Exposition of Ardisia Humilis Vahl in Ameliorating Hepatic Manifestations

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By

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To my students who love and care for nature ….

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Dr. Biman Bhuyan
Place: Dibrugarh
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BOOK DESCRIPTION

Herbal medicines are fast becoming the default choice for the treatment of diseases. They are gaining widespread popularity due to their general lack of side effects. They are extensively used in the developing world, where, in many places, they offer a more widely available and affordable alternative to synthetic conventional drugs. Most research has focused on clinical and experimental medicine (safety, efficacy, and mechanism of action) and regulatory issues, to the general neglect of public health dimensions.

The book titled "EXPOSITION OF *ARDISIA HUMILIS* VAHL IN AMELIORATING HEPATIC MANIFESTATIONS" is an illustrative account of the usage of *Adrisia humilis* Vahl. in the traditional medicinal system and its subsequent systematic validation and experimental standardization with special emphasis on its prospects in stabilizing hepatic manifestations. It tries to cover the basic aspects regarding phytocomposition, pharmacopoeial standardization, *and in vitro* and *in vivo* studies aimed at verifying its use in the traditional system of medicine. The work embedded in the book successfully upheld the folkloric claims made by traditional practitioners in the treatment of jaundice.

PREFACE

Herbal medicines are fast becoming the default choice for the treatment of diseases. They are gaining widespread popularity due to their general lack of side effects. They are extensively used in the developing world, where, in many places, they offer a more widely available and affordable alternative to synthetic drugs. Most research has focused on clinical and experimental medicine (safety, efficacy, and mechanism of action) and regulatory issues, to the general neglect of public health dimensions. Public health research must consider social, cultural, political, and economic contexts to maximize the contribution of herbal medicine to health care systems globally. Herbal plants or botanical medicines have been traditionally used by herbalists worldwide for the prevention and treatment of liver disease. Considerable studies have been carried out on ethnomedical plants; however, only a few medicinal plants have attracted the interest of scientists to investigate them for a remedy for jaundice.

Assam, one of the biodiversity hotspots, occupies a special place in north-east India. Various medicinal plants for health care are being used for treatments and therapies by village-level healers and practitioners widely in Assam. There are many plants used traditionally for the treatment of liver disorders, but research in this area is still at a primitive stage. The ethnomedicinal documentation, along with the evaluation of the hepatoprotective activity of those plants, may lead to the discovery of new remedies or isolated active compounds.

Ardisia humilis Vahl (Dhsponpoita) is used by traditional healers in the treatment of jaundice and also in fever, diarrhoea, rheumatic arthritis, rheumatism, skin soreness, and vertigo. Roots are used in fever, diarrhoea, and rheumatism, and they have antibacterial activity. Evaluation of hepatoprotective activity will authenticate its traditional use in liver disorders and ensure future scope for this very plant.

The preliminary study involved the evaluation of macroscopic and microscopic characteristics along with the determination of various physicochemical parameters such as ash values and extractive values, in addition to a qualitative analysis of the phytocomponent environment of

the plant. Quantitative estimation of phenols and flavonoids was also carried out to add value to the traditional claims.

The effectiveness of the plant in biological systems was successfully analyzed using *in vitro* and *in vivo* methods. To curtail any toxic manifestations that the plant may have, toxicity studies in compliance with OECD guidelines along with histopathological examinations were also done.

The findings of the study were encouraging, as it was successful in backing the claims put forward by traditional healers in the treatment of jaundice.

The entire work has been formatted in the form of a book to enable research workers working in this area to draw on suitable references to carry the work to the next level.

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INTRODUCTION

BIMAN BHUYAN AND JENTINOCHET AMRI

Medicinal plants play an important role in the development of potent therapeutic agents. Today it is estimated that about 80% of people in developing countries still rely on traditional medicine based largely on species of plants and animals for their primary health care. Herbal medicines are currently in demand and their popularity is increasing day by day. About 500 plants with medicinal use are mentioned in ancient literature and around 800 plants have been used in indigenous systems of medicine.

Herbal drug referred to as plant materials or herbals, involves the whole plants or part of plants, to treat injuries or illnesses (**Winslow & Kroll1 1998, 2192-2199**). Herbal drugs are the use of therapeutic herbs to prevent and treat diseases and ailments or to support health and healing (**Gossell-Williams & West 2006, 217-218**). These drugs or preparations are made from a plant or plants and used for any of such purposes. Herbal drugs are the oldest form of health care known to mankind (**De Smet 1997, 801-840**). World Health Organisation (WHO) has distinct herbal drugs as complete, labeled medicinal products that have vigorous ingredients, aerial or secretive parts of the plant, or other plant material or combinations. The World Health Organisation has precise guidelines for the evaluation of the safety, efficacy, and quality of herbal medicines (**WHO 1991, 1-4**).

The World Health Organisation (WHO) has listed 21,000 plants, which have been used for medicinal purposes around the world. Among these, 2,500 species are found in India, out of which 150 species are used commercially on large scales. India is the largest producer of medicinal herbs and it is called the botanical garden of the world (**Kumar & Kumar 2009, 1-20**).

Herbal drug is a chief constituent in traditional medicine and a common constituent in Ayurvedic, homeopathic, naturopathic, and other medical system. Herbs are usually considered safe since they belong to natural sources (**Rane Jadhao Bakal 2016, 24-36**). The use of herbal drugs due to the toxicity and side effects of allopathic medicines has led to a rapid increase in the number of herbal drug manufacturers. For the past few decades, herbal drugs have been more and more consumed by people with no prescription. These drugs have survived real-world testing and thousands of years of human testing. Some drugs have been discontinued due to their toxicity while others have been modified or combined with additional herbs to counterbalance side effects (**Atmakuri & Dathi 2010, 109-113**).

1.1 Herbal medicine market

As per available records, the herbal medicine market in 1991 in the countries of the European Union was about $ 6 billion (maybe over $20 billion), with Germany accounting for $ 3 billion, France $1.6 billion, and Italy $0.6 billion. Incidentally in Germany and France, herbal extracts are sold as prescription drugs and are covered by national health insurance. In 1996, the US herbal medicine market was about $ 4 billion and the current growth rate may be double by the turn of the century (**Nirali & Shankar 2016, 59-65**). Thus a reasonable guesstimate for the current herbal medicine market worldwide may be around $ 30$ to 60 billion. Global Industry Analysts, Inc. (GIA) announced the release of a comprehensive global report on the Herbal Supplements and Remedies Market. According to GIA, the global herbal supplements and remedies market is projected to reach US$115 billion by 2020, driven by the growing adoption of preventive care as a safe and effective strategy to improve health and well-being. The Indian herbal drug market is about $ 1 billion and the export of herbal crude extracts is about $80 million (**Kamboj 2000, 35-39**).

1.1.1 Overview of herbal medicine

Herbal medicines are a part of Traditional Medicine or alternative or complementary medicine. Herbal medicines are classified as herbs, herbal supplements, herbal preparations, and finished herbal products that contain parts of herbal plants or other herbal plant materials as key ingredients (**Nirali & Shankar 2016, 59-65**). Herbal medicines are extensively used in the developing world, where in many places they offer a more widely available and more affordable alternative to pharmaceutical drugs. Herbal

medicine is the use of plants, plant parts, their water or solvent extracts, essential oils, gum, resins, exudates, or other form of advanced products made from plant parts. About 70-80% of the world population, particularly in developing countries rely on non-conventional medicine in their primary healthcare as reported by the World Health Organisation (**Akerele1993, 9-13**). The desire to capture the wisdom of traditional healing systems has led to a resurgence of interest in herbal medicines, particularly in Europe and North America, where herbal products have been incorporated into so-called 'alternative', 'complementary', 'holistic' or 'integrative' medical systems (**Nirali & Shankar 2016, 59-65**).

During the latter part of the twentieth century, increasing interest in self-care resulted in an enormous growth in the popularity of traditional healing modalities, including the use of herbal remedies, this has been particularly true in the USA. Consumers have reported positive attitudes towards these products, in large part because they believe them to be of 'natural' rather than 'synthetic' origin, they believe that such products are more likely to be safe than are drugs, they are considered part of a healthy lifestyle and they can help to avoid unnecessary contact with conventional 'western medicine'(**Awang 1997,341-344**).

A large number of plants and formulations have been claimed to have hepatoprotective activity. Nearly 160 phytoconstituents from 101 plants have been claimed by the Pharmacopoeia Foundation to possess liver-protecting activity. In India, more than 87 plants are used in 33 patented and proprietary multi-ingredient plant formulations (**Roy Das Shil Dutta 2012, 87-99**). Despite the tremendous advances made, no significant and safe Hepatoprotective agents are available in modern therapeutics. For eg. Silymarin is a potent hepatoprotective drug that has established a place in hepatology practice. Silymarin is a flavono-lignan mixture obtained from seeds of *Silybum marianum*. Silymarin is a mixture of silybin, isosilybin, and silychristinanssilydianin. Research on Indian medicinal herbs like *Picrorhiza kurroa* (Kutaki) and *Andrographis paniculata* (Kalmegh) has thrown light on hepatoprotective activity and it is more promising than silymarin (**Kamboj 2000, 35-39**).

1.1.2 Herbal Medicine Scenario in India

The turnover of herbal medicines in India as over-the-counter products, ethical and classical formulations, and home remedies of Ayurveda, Unani, and Siddha systems of medicine is about $1 billion with a meager

export of about $80 million **(Vaidya & Thomas 2007, 1-11)**. Psyllium seeds and husk, castor oil, and opium extract alone account for 60% of the exports. 80% of the exports to developed countries are of crude drugs and not finished formulations leading to a low revenue for the country. Thus the export of herbal medicines from India is equivalent to ¾ of its land-exclusive economic zone in the ocean harboring a large variety of flora and fauna, many of them with therapeutic properties. About 1500 plants with medicinal uses are mentioned in ancient texts and around 800 plants have been used in traditional medicine **(Dubey Kumar Tripathi 2004, 37–41).**

The major traditional sector pharma, namely Himalaya, Zandu, Dabur, Hamdard, Maharishi, etc., and modern sector pharma namely Ranbaxy, Lupin, Alembic, etc. are standardizing their herbal formulation by chromatography techniques like TLC/HPLC fingerprinting, etc. There are about 700 firms in the small-scale sector manufacturing traditional medicines with or without standardization. However, none of the pharma has standardized herbal medicine using active compounds as markers linked with confirmation of the bioactivity of herbal drugs in experimental models **(Shi Song Li Yang *et al.* 2014, 78-85).**

1.1.3 Role of WHO in herbal medicine

Two decades ago, the WHO referred to traditional health systems (including herbal medicine) as 'holistic'- 'that a viewing man in his totality within a wide ecological spectrum and emphasizing the view that ill health or disease is brought about by an imbalance or disequilibrium of man in his total ecological system and not only by causative agent and pathogenic evolution, probably implying that the indigenous system drugs (including herbal medicine) restore the imbalanced or disequilibrium leading to the cure of ill health or disease. Such an attitude sent signals that WHO as an organization has failed to provide leadership to establish traditional systems of medicine that provide health care to about 80% of the world population. However, it helped the inclusion of proven traditional remedies in national drug policies and regulatory approvals by developing countries (**Kumar & Kumar 2009, 1-20**). The World Health Assembly continued to debate and adopted a resolution (WHA 42,43) in 1989 that herbal medicine is of great importance to the health of individuals and communities. The redefined definition of traditional medicine thus issued in the early nineties is given by supra. Consequently, in 1991 WHO developed guidelines for the assessment of herbal medicine, and the same

were ratified by the 6th International Conference of Drug Regulatory Authorities held in Ottawa in the same year (**WHO 1991, 1-4**).

1.1.4 Herbal medicine standardization

In indigenous/traditional systems of medicine, the drugs used are primarily dispensed as water decoctions or ethanolic extracts. Fresh plant parts, juice, or crude powder are a rarity rather than a rule. Thus medicinal plant parts should be authentic and free from harmful materials like pesticides, heavy metals, microbial or radioactive contamination, etc. The medicinal plant is subjected to a single solvent extraction once or repeatedly, or water decoction as described in ancient texts. The extract should then be checked for indicated biological activity in an experimental animal model. The bioactive extract should be standardized based on active principles or major compounds along with fingerprints. The next important step is the stabilization of the bioactive extract with a minimum shelf-life of over a year. The stabilized bioactive extract should undergo regulatory or limited safety studies in animals. Determination of the probable mode of action will explain the therapeutic profile. The safe and stable herbal extract may be marketed if its therapeutic use is well documented in indigenous systems of medicine, as also argued by WHO (**Kumar & Kumar 2009, 1-20**), (**WHO 1991, 1-4**). A limited clinical trial to establish its therapeutic potential would promote clinical use. The herbal medicines developed in this mode should be dispensed as prescription drugs or even OTC products depending upon disease consideration and under no circumstances as health foods or nutraceuticals (**Kamboj 2000, 35-39**).

1.2 Liver

The liver is a vital organ of vertebrates and some other animals (**Chattopadhyay & Bhattacharyya 2007, 151-156**). In humans, it is located in the upper right quadrant of the abdomen, below the diaphragm. The liver weighs about 1.5 kg and is made up of many lobes called hepatic lobes. Each lobe consists of many lobules called hepatic lobules. The hepatic lobule is the structural and functional unit of the liver. There are about 50,000 to 100,000 lobules in the liver. The lobule is a honeycomb-like structure and it is made up of liver cells called hepatocytes (**Sembulingam & Sembulingam 2010, 89-109**).

The liver is the main metabolic organ in the body. To be able to carry out its metabolic functions, a great part of the blood pumped out by the heart

is carried to the liver via the circulatory system. Studies have shown that about 1-1.5 liters of blood are transported to the liver every minute via the portal vein. The hepatic artery brings oxygen-rich blood to the liver while the portal vein transports nutrient-rich blood to the liver. The blood in the portal vein has already passed through the gastrointestinal tract and absorbed large amounts of nutrients (**Ramadori Moriconi Moriconi Dudas 2008, 107-117**).

1.2.1 Functions of Liver

As the main metabolic organ in the body, the liver has many different functions, including:

- Production of protein building blocks (amino acids), proteins (e.g. clotting factors, albumin), Cholesterol and bile acids
- Regulation of the blood sugar level by the production or use of glucose
- Production and supply of bile for digestion of fats
- The neutralization and elimination of waste products of the body's own metabolism and foreign substances such as drugs and environmental toxins
- Storage of nutrients (glycogen and sugar reserves), minerals (e.g. iron), or vitamins (e.g. vitamin B12)
  The liver has also many essential roles in keeping us alive including:
- Blood purification- As the journey of the liver starts throughout the body, the blood from the stomach and intestine is the liver and it prevents the contaminants and also removes the waste products from the body such as; drugs, bacteria, fungi, viruses, parasites, food additives, pesticides and herbicides, chemicals, fats, alcohol, dead cells.
- Detoxification also perform the function of detoxification as it detoxifies alcohol, heavy metal, drugs, chemicals, and toxic by-products from the blood. Housing an ingenious cleaning system, the liver detoxifies infectious organisms, alcohol, heavy metals, drugs, chemicals, toxic by-products, and other poisons from the blood
- Digestion-The liver produces bile, a substance needed to digest and absorb fats. Bile is used in digestion by helping the body absorbs fats and certain vitamins, including vitamin A, D, E, and K.

- Manufacturing- The liver manufactures variable proteins, including enzymes, hormones, blood proteins, clotting factors, and immune factors. The liver also produces cholesterol, which carries energy-supplying fats to the body. Processing- the liver performs most of its function via different organs like skin, mouth, and lungs, Considered to be the biochemical factory of the body, the liver metabolizes substances in the bloodstream.
- Storage- The cells of the liver also act as a powerhouse of the body for many substances, such as iron, vitamins, minerals, and glycogen until they are needed. When blood sugar levels drop and the body needs energy quickly, the liver converts the stored glycogen into glucose and releases it into the bloodstream. In this way, the liver supplies us with fast-acting energy (**Shivananda Julien Godwin Adogwa 2011, 1–5**), (**Akinloye & Moshood 2011, 237-241**).

1.2.2 Liver Disease

Although liver disease is stereotypically linked to alcohol or drugs, the truth is that there are 100 known forms of liver disease caused by a variety of factors affecting everyone from infants to older adults. Cirrhosis is often considered to be a form of liver disease and may be the only liver-related condition that results from permanent damage or scarring of the liver. It is the end stage of many different forms of liver disease and is known to cause several other health problems, including variceal bleeding, ascites, and hepatic encephalopathy (**Sembulingam & Sembulingam 2010, 89-109)**.

Many types of liver disease still have unknown causes but the most frequent liver diseases are generally caused by one of the following factors:

- **Viral hepatitis**

Caused by viruses that attack the liver, viral hepatitis comes in many forms. The most common forms worldwide are hepatitis A, B, and C. Although hepatitis A and B can be prevented by vaccines, there is no vaccine for hepatitis C. In Canada, hepatitis C is the leading cause of liver transplants.

- **Alcohol**

Factors such as gender, age, nationality, weight, and health can affect how a person's liver metabolizes alcohol. When the liver has too much alcohol to handle, normal liver function may be interrupted leading to a chemical imbalance. If the liver is required to detoxify alcohol continuously, liver cells may be destroyed or altered resulting in fat deposits (fatty liver) and more seriously, either inflammation (alcoholic hepatitis) or permanent scarring.

- **Genetics**

Several forms of liver disease are caused or thought to be caused, by defective genes. These forms of liver disease may be diagnosed in infancy or may not show up until later in life. Examples include Hemochromatosis, Wilson disease, Tyrosinemia, Alpha 1 Antitrypsin deficiency, and Glycogen Storage disease.

- **Drugs and toxins**

The liver is responsible for processing most of the chemicals and medications that enter your body- this leaves it vulnerable to acute or chronic liver disease caused by chemicals. In some cases, this is a predictable consequence of overexposure or over-consumption of certain chemicals such as acetaminophen or industrial toxins like polyvinyl chloride or carbon tetrachloride. In other cases, chemicals can cause an unpredictable reaction.

- **Autoimmune disorders**

Sometimes a body's immune system may begin to attack the liver or bile ducts causing inflammation and scarring which leads to a progressive form of liver disease. Examples of liver diseases believed to be caused by the immune system are primary biliary cholangitis (PBC), primary sclerosing cholangitis (PSC), and autoimmune hepatitis.

- **Cancer**

Although primary liver cancer is relatively uncommon, many other forms of cancer often metastasize in the liver. Because the liver filters a high volume of blood which may be carrying cancer cells, it is susceptible to

developing a form of secondary cancer. If cancer originates in the liver, it is often caused by hepatitis B and hepatitis C or it can develop in cases of advanced liver disease when cirrhosis is present (**Sembulingam & Sembulingam 2010, 89-109)**.

1.2.3 Signs and Symptoms of liver disease

Although the difference in signs and symptoms can vary so much from one drug to the next, and one patient to the next some common signs associated with drug-induced hepatotoxicity include; Non-specific symptoms: ones that may not directly pinpoint the problem.

- Fatigue
- Abdominal pain and swelling.
- Weakness
- Loss of appetite

Signs and symptoms specific for hepatotoxicity-

- Jaundice
- Itching
- Easily bruising

When the liver is severely damaged cirrhosis can occur. Symptoms of cirrhosis include

- Edema (oftentimes in the legs)
- Mental confusion
- Kidney failure
- Gastrointestinal bleeding
- Vulnerability to bacterial infection

Accumulated toxins in the body place a constant drag on the immune system, setting the stage for autoimmune diseases and cancer. If any organ is compromised, others will be affected, eventually leading to a cumulative negative effect on health. Some of the most toxic legal substances people flood their livers with are alcohol and medication. Alcohol- acetaldehyde is a highly toxic substance obtained from the metabolism of alcohol is the molecule that causes impairment or drunkenness. Scientists have discovered that when acetaldehyde is bound to human liver plasma membranes, liver cells die (**Rane Jadhao Bakal 2016, 24-36)**.

1.2.4 Drug-Induced Hepatotoxicity

Two drugs (Bromfenac and troglitazone) have been withdrawn from the market by the Food and Drug Administration (FDA) for causing severe liver injury, a potential danger that had not been fully recognized in the course of the preapproval clinical trials. Reports of adverse drug reactions of any type engender fear and skepticism in the public about the actions of the pharmaceutical industry and the FDA (**Lazarou Bruce Paul 1998, 1200-1205**). Drug-induced hepatic injury is the most frequent reason for the withdrawal from the market of an approved drug, and it also accounts for more than 50 percent of the cause of acute liver failure in the United States today. More than 75 percent of the case of idiosyncratic drug reactions results in liver transplantation or death (**Ostapowicz Fontana Schiødt Larson *et al.*2002, 947-954**). Recent efforts by the National Institute of Health and the FDA have been directed toward a better understanding of these occurrences to improve the outcomes (**Bissell Montgomery Gregory Debra *et al.* 2001, 1009-1013**), (**Weber Meinrad Stampfl 2003, 105-136**).

1.2.5 Mechanism

In some physiological conditions, the mitochondrial membrane can lose its structure and functional integrity by the opening of the mitochondrial permeability transitional pore (MPTP) and this can disturb ATP synthesis and storage. Impaired ATP synthesis causes a sudden increase in Intracellular calcium levels by the activation of plasma membrane calcium ATPase (PMCA (**Wallace Weiwei Procaccio2010, 297**). Sometimes MPTP opening also allows the release of proteins, such as caspases, Cytochrome C, and Apoptosis-inducing factor (**Masubuchi Chieko Toshiharu 2005, 110-116**), (**Pessayre Abdellah Berson Fromenty 2010, 311-365**). These components can bind with the Apaf protein leading to initiating the apoptosis pathway through caspase 9 and caspase 3 activation (**Fulda Lorenzo Guido 2010, 447-464**), (**Pessayre Delphine Fau Marie *et al.*1999, 760-770**). MPTM opening is mostly induced by several drugs, xenobiotics, or some endogenous by-products like calcium, and fatty acid of bile salt **(Malhi Gregory Lemasters 2006, S31-S44)**. Mitochondria produce Reactive Oxygen species (ROS) via the activity of the Mitochondrial Respiratory chain (MRC) (**Seifert Carmen Jian Harper 2010, 5748-5758**). In normal cases, ROS can be detoxified through glutathione peroxidase (GPx) using glutathione as a cofactor. Depletion in GSH causes H2O2 accumulation and this whole process turns