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### To the study of Bio-activity of Herbal Medicines

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**Abstract:** Research into less harmful, more environmentally friendly alternatives is urgently needed due to the fact that the careless application of synthetic pesticides has led to serious health risks, ecological imbalances, and insect resistance. To find a long-term, sustainable, and ecologically friendly substitute for conventional pesticides, this research looks at the insecticidal properties of bioactive chemicals extracted from herbs. The wide variety of chemicals found in herbal bio-actives allows them to effectively combat a wide range of pests with little impact on ecosystems and non-target creatures.

Keywords: sustainable, ecologically, friendly, conventional, pesticides, herbal

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#### INTRODUCTION

Phytotherapy, herbalism, or herbal medicine is the practice of using compounds derived from plants for medicinal reasons. The ancients did this because they respected plants for what they could do for health. Evidence of herbal medicine's historical importance may be seen within conventional medicine's (TCM, Ayurveda, etc.) Native American medicines.

A vital part of basic healthcare has always been medicinal herbs. It is estimated that about Eighty percent of the world's population uses herbal remedies because of their supposed health advantages. In 2022, the worldwide the projected value of the herbal medicine market was \$148.5 billion experts predict it will reach \$386.07 billion by 2032, up from \$165.13 billion in 2023. In the predicted time frame from 2023–2032, this trajectory shows a CAGR of 11.20%. An unprecedented and extraordinary global COVID-19 epidemic has broken records worldwide, driving up demand for herbal medication to levels not seen before the outbreak. As the benefits of herbal medicines have been better understood, along with the risks associated with allopathic pharmaceuticals, the demand for herbal medical items has increased. Another market factor driving growth is the increasing incidence of chronic illnesses and the growing population.

An essential aspect the use of traditional herbal therapy in human treatment has long been recognised and respected practices and culture for thousands of years. A wide variety of therapeutic procedures are included into it. have been handed down through the ages and is based on ancient knowledge. But nowadays, scientific research is putting more and more pressure on herbal medicines to prove their efficacy. The need to unite traditional medical practices with modern scientific understanding is underscored by the ever-changing relationship between the two bodies of knowledge.

The history of traditional herbal therapy is vast and varied, spanning many different civilizations and countries. Herbs and botanicals have a rich medicinal history that is echoed throughout several prehistoric societies. These methods often relied on first-hand experience, learning from mistakes, and the

accumulation of knowledge conveyed via oral or written traditions. Traditional medicine (CM), Native American herbalism, and India's Ayurveda all see herbs as part of a more complete whole, one that takes into account our bodily, psychological, and spiritual lives are all intricately related. The significance of preserving internal peace and the interdependence of all parts of health are highlighted by this holistic approach.

#### LITERATURE REVIEW

**Saira Khan et.al (2017)** The toxic effects of seven plant extracts were evaluated against the four major agricultural pest insects such as fruit flies, red flour beetles, armyworms, and pea aphids of the order Coleoptera (Hemiptera)—each of which represents a distinct order of insects. These extracts came from the following plant families: Lauralee, Eucalyptus sideroxylon, Rutaceae, Calotropis procera, Thymelaeaceae, and Asteraceae. The focus of this study was insecticides made from plants. Aphids, the most vulnerable insects, were killed by all of the plant extracts in a day of testing. Aphids were most severely affected by extracts from I. rugosus and D. mucronata, according to further bioassays that used lower concentrations of the plant extracts. These highly effective plant extracts were tested for their insecticidal efficiency after additional separation into various solvent fractions based on polarity.

**Suneel Kumar Singh et.al (2019)** Farmers throughout the world are using more and more agro-inputs every day. However, output, its due to the fact that agricultural expenditures are growing at a faster rate than increases in both quality and production, contemporary agriculture is rapidly losing its viability. The ecosystem and environment are being irreparably harmed by synthetic fertilizers and pesticides, and agriculture cannot continue to rely on their usage.

**Ravi Kant Upadhyay (2016)** Plants and many insect and pathogen groups have a long history together. Since the beginning of time, plants have evolved chemotypic and genotypic variances and adaptations in response to biological infections and infestations as well as changes in climate. In order to shield plants against herbivorous insects, it altered and enhanced their genomes to produce potent compounds that are poisonous, repulsive, or hinder the development of insects. Despite having some insect-specificity, the majority of these compounds are found in both host and non-host plants. Plants are mostly known for their anti-feedant or repellent compounds, which provide a defense function by discouraging insect pests from their hosts. These compounds produced from plants work at the cellular level by generating molecular interactions or reactions that cause organogenesis to default, resulting in the creation of deformed pupae with poor texture, weight loss, and structural abnormalities.

**Tamer Üstüner (2018)** GC and GC-MS were used in this work to analyse the chemical make-composed of Eucalyptus camaldulensis Dehnh's volatile oil. The sample under investigation had the following compositions: oxygenated sesquiterpenes, monoterpene hydrocarbons, and oxygenated monoterpenes. The oil's ability to kill the following storage insect pests were looked at: Sitophilus granaries L. (Col.: Curculionidae), Rhizopertha dominica F. (Col.: Bostrychidae), and Acanthoscelides obtectus Say., Tribolium confusum Duv., and Callosobruchus maculatus F. are all members of the Bruchidae family of corals. Additionally, our findings demonstrated that essential oil had a substantial impact on the development of weed stems and roots as well as fungal mycelial proliferation. The mycelial growth of In only seven days, S. sclerotiorum, F. oxyporum, P. debaryanum, and V. dahliae were all numbed down in

10 and 20  $\mu$ L, however R. solani showed no impact during this time. However, the effects of applying the oil to the weeds varied depending on the species under investigation. While the root and stem development of C. arvensis was unaffected by E. camaldulensis essential oil at concentrations of 5, 10, and 20  $\mu$ L, it did reduce the development of A. retroflexus and M. officinalis simultaneously concentration and time. Scientific evidence suggests that E. camaldulensis essential oil could have use as a natural fungicide and insecticide.

**Barro's dos Santos, Eliza de Jesus, et al. (2023)** Ingredients in essential oils made from plants in the Myrcia genus and their uses are thoroughly examined in this research. The combined findings demonstrate the biological activity and chemical variety oils, drawing attention to their possible importance for many medical and industrial applications. In addition to a variety of bioactive compounds, Yeasts and a variety of bacteria, both Gram-positive and Gram-negative, are included in the essential oils of Myrcia. many microorganisms that monoterpenes and sesquiterpenes have antibacterial properties against. Additionally, this research emphasizes these oils' phytotoxic action, suggesting that they may be used to manage weeds. The findings further demonstrate the effectiveness of Myrcia essential oils as a substitute for synthetic pesticides by demonstrating their insecticidal capabilities against a variety of pests. Furthermore, Myrcia species have shown encouraging hypoglycemic effects, indicating that they may be used to treat diabetes.

### INVESTIGATING THE POSSIBILITY OF USING A CRUDE EXTRACT OF ANDROGRAPHIS PANICULATA AS A BIO-PESTICIDE IN FARMING

Synthetic pesticides are employed in today's farming system to control weeds, insects, and illnesses. Around two million metric tonnes of pesticides are used every year on a global scale. Nevertheless, the consumption of pesticides and insecticides is expected to reach an estimated 4.11 million kilograms and 601 kilograms per hectare of agricultural by 2030, with a steady annual increase. The FAOSTAT database contains information pertaining to food and agriculture compiled by the UN Food and Agriculture Organisation. (Join, 2017). Although the traditional technique is advantageous from a crop protection and yield production standpoint, there is a major concern posed by excessive usage owing bio-magnification, bio-accumulation, and a very long half-life in nature. Consequently, bio-pesticides should be supported in order to reduce pesticide and insecticide burdens from the environment. Bio- Chemicals that are used to manage pests are known as pesticides and are produced from natural sources. These materials might be plants, animals, microorganisms, or minerals.

Insects, diseases, and weeds are just a few of the agricultural problems that these biological control agents may help manage. Biopesticides are an alternative to traditional chemical pesticides that are safer for people and other non-target creatures, easier on the environment, and more effective in eliminating the target pest. A variety of bio-pesticides are available, such as: [i] Pesticides produced by microbes: These include microbes that infect or parasitize the intended pests, protozoa, as well as fungi, viruses, and bacteria. As an example, a bacterial bio-pesticide called Bacillus thuringiensis (Bt) is often used to combat certain insect pests. (ii) To make them more resistant to pests, plants may create chemicals called plant-incorporated pesticides (PIPs). One such Case in point: Bt crops, which use plants endowed with the Bt toxin to protect themselves from harmful insects. (iv) Biochemical pesticides: These are compounds with pesticidal characteristics that exist naturally; they are often extracts from plants or animals. Neem oil, for

instance, has a yearly pesticide consumption rate that is comparable to that of many Asian nations (Table 1).

S. No.	Country	Pesticides used in tonnes	Country Area (km <sup>2</sup> )
1.	China	1,807,000	9,615,222
2.	India	56,120	3,287,263
3.	Malaysia	49,199	330,803
4.	Pakistan	27,885	881,913
5.	Thailand	21,800	513,120
6.	South Korea	19,788	100,210
7.	Vietnam	19,154	331,212
8.	Bangladesh	15,833	147,570
9.	Myanmar	5583	676,578
10.	Nepal	454	147,181
11.	Bhutan	12	38,394

Table 1 The yearly pesticide use in several Asian nations is shown

<sup>a</sup> Source: Sharma et al#,2019

qualities that kill insects. Bio-pesticides are a greener and more long-term viable option than conventional pesticides. They are an essential part of IPM tactics, which aim in an effort to lessen the reliance on synthetic pesticides and associated negative effects on ecosystems.

In ancient Ayurvedic texts, it is mentioned as kalmegh (meaning "dark cloud"), kalmegha (meaning "neem of the ground"), and "Indian echi-nacea." There are other names for it as well. 'Mahatita,' meaning king of bitter, is a popular name for the plant in northeastern India. The common names of kalmegh vary across Indian languages. For example, in Hindi it is called kirayat, in Sanskrit it is called bhunimba, in Bengali it is called kalmegh, in Telugu it is called naelavemu, in Tamil it is called nilavaembu, in Malay it is called hempendu bhumi, in Punjabi In Marathi, it is known as chooraita, whereas in Gujarati it is called kariyatu. oli-kiryata.

#### Uses in ethnobotany

One well-respected homoeopathic remedy is Kalmegh. Kalmegh is mentioned often in the ancient Ayurvedic texts. It is the most significant plant for treating a wide range of illnesses due to its adaptable pharmacological characteristics. When children in India get stomachaches, a common home remedy called

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"Alui" is given to them. This remedy is prepared from fresh kalmegh leaves and is popular in the Bengal area. Babies who experience fussiness, loss of appetite, or irregular bowel movements may find relief by drinking an infusion or drinking the juice of the leaves. Symptomatic relief from fever, gaseous distension-induced dyspepsia, and severe diarrhea are all possible with the use of the leaves and roots. When it comes to treating chronic hepatitis, the Unani medical system recommends it. The Indian Pharmacopoeia states that it is the active component in around 26 Ayurvedic medicines.

#### Synthesis and phytoconstituents

Andrographolide (C20H30O5) is a bitter, water-soluble diterpene lactone first found in Andrographis paniculata leaves. by Gorter in 1911. Andrographolide and neo-andrographolide were identified as diterpenes in subsequent chemical investigations. There was also the discovery of a novel flavone 2'-glucoside in A. alata and A. paniculata. Each part of the plant contains some amount of diterpenes, lactones, flavonoids, alkanes, ketones, or aldehydes. Diterpenes, aldehydes, ketones, and alkanes are abundant in the plant's aerial sections. Among the active compounds found in the leaves are lactones, flavonoids, and diterpenes; the first two classes of flavonoids are mostly accessible in the root section, while the latter two classes of lactones are found in the leaves as well.

The lactone andrographolide known as "Kalmeghin" is responsible for the bitter taste of the leaves. Both 3.4 and 3.5 include information on A. paniculata's chemical components and phytoconstituents. The leaves of the kalmegh plant contain the diterpene glucoside known as deoxy andrographolide 19  $\beta$ -D glucosides. found 12 compounds in the plant's aerial portions, including 6 ent-labdane type 2 diterpenoids, 4 dimers of diterpenes (glucosides), and A, B, C, and D bis-andrographolides. An ethanolic extract yielded three new ent-labdane diterpenoids: 19-norandrographolides A, B, and C. The 55 ent-labdane diterpenoids found in the aerial portions were identified using 1D and 2D NMR spectroscopy. These compounds included andrographic acid, three-O- $\gamma$ -D-glucosyl-14-deoxyandrographiside, and three-O- $\beta$ -D-glucosyl-14-deoxy-11,12-didehydroandrographiside. Extracted in 2004 by Koteswara et al., these flavonoids have the following functional groups: 5,7,2',3'-tetra methoxy and 5, hydroxy-7,2',3'-trimethoxy. In addition to the thirty flavonoids, twelve other compounds were identified, including two flavonoid glycosides and five hydroxy-7,8,2',5'-tetra methoxy. The 5-hydroxy-7,8-dimethoxy (2 R) derivatives of flavanone are also manufactured.5-flavone-5-O- $\beta$ -D-glucopyranoside and - $\beta$ -D-glucopyranoside.

#### The possibility of using ginger extracts (Zingiber officinale Rosc.) as a biological insecticide

In addition to its common culinary application, ginger contains medicinal properties that help with nausea, vomiting, flatulence, vomiting, and pain. Aphrodisiacs, sex stimulants, respiratory infections, gastrointestinal tract disorders, and blood purification are all greatly improved by crude ginger rhizome extract. In addition to regulating apoptosis, inhibiting vascular endothelial growth factors, and reducing inflammatory responses, the bioactive components of ginger have antioxidant characteristics.

The volatile oils included in ginger, which range from 1% to 3%, give it a strong aroma. These oils include sesquiterpene, zingerone, kaempferol, and zingiberene. A large body of evidence suggests that ginger's crude extract has insecticidal effects.

Numerous ginger types have emerged as a result of natural hybridization and thoughtless assortment of

ginger clones. The output of rhizomes, dry weight, quantity of extractive, fibre, and volatile oils might vary greatly across Indian species. The many ginger crop types in our nation may be attributed mainly to agroclimatic circumstances and spontaneous mutations via natural selection.

Bioactive	Phenolic	Terpenes	Others	Ref.
components				
	Gingerol (6, 8, 10-	β-bisabolene, α-	Polysaccharides,	
Fresh	gingerol), paradols,	farnesene,	raw fibers, lipids,	[20]
ginger	shogaol, 6-	zingiberene, α-	and organic acids	
	dehydrogingerdione	curcumene, and β-		
	quercetin, zingerone, and	sesquiphellandrene		
	gingerenone-A,			
Dried	Shogaol (6,8,10-shogaol)			[21]
ginger	and gingerol			
		ingiberene, $\beta$ -	amino acids,	
Ginger	gingerol,paradols, and	sesquiphellandrene,	protein, raw fiber,	[12]
powder	shogaol	$\beta$ -bisabolene, $\alpha$ -	ash, vitamins, and	
		curcumene, and a-	phytosterols,	
		farnesene,		

Table 2: Ginger and its bioactive components in various forms

#### **Discussion and results**

#### Identification of compounds (1-4)

A light-yellow solid that showed a favorable reaction with Dragendorff's reagent was identified as compound 1. It was shown using 1H NMR data that a benzene ring that had been monosubstituted was found the hydrogen bond lengths at  $\delta$ H 7.45 (dd, J = 7.8, 1.2 Hz, 2H-2', 6'), 7.36 (t, J = 7.8 Hz, 2H-3', 5'), and 7.19 (t, J = 7.8 Hz, H-4').

In addition, at  $\delta$ H 7.82 (s, H-4), there is a proton located downstream of the pyrazole ring. Moreover, three separate aliphatic methylene groups were identified at 2.69 (m, 2H-6), 3.10 (t, J = 7.5 Hz, 2H-7), and 4.19 (t,a J = 7.5 Hz, 2H-5). These results confirmed those that have been associated with withasomine before. The second compound was found to have a sheet crystal structure and a melting point ranging from 65 to 67 degrees Celsius. Analyses compared to the individual data further supported the identification of methyl isoferulate 2 as indicated by the aforementioned data.

The third component, the results of Dragendorff's analysis showed that the amorphous powder, was isolated. The signals for 28 carbon atoms were shown by the 13C NMR on top of heteronuclear single quantum coherence (HSQC) spectra. These atoms included 4 methyl groups, 9 methylene groups, 8 At  $\delta$ C 166.3 ppm, there is one carbonyl group, seven quaternary carbon atoms, and methine groups. The HRESIMS spectra of Molecular 3 confirmed the molecular formula as C28H43O6 by showing a [M +H] + peak at m/z 475.3199. In the infrared investigation, a six-membered lactone ring that is unsaturated was found between 1696 and 1601 cm–1. Four proton singlets at  $\delta$ H 0.86, 1.02, 1.28, and 2.06 ppm were observed in the 1H NMR spectra of compound 3, corresponding to quaternary methyl groups, for the C-18, C-19, C-21, and C-28 protons, respectively. On C-27, the methylenic protons are responsible for the 4.39 ppm and 4.33 ppm signals (J = 12.5 Hz), respectively. A signal at  $\delta$ H 4.28 (dd, J = 13.3, 3.3 Hz) was picked up when the C-22 proton was connected to the lactone group. The aforementioned findings

provided further evidence that a lactone substituent was present on the tetracyclic steroidal skeleton.



Figure 1: The molecular blueprint of chemicals (1-4)

At 3.97 (m, H-3) and 3.83 (t, W1/2 = 5.0 Hz, H-1), two oxygenated methine protons associated with C-1 and C-3, respectively, were detected. The stereochemistry of the C-l ( $\alpha$ ) and C-3 ( $\beta$ ) hydroxyl groups could be determined by comparing the chemical shifts and coupling constant values to those of the previously recorded withanolide 1 $\alpha$ , 3 $\beta$ , 20-trihydroxy (20R, 22R)-witha-5,24-dienolide. Plus, the proton of C-6 was assigned the down-field proton signal at  $\delta$ H 5.57 (d, J = 5.0 Hz). With the use of the H-H COSY spectrum, we were able to discern the cross peaks between (H-22, 4.28) and (H-23, 2.53), and between (H-3, 3.97) and (H-2, 1.73 and 2H-4, 2.37, 2.30). The relationship between 2H-27 at  $\delta$ H 4.33 and 4.39 ppm and C-24, C-25, and C-26 at  $\delta$ C 153.3, 125.8, and 166.3 ppm was validated by the heteronuclear multiple bond association (HMBC) spectra. Additionally, the proton was connected to C-5 at 137.6 ppm and C-3 at  $\delta$ H 3.83 (H-1) at  $\delta$ C 66.5 ppm. The HMBC analysis further confirmed places where the four methyl groups are located. Based on the information given, Sominone A. was assigned structure (3), which is ((20R,22R)-1 $\alpha$ ,3 $\beta$ ,20,27)."- tetrahydroxywitha-5,24-dienolide".

Analyzed using Dragendorff's method, Compound 4 was found to be an amorphous powder with an orange hue. The existence of 34 carbon atom signals was confirmed by 13C NMR and HSQC investigations. These signals belong to contains there is a carbonyl group at 168.9 ppm, five methyl groups, nine methylene groups, thirteen methine groups, and seven quaternary carbon atoms. Compound 4 has a structural similarity to compound 3, according to the 1H NMR investigation. The distinctive feature of compound 4 is its glucose moiety, which is linked to the withanolide's C-3  $\beta$ -OH at  $\delta$ H 4.05 (m, H-3). Furthermore, molecule 4 does not have an oxygenated methylene group at  $\delta$ H 1.85 (s, 3H-27), but rather a methyl group singlet signal. According to the HMBC spectra, The C-3 of withanolide was connected to the anomeric proton of glucose at 74.9 ppm at  $\delta$ H 4.19 (d, J = 7.8 Hz, H-1'). The structure of Coagulin Q aligned with the previously reported findings. Compounds 2, 3, and 4 did not include any alkaloids, but they did contain compounds that showed  $\alpha$ ,  $\beta$ -unsaturated ester characteristics, especially withanolide derivative alkaloids, and they worked well with Dragendorff's reagent.

We evaluated and compared the effects of Azadirachtin (Okios 3.2% EC), parts of W. somniferous that are alkaloid, as well as parts of T. urticae, N. viridula, B. tabaci, and A. craccivora (2). The alkaloid fraction and its constituents clearly have a harmful impact on the four pests in varying proportions. Table 3 shows that out of all the compounds tested, withasomine 1 had the highest LC50 value (15.44 ppm) against A. craccivora. This molecule outperformed the alkaloid fraction and the others. With LC50 values of 36.61 and 36.78 ppm against B. tabaci and 85.11 and 98.26 ppm against N. viridula, respectively, withasomine 1 and Azadirachtin proving to be the most effective therapies, as shown in Table 4.

Table 5 shows that withasomine 1, with an LC50 value of 128.29 ppm, was the most hazardous chemical towards T. urticae. Aspartamine 1, azadirachtin, isoferulate 2, and the alkaloid component clearly poison A. craccivora, B. tabaci, and N. viridula, while withanolides, sominone A, and coagulin Q have mild effects. Isoferulate 2 and withas-omine 1 were both effective in treating T. urticae. the most effective options.

The insecticidal effect on Callosobruchus maculatus by use of a hydroethanol extract of Withania adpressa was discovered by Salamatullah (2022). The rose aphid, Macrosiphum rosae, was targeted by an aphicide made from an extract of Willow somnifera in methanol. Spodoptera frugiperda, Spodoptera litura, and Fall armyworm pupae and larvae were all poisoned by W. somnifera root extract. A pesticide investigating the efficacy of Withania cocoana, a member of the same species as In contrast to B. tabaci and Myzus persicae, W. somnifera attacks green peach aphids. The significant function of alkaloids as pesticides was elucidated by Al-Ani et al. Plants may be protected from several pests by using phenolic chemicals, such as ferulic acid.

Treatment	LC50 (ppm)	Confidence limit 95% (ppm)		LC90 (ppm)	Confidence limit 95% (ppm)		Slope ± <u>S.E</u>	Toxicity index (%)at LC50
		Lower	Upper		Lower	Upper		value
Alkaloid fraction	28.97	20.35	39.15	164.32	103.61	378.01	1.70± 0.29	53.30
Withasomine 1	15.44	11.12	20.67	81.48	52.26	178.69	1.77± 0.29	100
Isoferulate 2	55.80	47.42	65.92	140.34	108.30	219.81	3.20± 0.49	28.67
Sominone A 3	137.15	111.43	168.51	431.17	303.87	878.62	2.58± 0.49	11.26
Coagulin Q 4	130.55	107.61	156.25	362.87	271.52	625.54	2.89 ± 0.50	11.83
Azadirachtin (Okios 3.2% EC)	27.40	17.56	45.64	178.28	83.30	2003.89	1.58± 0.44	56.35

### Table 3: The acute toxicity to A. craccivora after 24 hours of exposure to the alkaloid fraction and isolated chemicals from W. somnifera under controlled laboratory settings

## Table 4: The in vitro toxicity of the alkaloid portion of W. somnifera and its separated componentsto B. tabaci after 24 hours

Treatment	LC50 (ppm)	Confi limit (pp	dence 95% om)	ce LC90 Confidence limit (ppm) 95% (ppm)		Slope± <u>S.E</u>	Toxicity index (%)at LC50	
		Lower	Upper		Lower	Upper		value
Alkaloid fraction	47.19	35.82	60.83	189.62	131.93	343.36	2.12 ± 0.31	77.58
Withasomine 1	36.61	26.06	50.61	228.80	135.37	612.10	1.61± 0.28	100
Isoferulate 2	69.93	57.93	84.36	197.02	144.78	356.76	2.85± 0.51	52.35
Sominone A 3	19383	161.06	229.71	508.75	389.73	825.57	3.06± 0.51	18.89
Coagulin Q 4	199.67	164.14	240.54	572.14	421.52	1028.16	2.80 ± 0.50	18.34
Azadirachtin (Okios 3.2% EC)	36.78	27.22	49.93	184.33	118.04	401.99	1.83 ± 0.29	99.54

# Table 5: Analysis of the 3-day in vitro toxicity of the alkaloid fraction and separated chemicals from W. somnifera to N. viridula

Treatment	LC <sub>50</sub> (ppm)	Confidence limit 95% (ppm)		LC <sub>90</sub> (ppm)	Confidence limit 95% (ppm)		Slope±S.E	Toxicity index (%)at LC <sub>50</sub>
		Lower	Upper		Lower	Upper		value
Alkaloid fraction	273.92	225.61	331.22	790.49	577.59	1453.13	2.78±0.50	31.07
Withasomine 1	85.11	63.99	111.29	375.03	252.38	729.61	1.99±0.30	100
Isoferulate 2	203.91	158.05	263.50	792.10	545.72	1456.76	2.17±0.31	41.74
Sominone A 3	399.86	334.73	472.45	1021.10	786.31	1637.26	3.15±0.52	21.28
Coagulin Q 4	440.08	366.68	532.57	1224.52	895.86	2246.73	2.88±0.52	19.34
Azadirachtin (Okios 3.2% EC)	98.26	70.44	132.88	550.67	343.85	1287.89	1.71±0.28	86.62

## Table 6: Analysis of the 3-day in vitro toxicity of the alkaloid portion of W. somnifera and itsseparated components to Trichoderma urticae

Treatment	LC <sub>50</sub> (ppm)	Confidence limit 95% (ppm)		LC <sub>90</sub> (ppm)	Confidence limit 95% (ppm)		Slope±S.E	Toxicity index (%)at LC <sub>50</sub>
		Lower	Upper		Lower	Upper		value
Alkaloid fraction	467.79	356.03	586.11	1784.56	1195.73	4327.56	2.20±0.46	27.42
Withasomine 1	128.29	104.86	154.36	370.47	273.93	659.20	2.78±0.49	100
Isoferulate 2	266.69	222.77	315.88	691.05	529.22	1123.24	3.10±0.52	48.10
Sominone A 3	606.02	487.11	732.91	1836.41	1345.69	3352.52	2.66±0.48	21.17
Coagulin Q 4	641.48	522.85	774.01	1883.55	1383.57	3412.49	2.74±0.49	20.00
Azadirachtin (Okios 3.2% EC)	749.85	392.26	3765.36	21,305.20	4069.86	85,416.21	0.88±0.26	17.11

Treatments	a-esterase	β-esterase	Chitinase	Acetyl	GST activity	Peroxidase
	activity (µg	activity (µg	Activity (µg	choline	(mmol sub-	activity ( $\Delta$
	α-naphthol∕	β-naphthol∕	NAGA /min/g	esterase	conjugated	mO.D./min/mg
			b	activity		
	min/g b wt.)±	min/g b wt.)±	wt.) ± <i>SE</i>	<i>(</i> ug AchBr	/min/mg	protein) ± SE
	SF	SF	,	/min/gm		<b>.</b> ,
	52	52		/mm/gm		
				b wt.) ± SE	protein) ± SE	
Withasomine	$38.83 \pm 6.14$	72.86 ± 5.45m	31.45 ± 3.25*	506.4 ± 3.29**	$2.62 \pm 0.05^{**}$	$251.0 \pm 2.08^*$
1	ns					
Control	$31.13 \pm 3.54$	$64.03 \pm 6.54$	$10.53 \pm 0.67$	$643.8 \pm 3.18$	$1.92 \pm 0.04$	$242.3 \pm 1.45$
T-value	1.09	1.04	6.30	-30.02	0.74	-3.41
Divalue	0.26	0.20	0.02	0.00	0.00	0.04
P-value	0.36	0.38	0.02	0.00	0.00	0.04

SE stands for standard error, and a two-sample If the two differed in ways that were statistically significant means, we utilized a T-test to find them. Given that P < 0.05, then the differences are considered significant; otherwise, they are considered non-significant.

#### CONCLUSION

Herbal medicine is expected to further establish itself as an important part of comprehensive healthcare as more and more details about herbal bioactivity are revealed by continuing research. This will meet the changing demands of people who are looking for more natural and holistic ways to take care of themselves.

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