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## Harnessing the phytochemistry and pharmacological roles of *Andrographis paniculata*

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**Abstract:** Herbs and natural product have extremely high potential as therapeutic agent without causing serious side effects on human body. In this review article the medicinal uses of *Andrographis paniculata* an annual herb found in Asian continent and other parts of world is being discussed along with its traditional uses, phytochemistry and pharmacology.

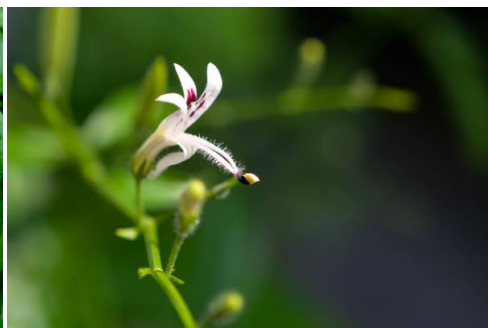
**Key words:** *Andrographis paniculata*, Traditional medicinal uses, phytochemistry, pharmacology

### **Introduction:**

*A. paniculata*, commonly known as King of Bitters or kalmegh, is an annual, branched, erect herb. It is native to peninsular India and Srilanka and is also distributed in different regions of Southeast Asia, China, America, West Indies and Christmas Island. It is cultivated because of its well known medicinal value and it is widely distributed since it grows well in most soil types<sup>1</sup>. The aerial parts and roots of the plant have been widely used as traditional medicine in China, India, Thailand and other Southeast Asian countries to treat many maladies. Its common names are King of Bitters (English), Mahatikta (Sanskrit), Kiryato (Gujarati), Mahatita (Hindi), Kalmegh (Bengali)<sup>2</sup>. It is also known as ‘Bhui Neem’ as the plant although being very small has similarity in taste and appearance as Neem (*Azadirachta indica*). A wide array of studies has been conducted by researchers, especially in Asia, following reports about the medicinal properties possessed by this plant mostly according to traditional medical practitioners in ayurvedic medical system. Phytochemical studies have revealed that *A. paniculata* contains diverse compounds including diterpenoid lactones, flavonoids and miscellaneous compounds. It has been shown to possess wide spectrum of pharmacological properties<sup>3,4</sup>.



**Figure 1:** *A. Paniculata* leaves



**Figure 2:** *A. Paniculata* flower

### **Traditional Medicinal Uses**

The aerial parts, roots and whole plant of *A. paniculata* have been used for centuries in Asia as traditional medicine for the treatment of various ailments. It has been used by traditional medical practitioners for stomachaches, inflammation, pyrexia, and intermittent fevers<sup>5-8</sup>. The whole plant has been used for several applications such as antidote for snake-bite and poisonous stings of some insects, and to treat dyspepsia, influenza, dysentery, malaria and respiratory infections<sup>5,6</sup>. The leaf

extract is a traditional remedy for the treatment of infectious disease, fever causing diseases, colic pain, loss of appetite, irregular stools and diarrhea<sup>9</sup>. In Malaysia, a decoction of the aerial parts is used to treat common cold, hypertension, diabetes, cancer, malaria and snake bite<sup>10</sup>. It is an important constituent of at least 26 Ayurvedic formulas in Indian pharmacopoeia. In traditional Chinese medicine, it is seen as the cold-property herb used to rid the body of heat and fever and to dispel toxins from the body<sup>11</sup>. In Ayurvedic medicinal system, tribals of Tamilnadu ( India use this herb for a variety of ailments like dysmenorrhoea, leucorrhoea, pre-natal and post-natal care, complicated diseases such malaria, jaundice, gonorrhoea and general ailments like wounds, cuts, boils and skin diseases<sup>12-14</sup>.

*A. paniculata* is being widely used by the Local Traditional Healers of Chhattisgarh also mainly in the treatment of Cough & Cold, Fever, Jaundice and Diabetes apart from other ailment treatments. The plant is generally used in combination with some other medicinal plants or their parts as a compound drug<sup>15</sup>.

### **Phytochemistry**

*A. paniculata* has various compounds in its aerial parts and roots and these are often used in extracting its active principles. Diverse factors such as geographical region, harvest time and processing method account for the variability in its chemical content. Phytochemical studies of *A. paniculata* has led to the isolation of various plant metabolites. Notable among these metabolites are the terpenoids (entolabdane diterpene lactones) which account for a large proportion of its components and therapeutic activities<sup>16-18</sup>. Other categories of compounds that have also been isolated include flavonoids (flavones), noriridoids, xanthenes, polyphenols and trace and macro elements<sup>19</sup>.

### **Terpenoids**

Diterpenoid lactones are the commonest terpenoid compounds isolated from *A. paniculata* (Table 1). Diterpenoids are distributed in and have been isolated from the aerial parts and roots of this plant. of the diterpenoids that have been identified and isolated from *A. paniculata*, andrographolide is the most prominent in occurrence and quantity. Andrographolide has a very bitter taste, and it is colourless and crystalline in appearance<sup>16</sup> and was first isolated in pure form by Gorter in 1911. Dominant diterpenoids other than andrographolide which have been isolated mostly from the aerial parts of *A. paniculata* include deoxyandrographolide and neoandrographolide. These diterpenoids (Table 1) have been isolated by several workers<sup>17-18</sup>.

### **Flavonoids**

Flavones are the major flavonoids that have been isolated from the aerial parts, roots and whole plant of *A. paniculata* (Table 1)<sup>19,20</sup>.

### **Other compounds**

Several other compounds (Table 1) have also been isolated, especially, from the roots of *A. paniculata*. Four xanthenes were isolated from the roots identified as 1, 8 -dihydroxy- 3, 7 -dimethoxy-xanthone, 4, 8 -dihydroxy- 2, 7 -dimethoxy-xanthone, 1, 2 -dihydroxy- 6, 8 -dimethoxy- xanthone and 3, 7, 8 -trimethoxy- 1 -hydroxy-xanthone<sup>21</sup>. Five rare noriridoids designated as andrographolide A - E, along with curvifloruside were isolated from the roots of *A. paniculata*<sup>22</sup>.

SN.	Compound	Plant part from where isolated	Compound type
1.	Andrographolide	Leaves/aerial	Diterpenoid lactone
2.	Neoandrographolide	Leaves/aerial	Diterpenoid lactone
3.	14-deoxyandrographolide	Aerial parts	Diterpenoid lactone
4.	Andrographoside	Leaves/aerial parts	Diterpene
5.	14-deoxy-11, 12-didehydroandrographolide	Aerial parts	Diterpenoid lactone
6.	5, 7, 2', 3'-tetramethoxyflavone	Whole plant	Flavonone
7.	5-hydroxy-7, 2', 3'-trimethoxy flavones	Whole plant	Flavone
8.	6'trimethoxyflavone	Root	Flavone
9.	5-hydroxy-7, 2', 7-O-methylidihydrowogonin	Root/aerial part	Flavone
10.	7-O-methylwogonin	Root/aerial part/whole plant	Flavone
11.	Flavone-1, 2'methylether	Root/aerial part/whole plant	Flavone
12.	7-O-methylwogonin-5-glucoside	Root/aerial parts	Flavones
13.	Flavone-1, 2'-O-glucoside	Root /aerial part/whole plant	Flavonoids
14.	5-hydroxy-7, 8, 2', 5'-tetramethoxyflavone	Whole plant	Flavonoids

### Pharmacology

The use of the different parts of *A. paniculata* plant in folk medicine, especially, in Asia led scientists to study its pharmacological properties to explore the use of this plant as a therapeutic agent in the remedy of various ailments. Several studies showed that this plant exhibited various biological activities such as anti-microbial, cytotoxicity, anti-protozoan, anti-inflammatory, anti-oxidant, immunostimulant, anti-diabetic, anti-infective, anti-angiogenic, hepato-protective, sex hormone modulatory, liver enzymes modulatory and insecticidal and toxicity activities and cardiovascular therapy.

### Anti-microbial activity

Aqueous extract, andrographolides and arabinogalactan proteins isolated from the dried herb of *A. paniculata* were screened for anti-microbial activity. The result showed that the aqueous extract and arabinogalactan proteins have antibacterial activity against *Bacillus subtilis* (*B. subtilis*), *Escherichia coli* (*E. coli*), *Pseudomonas aeruginosa* while andrographolide was only active against *B. subtilis*. All three were also reported to possess anti-fungal activity against *Candida albicans*<sup>23</sup>. Five rare noriridoides, andrographidoides A - E were screened for anti-bacterial activity against *E. coli*,

Staphylococcus aureus, Staphylococcus epidermidis, Pseudomonas aeruginosa and *B. subtilis*. None of the compounds showed any inhibitory activity (MIC>100 µg/mL). Gentamycin, chloramphenicol and Ciprofloxacin were used as positive controls<sup>22</sup>.

#### **Anti-inflammatory/anti-allergic activity**

The *A. paniculata* leaves combined aqueous and methanol extract showed significant improvement of lipopolysaccharide inducer release of pro-inflammatory mediators (NO, IL-1β and IL-6), inflammatory mediators (PGE 2 and TXB2) along with allergic mediators (LTB 4)<sup>24</sup>. Different photochemicals: andrographolide, neoandrographolide, isoandrographolide, andrograpanin, 7-O-methylwogonin, skullcapflavone and 14 -deoxy- 11, 12 - didehydroandrographolide isolated from *A. paniculata* leaves were screened for *in vitro* anti-inflammatory and anti-allergic potential. It was observed that andrographolide, isoandrographolide, 7-O-methylwogonin and skullcapflavone-1 significantly inhibited inflammatory mediators NO and PGE2 release from lipopolysaccharide (LPS) stimulated cultured macrophages. Andrographolide, isoandrographolide and 7- O-methylwogonin inhibited the production of IL-1β in LPS stimulated macrophages. In addition IL - 6 production from LPS induced macrophages was significantly (P<0.01) inhibited by andrographolide, isoandrographolide and skullcapflavone-1 in a concentration dependent manner. The results also showed that andrographolide, isoandrographolide and skullcapflavone-1 significantly suppressed TXB 4 released in A 23187 activated HL - 60 promyelocytic leukemia cells. Andrographolide, dehydroandrographolide and neoandrographolide isolated from the aerial parts of *A. paniculata* exhibited anti-inflammatory effects by interfering with COX enzyme activity. Remarkable inhibition of COX-1 by andrographolide (30.1 µM) and dehydroandrographolide (28.5 µmol/ L) was found in ionophore A 23187–induced human platelets. Dehydroandrographolide (28.5 µmol/ L ) and neoandrographolide (20.8 µmol/ L) strongly suppressed the LPS -stimulated COX - 2 activity in human blood and dehydroandrographolide modulated the level of LPS -induced TNF -α, IL - 6, IL - 1β, and IL - 10 secretion in human blood in a concentration dependent manner. It indicates that dehydroandrographolide has the highest efficacy<sup>25</sup>. Andrograpanin (15 - 90 µmol/L) isolated from the ethanol extract of the leaves inhibited NO and pro-inflammatory cytokines in a dose dependent manner from lipopolysaccharide activated macrophages. Significant (p< 0.05) inhibition of NO was evident at a concentration of 30 µmol/L and at a concentration of 75 µmol/L. Andrograpanin almost completely inhibited NO production. Significant inhibition of proinflammatory cytokines was evident at a concentration of 1.5µmol/L and there was an almost complete inhibition at a concentration of 90 µmol/ L<sup>25</sup>.

In another study it was found that andrographolide has shown to have a strong anti-inflammatory activity. LPS-induced TNF-α and GM-CSF release from mouse peritoneal macrophages was inhibited by andrographolide in a concentration-dependent manner. The concentration of the drug producing 50% inhibition was 0.6 µM for TNF-α and 3.3 µM for GM-CSF. The maximal inhibition achieved (at 50 µM) was 77% and 94%, respectively, for the two cytokines. In the *in vivo* study, intra-peritoneal treatment of ovalbumin-immunized and nasally-challenged mice with andrographolide significantly inhibited the elevation of bronchoalveolar fluid (BAF) levels of TNF-α and GM-CSF in a dose-dependent manner, with 30 mg/kg produced an inhibition of 92% and 65% of the cytokines and almost completely abolished the accumulation of lymphocytes and eosinophils<sup>26</sup>.

The anti-allergic properties of the phytoconstituents was investigated on A23187 induced LTB4 production. The result showed 30.5% and 19.6% inhibition of LTB 4 production in A 23187 induced HL - 60 promyelocytic leukemia cells at concentrations of 63 µmol/L and 33.5 µmol/L for

skullcapflavone and 7-O-methylwogonin respectively. The IC 50 value for the reference standard captopril was 48  $\mu\text{mol/L}$ . 7-O-methylwogonin was the only phytoconstituent that potently inhibited A23187 induced histamine release in RBL-2H3 rat basophil leukemic cells in a dose dependent manner<sup>27</sup>.

In another study diterpenes, andrographolide and neoandrographolide, isolated from the plant, were evaluated for antiallergic activity. These were tested for anti-PCA (Passive cutaneous anaphylaxis) and mast cell stabilizing activities in rats have shown significant anti-PCA activity<sup>28</sup>.

### Cytotoxicity

The methanol extract, petroleum ether, dichloromethane fraction and aqueous fraction of the methanol extract of *A. paniculata* were screened for anti-proliferation activity against HT-29 (colon cancer) cells. The methanol extract inhibited the proliferation of HT-29 cells by 50% at a concentration of 10  $\mu\text{g/mL}$ . The petroleum ether and dichloromethane fractions inhibited proliferation of HT-29 cells with a GI50 value of 46  $\mu\text{g/mL}$  and 10  $\mu\text{g/mL}$  respectively. The aqueous extract did not inhibit the proliferation of HT-29 cells. The diterpenes andrographolide isolated from the dichloromethane fraction, inhibited the proliferation of all cancer cells screened. 14-deoxy-andrographolide showed moderate inhibition against the proliferation of two cancer cell out of the entire cell screened. 14-deoxy-11, 12-didehydroandrographolide did not inhibit the proliferation of any of the cancer cell line tested<sup>27</sup>. These findings are in accordance with earlier reports, showing the cytotoxic activity of andrographolide against human epidermoid carcinoma and lymphocytic leukaemia cells<sup>18</sup>. The growth inhibitory activity on mouse myeloid leukemia cells by the methanol extract of the aerial parts of *A. paniculata* and some of the isolated compounds has also been reported<sup>29</sup>. Andrographolide and its semi-synthetic analogues namely 3, 19-isopropylideneandrographolide, 14-acetyl-3, 19-isopropylideneandrographolide and 14-acetylandrographolide were screened *in vitro* for anti-tumor activity against MCF-7 human breast cancer and HCT-116 colon cancer cell lines. 19-isopropylideneandrographolide and 14-acetylandrographolide showed cytotoxic activity against the two cell lines tested and they were equally potent when compared to parent andrographolide. In a similar study at the national cancer institute in the USA, 19-isopropylideneandrographolide and 14-acetylandrographolide were also screened and found to be cytotoxic against 60 human cancer cell lines<sup>30</sup>. Xanthenes isolated from the chloroform fraction of the roots were screened for cytotoxicity and the results showed that all the compounds have IC 50 values  $>16 \mu\text{g/mL}$ , which can be considered as non-cytotoxic as per WHO criteria<sup>22</sup>.

### Antidiabetic effect:

*A. paniculata* has also been studied for its possible impact on the management of Diabetes Mellitus in humans<sup>31-34</sup>. Andrographolide is a potential bioactive phytochemical in *A. paniculata*, which possesses antidiabetic properties<sup>31, 35-39</sup>. Oral andrographolide and *A. paniculata* lowered the blood glucose levels of streptozotocin-induced diabetic rats in a dose-dependent manner<sup>40</sup>. Furthermore, *A. paniculata* extract was reported to reduce hyperglycemia by inhibiting  $\beta$ -cell dysfunction in alloxan-induced diabetic rats<sup>41</sup>. An ethanolic extract of *A. paniculata* and andrographolide lowered the plasma glucose levels by enhancing the translocation of glucose-transporter-4 in insulin-resistant obese mice<sup>42, 43</sup>.

### Effects on Cardiovascular Disease

An increase of blood-clotting time has been demonstrated by *A. paniculata*; thus making it useful for the pre- and post-treatments by extract of *A. paniculata* after surgery which prevent the contraction of blood vessels, hence decreasing the risk of the consequent closing of blood vessels after angioplasty operations<sup>44</sup>. Several studies have used animal models to study the effects of aqueous extracts and active constituents of *A.*

*paniculata*, both before and after experimental myocardial infarction. An extract of the plant produced antihypertensive effects because it relaxed smooth muscles in the walls of blood vessels and prevented the blood vessels from constricting and limiting blood flow to the brain, heart, and other organs<sup>45</sup>. It was demonstrated that the dichloromethane extract of *A. paniculata* significantly reduced coronary perfusion pressure by up to  $24.5 \pm 3.0$  mmHg at a 3 mg dose and also reduced the heart rate by up to  $49.5 \pm 11.4$  beats/min at this dose<sup>46</sup>. *A. paniculata* was also found to reduce the arterial constriction caused by high cholesterol in the diet and by injury to the inner lining of the blood vessel<sup>47</sup>. The pretreatment of andrographolide; led to a time-dependent protection of rat cardiomyocytes against hypoxia injury which was reported to be caused by this effect was reported to be linked with regulation of cellular reduced glutathione (GSH) level and antioxidant enzyme activities<sup>48</sup>. Dogs administered with *A. paniculata* one hour after the development of myocardial infarction were reported to have decreased damage of the heart muscle<sup>49</sup>. These findings infer the promising use of *A. paniculata* as a favorable agent for cardiovascular therapy.

### Hepatoprotective activity:

Among the phytochemicals obtained from the plant, andrographolide the main constituent of *A. paniculata* was found to be effective in preventing carbon tetrachloride induced liver damage (Rats and mice). Andrographolide has also exhibited significant hepatoprotective effect against different types of liver damages induced by paracetamol or galactamine,<sup>50</sup> and had a higher capacity than a classical antioxidant silymarin in preventing a decrease of bile production induced by paracetamol.<sup>51</sup> The paracetamol induced enhanced levels of serum marker enzymes such as serum glutamate pyruvate transaminase (GPT), serum glutamate oxaloacetate transaminase (GOT), alkaline phosphatase (ALP), and bilirubin in peripheral blood serum and distorted hepatic tissue architecture along with increased levels of lipid peroxides (LPO) and reduction of superoxide dismutase (SOD), catalase, reduced glutathione (GSH) and glutathione peroxidase (GPx) in liver tissue. Administration of the plant extracts after paracetamol insult restored the levels of these parameters to control (untreated) levels.<sup>52</sup>

It also played a great hepatoprotective role by reducing a lipid peroxidation product malondialdehyde (MDA), in which the lowering of MDA formation conveyed the free radical scavenging property of diterpene lactones of andrographolide, as well as by maintaining high level of glutathione, glutamic pyruvate transaminase, and alkaline phosphatase in carbon tetrachloride or tert-butylhydroperoxide treated mice.<sup>53</sup> Antihepatotoxic action of andrographolide was reported against *Plasmodium berghei* K173-induced hepatic damage of *mastomys natalensis*.<sup>54</sup> Andrographolide was found to play an important role as effective stimulator of gall bladder function by producing a significant increase in bile flow, bile salt and bile acid in conscious rats and anesthetized guinea pigs. There was distinct improvement in the majority of infective hepatitis patient after continuous treatment with *A. paniculata* in regular recovery from jaundice, improvement of appetite and liver function tests, as well as subsidence of fever.<sup>55</sup>

**Conclusion:** *A. paniculata* has been an important herb for treatment of various ailments in the traditional medicinal system. Plant extract of the plant contains various phytochemicals exhibiting promising pharmacological effects a few of them are discussed here.

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