conducted.
(*Altern Med Rev* 2000;5(4) 334-346)

Introduction

Withania somnifera Dunal (ashwagandha, WS) is widely used in Ayurvedic medicine, the traditional medical system of India. It is an ingredient in many formulations prescribed for a variety of musculoskeletal conditions (e.g., arthritis, rheumatism), and as a general tonic to increase energy, improve overall health and longevity, and prevent disease in athletes, the elderly, and during pregnancy.[1,2] Many pharmacological studies have been conducted to investigate the properties of ashwagandha in an attempt to authenticate its use as a multi-purpose medicinal agent. For example, anti-inflammatory properties have been investigated to validate

Lakshmi-Chandra Mishra, MD (Ayur), PhD – Professor of Research (Adj.) Los Angeles College of Chiropractic (LACC). Correspondence address: 16200 E Amber Valley Dr, Whittier, CA 90609-1166. E-mail: lakshmimishra@lacc.edu

Betsy B. Singh, PhD – Dean of Research, LACC

Simon Dagenais, BA – Research Assistant, LACC

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

***Withania somnifera* (Ashwagandha)**

its use in inflammatory arthritis,[3-6] and animal stress studies have been performed to investigate its use as an antistress agent.[7-10] Several studies have examined the antitumor and radiosensitizing effect of WS.[11-15] The purpose of this paper is to review the literature regarding WS and report on clinically relevant studies, in an attempt to establish a scientific basis for the therapeutic use of WS. Results of studies investigating the chemistry and toxicity of WS will also be discussed.

Methods

This literature review was limited to published articles and books in the English language. Four computerized medical databases (MEDLINE, CINAHL, EMBASE, Mantis) were searched for the entire duration of each database as available on the OVID computer search service. The following keywords were used for the search: ashwagandha and common misspellings (ashwaganda, aswaganda, aswagandha), withania, somnifera, dunal, withaferin, sitoindoside, solanaceae, Indian ginseng, and winter cherry. Results of these searches were reviewed to identify relevant articles.

Results

A total of 58 articles were found using the search method described above. Research reveals ashwagandha possesses anti-inflammatory, antitumor, antistress, antioxidant, immunomodulatory, hemopoetic, and rejuvenating properties. Ashwagandha also appears to benefit the endocrine, cardiopulmonary, and central nervous systems. Few articles were discovered on the mechanism of action for these effects. Several preliminary studies have been conducted on animals. A summary of the findings of these studies is presented below.

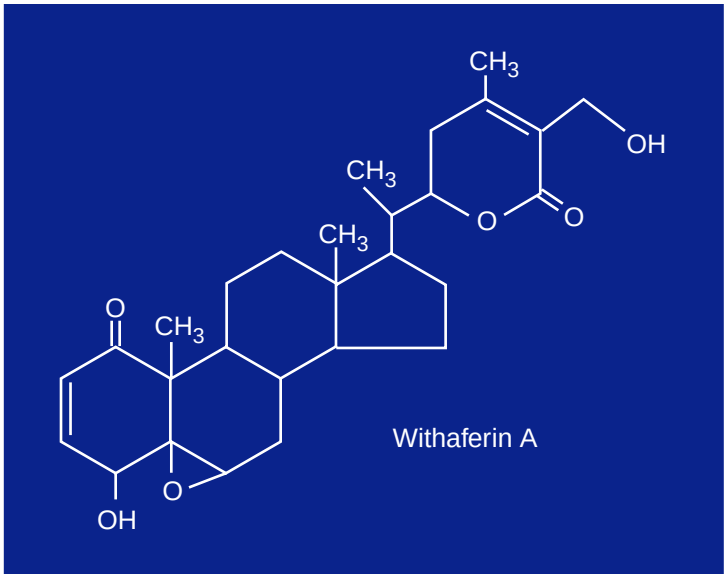
Chemistry

Since many of ashwagandha's uses have not been scientifically validated, skepticism can naturally be expected when presented with an herb purportedly useful in so many ailments. In Ayurvedic medicine there is a class of herbs, including WS, known as adaptogens or vitalizers. Adaptogens cause adaptive reactions to disease, are useful in many unrelated illnesses, and appear to produce a state of nonspecific increased resistance (SNIR)[10,16] to adverse effects of physical, chemical, and biological agents. They are relatively innocuous, have no known specific mechanism of action, normalize pathological effects, and are usually glycosides or alkaloids of a plant.[17,18]

The chemistry of WS has been extensively studied and over 35 chemical constituents have been identified, extracted, and isolated.[19] The biologically active chemical constituents are alkaloids (isopelletierine,

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

Figure 1: Chemical Structure of Withaferin A

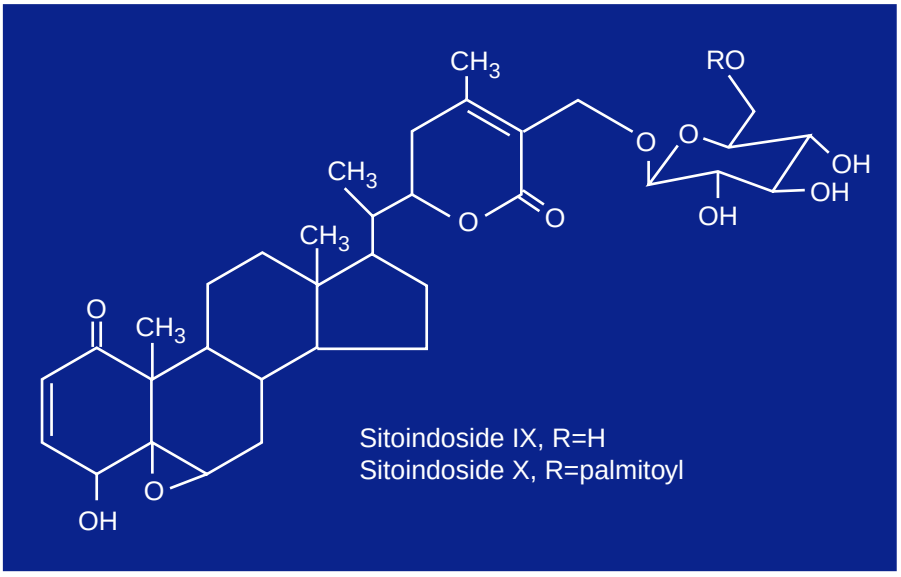


anaferine), steroidal lactones (withanolides, withaferins), saponins containing an additional acyl group (sitoindoside VII and VIII), and withanolides with a glucose at carbon 27 (sitoindoside IX and X). WS is also rich in iron. See Figure 1 for the chemical structure of withaferin A, and Figure 2 for sitoindosides IX and X.

Anti-inflammatory Properties

The effectiveness of ashwagandha in a variety of rheumatologic conditions may be due in part to its anti-inflammatory properties, which have been studied by several authors. In a study by Anbalagan et al,[3] powdered root of WS (1 g/kg suspended in 2% gum acacia, 50 mg/mL) was given orally one hour before the induction of inflammation by injection of Freund's complete adjuvant in rats and continued daily for three days; phenylbutazone (100mg/kg) was given as a positive control. WS was found to cause considerable reduction in inflammation. Acute phase reactants of the blood monitored by crossed immuno-electrophoresis showed changes in the concentration of many serum proteins (α2-glycoprotein, major acute phase α1-protein, and pre-albumin) in the WS group. The α2-glycoprotein found only in inflamed rat serum was decreased to undetectable levels in the WS group. Phenylbutazone, on the other hand, caused a considerable increase in the α2-glycoprotein in both arthritic and healthy rats. Another acute phase protein (peak 2, α-1 major acute phase) which increased approximately 200 percent by inflammation was brought back to normal levels by WS treatment but only to 132 percent of normal by phenylbutazone. WS influenced several modulator proteins in normal rats, suggesting that several plant chemicals in WS possibly interact with the liver protein synthesis process. Another study by Anbalagan

Figure 2: Chemical Structures of Sitoindosides IX and X.



Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

et al[4] found WS caused dose-dependent suppression of α2-macroglobulin (an indicator for anti-inflammatory drugs) in the serum of rats inflamed by sub-plantar injection of carrageenan suspension. The doses of WS root powder were 500, 1000, 1500, or 1200 mg/kg given as suspension orally 3-4 hours prior to induction of inflammation. Maximum effect (about 75%) was seen at 1000 mg/kg. Actual measurements of inflammation were not conducted.

In a study by Begum et al,[20] air pouch granuloma was induced by subcutaneous injections of 4 mL of two-percent (w/v) carrageenan on the dorsum of male Wistar rats (150-200 g) which had been subcutaneously injected one day prior with 6 mL of air on the dorsum. WS root powder (1000 mg/kg) was given orally from day 7 to day 9. Radioactive $Na_2{}^{32}SO^4$/100 g was injected intraperitoneally on day 9; ^{35}S incorporation in glycosaminoglycan, oxidative phosphorylation (ADP/O ratio), Mg^{2+} dependent-ATPase enzyme activity, and succinate dehydrogenase activity were determined in the mitochondria of the granuloma tissue. WS decreased the glycosaminoglycans content in the granuloma tissue by 92 percent, compared with 43.6 percent by hydrocortisone (15 mg/kg) treatment and no effect by phenylbutazone treatment (100 mg/kg). WS also uncoupled the oxidative phosphorylation by significantly reducing the ADP/O ratio in mitochondria of granuloma tissue. It increased the Mg^{2+} dependent-ATPase enzyme activity and also reduced the succinate dehydrogenase activity in the mitochondria of the granuloma tissue; no such effect was produced by the reference drugs. No physical measurements of the inflammation were carried out.

Another study by Begum et al[21] examined the effect of WS (root powder, 1000 mg/kg, orally daily for 15 days) on paw swelling and bony degenerative changes in Freund's adjuvant-induced arthritis in rats. WS caused significant reduction in both paw swelling and degenerative changes as observed by radiological examination. The reductions were better than those produced by the reference drug, hydrocortisone (15 mg/kg). No biochemical parameters were reported in this study. A study by al Hindawi et al[22] found WS inhibited the granuloma formation in cotton-pellet implantation in rats and the effect was comparable to hydrocortisone sodium succinate (5 mg/kg) treatment. Methanol extract of WS (10 mg/kg, which is one-tenth the LD_{50} dose) was given one hour before the cotton-pellet implant and continued daily until the pellets were harvested on day 4.

One clinical trial supports the possible use of WS for arthritis. In a double-blind, placebo-controlled cross-over study, 42 patients with osteoarthritis were randomized to receive a formula containing ashwagandha (see Table 1 for formula) or placebo for three months. Patients were evaluated for one month, pretreatment, during which time all previous drugs were withdrawn. During both the pretreatment and treatment phase, pain and

Table 1: Formula Used for Treatment of Osteoarthritis

Ingredient	Plant part	Dosage/capsule
Ashwagandha	root	450 mg
Boswellia	oleo gum-resin	100 mg
Turmeric	rhizome	50 mg
Zinc complex*		50 mg

* an Ayurvedic zinc complex (Jasad Bhasma) prepared by the traditional method described in Sharanghar Sambhita.

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

disability scores were evaluated weekly while erythrocyte sedimentation (SED) rate and radiological studies were conducted monthly. The herbal formula significantly reduced the severity of pain ($p<0.001$) and disability ($p<0.05$) scores, although no significant changes in radiological appearance or SED rate were noted.[23]

Few studies have been conducted on the mechanism of action for the anti-inflammatory properties of WS. In one study, rats injected with 3.5-percent formaline in the hind leg footpad showed a decrease in absorption of ^{14}C-glucose in rat jejunum.[5] Glucose absorption was maintained at the normal level by both WS and the cyclooxygenase inhibitor oxyphenbutazone. Both drugs produced anti-inflammatory effects. Similar results were obtained in parallel experiments using ^{14}C-leucine absorption from the jejunum.[6] These studies suggest cyclooxygenase inhibition may be involved in the mechanism of action of WS.

Antitumor Properties

To investigate its use in treating various forms of cancer, the antitumor and radiosensitizing effects of WS have been studied. In one study, WS was evaluated for its anti-tumor effect in urethane-induced lung adenomas in adult male albino mice.[11] Simultaneous administration of WS (ethanol extract of whole plant, 200 mg/kg daily orally for seven months) and urethane (125 mg/kg without food biweekly for seven months) reduced tumor incidence significantly (tumor incidence: untreated control, 0/25; urethane treated, 19/19; WS treated, 0/26, and WS plus urethane treated, 6/24, $p<0.05$). The histological appearance of the lungs of animals protected by WS was similar to those observed in the lungs of control animals. No pathological evidence of any neoplastic change was observed in the brain, stomach, kidneys, heart, spleen, or testes of any treated or control animals. In addition to providing protection from carcinogenic effects, WS treatment also reversed the adverse effects of urethane on total leukocyte count, lymphocyte count, body weight, and mortality.

The growth inhibitory effect of WS was also observed in Sarcoma 180 (S-180), a transplantable mouse tumor.[12] Ethanol extract of WS root (400 mg/kg and up, daily for 15 days) after intra-dermal inoculation of $5x10^5$ cells of S-180 in BALB/c mice produced complete regression of tumor after the initial growth. A 55-percent complete regression was obtained at 1000 mg/kg; however, it was a lethal dose in some cases. WS was also found to act as a radio- and heat sensitizer in mouse S-180 and in Ehrlich ascites carcinoma.[12,14] Antitumor and radiosensitizing effects of withaferin (a steroidal lactone of WS) were also seen in mouse Ehrlich ascites carcinoma *in vivo*.[15] Withaferin A from WS gave a radiosensitizer ratio of 1:5 for *in vitro* cell killing of V79 Chinese hamster cell at a non-toxic concentration of about 2 mM/L.[12-14] These studies are suggestive of antitumor activity as well as enhancement of the effects of radiation by WS.

Antistress Effect

To evaluate the antistress effect of WS, an alcohol extract from defatted seeds of WS dissolved in normal saline was given (100 mg/kg intraperitoneally as a single dose) to 20-25 g mice in a swimming performance test in water at 28º-30ºC.[10] Controls were given saline. The extracts approximately doubled the swimming time when compared to controls. In another study, WS prevented both a weight increase of the adrenals and a reduction in ascorbic acid content of the adrenals normally caused by this swimming test. The authors suggested that WS induced a state of nonspecific increased resistance during stress.

Glycosides of WS (sitoindosides VII and VIII, 50 to 100 mg/kg) exhibited significant antistress activity in forced swimming-induced immobility in mice, restraint stress-induced gastric ulcers in rats, restraint-induced

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

auto-analgesia in rats, restraint stress effect on thermic response of morphine in rats, and morphine-induced toxicity in aggregated mice.[24] The alcohol extract of WS (100 mg/kg, twice daily orally on day 1, 4 or 7) reduced stress-induced increases in blood urea nitrogen levels, blood lactic acid, and adrenal hypertrophy, but did not affect changes in thymus weight and hyperglycemia in rats.[7] WS reversed the cold swimming-induced increases in plasma corticosterone, phagocytic index, and avidity index to control levels. WS root powder (100 mg/kg orally as an aqueous suspension daily for seven days) given before the swimming test in water at 10ºC also increased total swimming time, indicating better stress tolerance in rats.[8] These results indicated a significant increase in plasma corticosterone level, phagocytic index, and avidity index in control rats, whereas these levels were near normal in WS rats subjected to the same test.

In a comparative study for antistress activity, finely powdered roots of WS and *Panax ginseng* (PG), suspended in 2-percent acacia (100 mg/kg in 1.00 mL orally) were given to 18-20 g mice daily for seven days; the swimming test was given on day 8.[25] Significant antistress activity, as measured by the swimming endurance test, was found for both compounds. The swimming time was 536.6 minutes for PG, 474.1 minutes for WS, and 163.3 minutes for controls; all differences between groups were significant ($p<0.05$). Anabolic activity, measured as an increase in body weight, was significant for both herbal extracts but was better in the WS group than in the PG group. If these results could be reproduced in humans, it would support the use of WS in nervous exhaustion due to stress and in cachexia to increase body weight.

Antioxidant Effect

The brain and nervous system are relatively more susceptible to free radical damage than other tissues because they are rich in lipids and iron, both known to be important in generating reactive oxygen species.[26] The brain also uses nearly 20 percent of the total oxygen supply.[27] Free radical damage of nervous tissue may contribute to neuronal loss in cerebral ischemia and may be involved in normal aging and neurodegenerative diseases, e.g., epilepsy, schizophrenia, Parkinson's, Alzheimer's, and other diseases.[28,29] Since traditional Ayurvedic use of WS has included many diseases associated with free radical oxidative damage, it has been considered likely the effects may be due to a certain degree of antioxidant activity. The active principles of WS, sitoindosides VII-X and withaferin A (glycowithanolides), have been tested for antioxidant activity using the major free-radical scavenging enzymes, superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPX) levels in the rat brain frontal cortex and striatum. Decreased activity of these enzymes leads to accumulation of toxic oxidative free radicals and resulting degenerative effects. An increase in these enzymes would represent increased antioxidant activity and a protective effect on neuronal tissue. Active glycowithanolides of WS (10 or 20 mg/kg intraperitoneally) were given once daily for 21 days to groups of six rats. Dose-related increases in all enzymes were observed; the increases comparable to those seen with deprenyl (a known antioxidant) administration (2 g/kg/day intraperitoneally). This implies that WS does have an antioxidant effect in the brain which may be responsible for its diverse pharmacological properties.[30] Further studies on other parts of the brain (e.g., cerebellum, medulla, and hypothalamus) may provide information with respect to the effects of WS on cognitive behavior and other functions of the brain, in both healthy and diseased individuals.

In another study, an aqueous suspension of WS root extract was evaluated for its effect on stress-induced lipid peroxidation (LPO) in mice and rabbits.[9] LPO blood levels were increased by IV administration of 0.2 mg/

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

kg of lipopolysaccharides (LPS) from *Klebsiella pneumoniae* and 100 mg/kg of peptidoglycans (PGN) from *Staphylococcus aureus*. Simultaneous oral administration of WS extract (100 mg/kg) prevented an increase in LPO. The authors indicated that the almost innocuous doses of LPS and PGN used in this study that induced elevated levels of LPO were comparable to a mild bacteremia which may follow tooth extraction, streptococcal angina, etc.

Immunomodulatory Properties

The use of WS as a general tonic to increase energy and prevent disease may be partially related to its effect on the immune system. Glycowithanolides and a mixture of sitoindosides IX and X (Figure 2) isolated from WS were evaluated for their immunomodulatory and central nervous system effects (antistress, memory, and learning) in Swiss mice (15-25 g, 5-6 months old) and Wistar strain albino rats (120-150 g and 250-300 g).[31] Both materials produced statistically significant mobilization and activation of peritoneal macrophages, phagocytosis, and increased activity of the lysosomal enzymes. Both compounds (50-200 mg/kg orally) also produced significant antistress activity in albino mice and rats, and augmented learning acquisition and memory retention in both young and old rats.

Root extract of WS was tested for immunomodulatory effects in three myelosuppression models in mice: cyclophosphamide, azathioprin, or prednisolone.[32] Significant increases ($p<0.05$) in hemoglobin concentration, red blood cell count, white blood cell count, platelet count, and body weight were observed in WS-treated mice compared to untreated control mice. The authors also reported significant increases in hemolytic antibody responses toward human erythrocytes which indicated immunostimulatory activity.

The effect of WS was also studied on the functions of macrophages obtained from mice treated with the carcinogen ochratoxin A (OTA).[33] OTA treatment of mice for 17 weeks significantly decreased the chemotactic activity of the macrophages. Interleukin-1 (IL-1) and tumor necrosis factor alpha (TNF-α) production was also markedly decreased.

Hemopoetic Effect

Administration of WS extract was found to significantly reduce leukopenia induced by cyclophosphamide (CTX) treatment in Swiss albino mice.[34] Total white blood cell count on the 12th day of the CTX-treated group was 3720/mm^3; that of the CTX-plus-WS group was 6120/mm^3. In the CTX-plus-WS mice, the cellularity of the bone marrow was significantly increased (13.1 x 10^6 /femur) ($p<0.001$) compared to the CTX-alone treated group (8 x 10^6/femur). Similarly, the number of alpha-esterase positive cells (1130/4000 cells) in the bone marrow of the CTX-plus-WS mice increased compared to the CTX-alone mice (687/4000 cells).

The major activity of WS may be the stimulation of stem cell proliferation. These studies indicated WS reduced CTX-induced toxicity and may prove useful in cancer chemotherapy. Further studies need to be conducted to confirm the hemopoetic effect with other cytotoxic agents and to determine its usefulness as an adjuvant in cancer chemotherapy.

Rejuvenating Effect

The growth-promoting effect of WS was studied for 60 days in a double-blind study of 60 healthy children, age 8-12 years, who were divided into five groups of 12.[35] Group 1 was given purified and powdered WS 2 g/day fortified in 100 cc of milk (no details about purification and powdering methods were disclosed). Similarly, Group 2 received 2 g daily of a mixture of equal parts WS and punarnava (*Boerhaavia diffusa*), Groups 3 and

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

4 were given ferrous fumarate 5 mg/day and 30 mg/day, respectively, and Group 5 received placebo.

Group 1 experienced a slight increase in hemoglobin, packed cell volume, mean corpuscular volume, serum iron, body weight, and hand grip, and significant increases in mean corpuscular hemoglobin and total proteins ($p<0.01$) at the end of 60 days when compared to the initial level and the placebo group. There was an increase in body weight in all groups over the control group.

Group 2, treated with WS and punarnava, showed a significant increase in the level of hemoglobin at the end of 30 days compared to the initial value. Marked increases in the levels of hemoglobin, packed cell volume, mean corpuscular volume, mean corpuscular hemoglobin, serum iron, and hand grip were also observed at the end of 60 days when compared to initial levels. However, when compared with the placebo group, only hemoglobin and handgrip showed significant increase ($p<0.05$). No change was seen in other parameters. It was noted that 13 of 15 children had an increase in body weight, 10 children had an increase in hemoglobin and packed cell volume, and 11 children had an increase in serum iron.

Group 3 (5 mg ferrous fumarate) had no significant change in any parameters, while Group 4 (30 mg ferrous fumarate) showed a significant increase in hemoglobin ($p<0.01$), mean corpuscular hemoglobin ($p<0.05$), mean corpuscular hemoglobin concentration ($p<0.01$), serum iron, ($p<0.05$), and hand grip ($p<0.05$), and a marked increase in packed cell volume. Group 5 (placebo) had no significant change in any parameter. The study demonstrated that WS may be useful as a growth promoter and hematinic in growing children.

In another clinical trial, WS purified powder was given 3 g/day for one year to 101 normal healthy male volunteers, age 50-59 years.[36] All subjects showed significantly increased hemoglobin and RBC count, and improvement in hair melanin and seated stature. They also showed decreased SED rate, and 71.4 percent of the subjects reported improvement in sexual performance. In summary, these studies indicate WS may prove useful in younger as well as older populations as a general health tonic.

Nervous System Effects

Total alkaloid extract (ashwagandholine, AG) of WS roots has been studied for its effects on the central nervous system.[37] AG exhibited a taming effect and a mild depressant (tranquilizer) effect on the central nervous system in monkeys, cats, dogs, albino rats, and mice. AG had no analgesic activity in rats but increased Metrazol toxicity in rats and mice, amphetamine toxicity in mice, and produced hypothermia in mice. It also potentiated barbiturate-, ethanol-, and urethane-induced hypnosis in mice.

Effects of sitoindosides VII-X and withaferin isolated from aqueous methanol extract of roots of cultivated varieties of WS were studied on brain cholinergic, glutamatergic and GABAergic receptors in male Wistar rats.[38] The compounds slightly enhanced acetylcholinesterase (AChE) activity in the lateral septum and globus pallidus, and decreased AChE activity in the vertical diagonal band. These changes were accompanied by enhanced M_1-muscarinic-cholinergic receptor-binding in lateral and medial septum as well as in frontal cortices, whereas the M_2-muscarinic receptor-binding sites were increased in a number of cortical regions including cingulate, frontal, piriform, parietal, and retrospinal cortex. The data suggest the compounds preferentially affect events in the cortical and basal forebrain cholinergic-signal-transduction cascade. The drug-induced increase in cortical muscarinic acetylcholine-receptor capacity might partly explain the cognition-enhancing and memory-improving effects of WS extracts in animals and in humans.

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

Ashwagandholine, total alkaloids extracted from extract of WS roots, caused relaxant and antispasmodic effects against various agents that produce smooth muscle contractions in intestinal, uterine, tracheal, and vascular muscles.[39] The pattern of smooth muscle activity was similar to that of papaverine, but several-fold weaker, which indicated a direct musculotropic action. These results are consistent with the use of WS to produce relaxation.

Effects on the Endocrine System

Based on the observations that WS provides protection from free radical damage in the mouse liver, studies were conducted to determine the efficacy of WS in regulating thyroid function.[40,41] Mice were given WS root extract (1.4 g/kg by gavage, daily for 20 days). The treatment significantly increased the serum levels of 3,3',5-triiodothyronine (T3) and tetraiodothyronine (T4), while the hepatic concentrations of glucose 6-phosphatase activity and hepatic iodothyronine 5'-monodeiodinase activity did not change significantly. WS significantly reduced hepatic lipid peroxidation and increased the activity of superoxide dismutase and catalase. The results suggest WS stimulates thyroidal activity and also promotes hepatic antioxidant activity.

A combination formula of WS, *Tinospora cordifolia, Eclipta alba, Ocimium sanctum, Picorrhiza kurroa,* and *shilajit* was found to cause a dose-related decrease in streptozotocin-induced hyperglycemia.[31] None of the herbs given individually, however, produced any effect on the hyperglycemia, indicative perhaps of why Ayurvedic medicine generally prefers combinations of herbs rather than single herbs.

Effects on the Cardiopulmonary System

WS may be useful as a general tonic, due in part to its beneficial effects on the cardiopulmonary system, as reported in the following studies. The effect of AG was studied on the cardiovascular and respiratory systems in dogs and frogs.[42] The alkaloids had a prolonged hypotensive, bradycardiac, and respiratory-stimulant action in dogs. The study found that the hypotensive effect was mainly due to autonomic ganglion blocking action and that a depressant action on the higher cerebral centers also contributed to the hypotension. The alkaloids stimulated the vasomotor and respiratory centers in the brain stem of dogs. The cardio-inhibitory action in dogs appeared to be due to ganglion blocking and direct cardio-depressant actions. The alkaloids produced immediate predominant but short-lived cardio-depressant effects and a weak but prolonged cardiotonic effect in isolated normal and hypodynamic frog hearts. The pharmacological actions of the total extract of WS roots on the cardiovascular and respiratory systems appeared to be due to its alkaloid content. The total alkaloids were more than twice as active as the 70-percent alcohol extract of WS root. These studies were found to be consistent with the use of WS as a tranquilizing agent.

General Toxicity Studies

An important consideration when investigating the medicinal properties of an unknown compound is diligent evaluation of its potential for harmful effects, usually evaluated through toxicity studies. For WS, no systematic study was found which included acute, sub-acute, sub-chronic or chronic toxicity of WS root powder, whole plant powder, or different extracts of the plant (e.g., water, alcohol, petroleum ether, purified alkaloids, and glycosides). The acute toxicity data found as a part of pharmacological studies are summarized here. Although one preliminary toxicity study of WS was conducted, it was of insufficient quality to support its findings as too few animals were used, body weight data was not collected, and

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

survival data was not reported.[43] In one central nervous system study, a two-percent suspension of ashwagandholine (total alkaloids from the roots of WS) prepared in ten-percent propylene glycol using two-percent gum acacia as suspending agent was used to determine acute toxicity.[39] The acute LD_{50} was 465 mg/kg (332-651 mg/kg) in rats and 432 mg/kg (299-626 mg/kg) in mice.

In an antistress-effect study, an alcohol extract from defatted seeds of WS dissolved in normal saline was used to study LD_{50} in albino mice.[10] The acute LD_{50} was 1750 +/- 41 mg (p.o). In another antistress-effect study, aqueous-methanol extracts of the root from one-year-old cultivated WS (SG-1) and equimolar combinations of sitoindosides VII and VIII and withaferin-A (SG-2) were studied for acute toxicity.[25] The acute LD_{50} of SG-1 and SG-2 by intraperitoneal administration in mice was 1076 +/- 78 mg/kg and 1564 +/- 92 mg/kg, respectively.

In one long-term study, WS was boiled in water and administered to rats in their daily drinking water for eight months while monitoring body weight, general toxicity, well being, number of pregnancies, litter size, and progeny weight.[44] The estimated dose given was 100 mg/kg/day. In the second part of the study, the estimated dose was 200 mg/kg/day given for four weeks as above while monitoring body temperature, body weight, cortisol value in heparinized plasma, and ascorbic acid content of the adrenals. The liver, spleen, lungs, kidneys, thymus, adrenals, and stomach were examined histopathologically and were all found to be normal. The initial average body weights of the WS-treated group (100 mg/kg/day) and control group on day 1 were 91 g and 106 g which, after four weeks, increased to 185 g (103%) and 178 g (67.9%), respectively. The WS-treated rats appear to have gained more weight than the control group (no p value given). The percent weight gain after eight weeks on the same WS treatment was 227 percent for the treated group and 145.3 percent in the control group. The relative body weight gain was significantly greater in the WS-treated group as compare to the control group ($p < 0.001$). While it is not clear when the rats were mated, the average weights of the progeny at one month of age were 70 g and 45 g in the WS-treated and control groups, respectively, indicating healthier progeny in the WS-treated group. Additional studies are necessary to confirm these findings.

In the four-week study, the weight gain in the treated group was comparable to that of the control group. The body temperature in the WS treated group was 1.7ºC lower than the controls. The WS treatment caused an increase in lung and liver weights and a decrease in adrenocortical activity as was evident from the reduction in adrenal weight and a significant reduction in plasma cortisol ($p<0.001$). Histopathologically, all organs were normal. The authors attributed the increase in liver weight to an increase in glycogen storage because WS contains many steroids and glucocorticoids known to enhance liver glycogen stores. Reduction in metabolic rate also leads to underutilization of glycogen stores in the liver, leading to its accumulation. The reduced adrenocortical activity may be attributed to steroid moieties in WS roots which may act like exogenous adrenocortical steroids, lowering the ACTH secretion and consequently, endogenous steroid production. The authors concluded the decoction of WS promoted growth especially during the active growth period and helped produce healthier progeny. The WS group was devoid of any toxic effects after eight months of daily dosing in this study.

Discussion

As outlined above, results from various studies indicate ashwagandha possesses many qualities, including anti-inflammatory, antitumor, and immunomodulatory properties, as well as exerting an influence on the endocrine, nervous, and cardiopulmonary systems. Further clinical studies should be

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

conducted, as well as studies in multiple animal-based models using a variety of suitable biochemical markers (e.g., urinary excretion of pyridinoline and deoxypyridinoline) to understand its mechanism of action. Any protective or prophylactic effect it may have on the development of arthritis should also be investigated, as well as effects it may have on cartilage degradation or regeneration. As for its use in fighting cancer, confirmatory studies in several other animal tumor systems must be conducted for more definitive findings. Studies should also be carried out to determine the effects, if any, of WS on existing antitumor agents when given in combination with WS. Regarding the effects observed in animals on the endocrine and cardiopulmonary systems, the therapeutic significance of these biochemical markers is not clear. Studies point to a possible benefit of WS in central nervous system-related ailments. The lack of systematic toxicity studies is of some concern, as is the poor quality of the existing toxicity studies.

The review indicates that WS may be useful in many ailments, including arthritis and other musculoskeletal disorders, stress-induced nervous exhaustion, and hypertension. There are a few preliminary studies available on the effects of WS on the immune system, central nervous system, hemopoetic system, and general growth promotion to form a basis for further studies but not enough evidence to provide a firm scientific basis for definitive therapeutic uses.

Conclusion

Although the results from this review are quite promising for the use of ashwagandha as a multi-purpose medicinal agent, several limitations currently exist in the current literature. While ashwagandha has been used successfully in Ayurvedic medicine for centuries, more clinical trials should be conducted to support its therapeutic use. It is also important to recognize that WS may be effective not only in isolation, but may actually have a potentiating effect when given in combination with other herbs or drugs.

Acknowledgment: The authors acknowledge the technical support provided by Fred Zarow, DC, MS in the preparation of this manuscript.

References

1. Chatterjee A, Pakrashi SC. *The Treatise on Indian Medicinal Plants* 1995;4:208-212.
2. Bone K. *Clinical Applications of Ayurvedic and Chinese Herbs. Monographs for the Western Herbal Practitioner*. Australia: Phytotherapy Press; 1996:137-141.
3. Anbalagan K, Sadique J. Influence of an Indian medicine (Ashwagandha) on acute-phase reactants in inflammation. *Indian J Exp Biol* 1981;19:245-249.
4. Anbalagan K, Sadique J. Role of prostaglandins in acute phase proteins in inflammation. *Biochem Med* 1984;31:236-245.
5. Somasundaram S, Sadique J, Subramoniam A. Influence of extra-intestinal inflammation on the in vitro absorption of 14C-glucose and the effects of anti-inflammatory drugs in the jejunum of rats. *Clin Exp Pharmacol Physiol* 1983;10:147-152.
6. Somasundaram S, Sadique J, Subramoniam A. *In vitro* absorption of [14C]leucine during inflammation and the effect of antiinflammatory drugs in the jejunum of rats. *Biochem Med* 1983;29:259-264.
7. Dadkar VN, Ranadive NU, Dhar HL. Evaluation of antistress (adaptogen) activity of Withania somnifera (Ashwagandha). *Ind J Clin Biochem* 1987,2:101-108.
8. Archana R, Namasivayan A. Antistressor effect of Withania somnifera. *J Ethnopharmacol* 1999;64:91-93.
9. Dhuley JN. Effect of ashwagandha on lipid peroxidation in stress-induced animals. *J Ethnopharmacol* 1998;60:173-178.
10. Singh N, Nath R, Lata A, et al. Withania somnifera (ashwagandha), a rejuvenating herbal drug which enhances survival during stress (an adaptogen). *Int J Crude Drug Res* 1982;20:29-35.

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

11. Singh N, Singh SP, Nath R, et al. Prevention of urethane-induced lung adenomas by Withania somnifera (L.) Dunal in albino mice. *Int J Crude Drug Res* 1986;24:90-100.
12. Devi PU, Sharada AC, Solomon FE, Kamath MS. *In vivo* growth inhibitory effect of Withania somnifera (Ashwagandha) on a transplantable mouse tumor, Sarcoma 180. *Indian J Exp Biol* 1992;30:169-172.
13. Devi PU. Withania somnifera dunal (ashwagandha): potential plant source of a promising drug for cancer chemotherapy and radiosensitization. *Indian J Exp Biol* 1996;34:927-932.
14. Devi PU, Sharada AC, Solomon FE. *In vivo* growth inhibitory and radiosensitizing effects of withaferin A on mouse Ehrlich ascites carcinoma. *Cancer Lett* 1995;95:189-193.
15. Sharad AC, Solomon FE, Devi PU, et al. Antitumor and radiosensitizing effects of withaferin A on mouse Ehrlich ascites carcinoma in vivo. *Acta Oncol* 1996;35:95-100.
16. Lazarev NV. *Farmacol Toxicol* 1958;21:81-86. As cited in Brekhman II, Dardymov IV. New substances of plant origin which increase non-specific resistance. *Annu Rev Pharmacol* 1969;9:419-430.
17. Brekhman II. Panax Ginseng. *Med Sci Serv* 1967;4:17-26.
18. Brekhman II, Dardymov IV. New substances of plant origin which increase non-specific resistance. *Annu Rev Pharmacol* 1969;9:419-430.
19. Rastogi RP, Mehrotra BN. *Compendium of Indian Medicinal Plants*, Vol.6. Central Drug Research Institute, New Delhi, 1998.
20. Begum VH, Sadique J. Effect of Withania somnifera on glycosaminoglycan synthesis in carrageenin-induced air pouch granuloma. *Biochem Med Metab Biol* 1987;38:272-277.
21. Begum VH, Sadique J. Long term effect of herbal drug Withania somnifera on adjuvant induced arthritis in rats. *Indian J Exp Biol* 1988;26:877-882.
22. al Hindawi MK, al Khafaji SH, Abdul-Nabi MH. Anti-granuloma activity of Iraqi Withania somnifera. *J Ethnopharmacol* 1992;37:113-116.
23. Kulkarni RR, Patki PS, Jog VP, et al. Treatment of osteoarthritis with a herbomineral formulation: a double-blind, placebo-controlled, cross-over study. *J Ethnopharmacol* 1991;33:91-95.
24. Bhattacharya SK, Goel RK, Kaur R, Ghosal S. Antistress activity of sitoindosides VII and VIII, new acylsterylglucosides from Withania somnifera. *Phytotherapy Res* 1987;1:32-39.
25. Grandhi A, Mujumdar AM, Patwardhan B. A comparative pharmacological investigation of Ashwagandha and Ginseng. *J Ethnopharmacol* 1994;44:131-135.
26. Halliwell B, Gutteridge JMC. *Free Radicals in Biology & Medicine*. 2nd ed. Oxford: Clarendon Press; 1989.
27. Ames BN, Shigenaga MK, Hagen TM. Oxidants, antioxidants, and the degenerative diseases of aging. *Proc Natl Acad Sci U S A* 1993;90:7915-7922.
28. Jesberger JA, Richardson JS. Oxygen free radicals and brain dysfunction. *Int J Neurosci* 1991;57:1-17.
29. Scarfiotti C, Fabris F, Cestaro B, Giuliani A. Free radicals, atherosclerosis, ageing, and related dysmetabolic pathologies: pathological and clinical aspects. *Eur J Cancer Prev* 1997;6:S31-S36.
30. Bhattacharya SK, Satyan KS, Chakrabarti A. Effect of Trasina, an Ayurvedic herbal formulation, on pancreatic islet superoxide dismutase activity in hyperglycaemic rats. *Indian J Exp Biol* 1997;35:297-299.
31. Ghosal S, Lal J, Srivastava R, et al. Immunomodulatory and CNS effects of sitoindosides IX and X, two new glycowithanolides from Withania somnifera. *Phytotherapy Res* 1989;3:201-206.
32. Ziauddin M, Phansalkar N, Patki P, et al. Studies on the immunomodulatory effects of ashwagandha. *J Ethnopharmacol* 1996 Feb;50:69-76.
33. Dhuley JN. Effect of some Indian herbs on macrophage functions in ochratoxin A treated mice. *J Ethnopharmacol* 1997;58:15-20.
34. Davis L, Kuttan G. Suppressive effect of cyclophosphamide-induced toxicity by Withania somnifera extract in mice. *J Ethnopharmacol* 1998;62:209-214.
35. Venkataraghavan S, Seshadri C, Sundaresan TP, et al. The comparative effect of milk fortified with aswagandha, aswagandha and punarnava in children – a double-blind study. *J Res Ayur Sid* 1980;1:370-385.

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission

36. Kuppurajan K, et al. *J Res Ayu Sid* 1, 247, 1980. As cited in Bone K. *Clinical Applications of Ayurvedic and Chinese Herbs. Monographs for the Western Herbal Practitioner*. Australia: Phytotherapy Press; 1996:137-141.
37. Malhotra CL, Mehta VL, Das PK, Dhalla NS. Studies on Withania-ashwagandha, Kaul. V. The effect of total alkaloids (ashwagandholine) on the central nervous system. *Indian J Physiol Pharmacol* 1965;9:127-136.
38. Schliebs R, Liebmann A, Bhattacharya SK, et al. 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* 1997;30:181-190.
39. Malhotra CL, Mehta VL, Prasad K, Das PK. Studies on Withania ashwagandha, Kaul. IV. The effect of total alkaloids on the smooth muscles. *Indian J Physiol Pharmacol* 1965;9:9-15.
40. Panda S, Kar A. Evidence for free radical scavenging activity of ashwagandha root powder in mice. *Indian J Physiol Pharmacol* 1997;41:424-426.
41. Panda S, Kar A. Changes in thyroid hormone concentrations after administration of ashwagandha root extract to adult male mice. *J Pharm Pharmacol* 1998;50:1065-1068.
42. Malhotra CL, Das PK, Dhalla NS, Prasad K. Studies on Withania ashwagandha, Kaul. III. The effect of total alkaloids on the cardiovascular system and respiration. *Indian J Med Res* 1981;49:448-460.
43. Arseculeratne SN, Gunatilaka AA, Panabokke RG. Studies of medicinal plants of Sri Lanka. Part 14: Toxicity of some traditional medicinal herbs. *J Ethnopharmacol* 1985;13:323-335.
44. Sharma S, Dahanukar S, Karandikar SM. Effects of long-term administration of the roots of ashwagandha (Withania somnifera) and shatavari (Asparagus racemosus) in rats. *Indian Drugs* 1986;23:133-139.

Copyright©2001 Thorne Research, Inc. All Rights Reserved. No Reprints Without Written Permission