

ASSESSMENT OF BEHAVIORAL AND PHYSIOLOGICAL RESPONSES IN WISTAR RATS EXPOSED TO *HYOSCYAMUS ALBUS* L.

Nour El Imène BOUBLATA^{1*}, Sarra HABBACHI¹, Alina Iuliana TABIRCA², Reda DJAOUHADOU³, Wafa HABBACHI¹, Khellaf REBBAS^{4,5}, Abedkrim TAHRAOUI¹

¹Laboratory of Applied Neuroendocrinology. Badji Mokhtar - Annaba University P.O Box.12, Annaba-23000 Algeria

²Faculty of Economics, Valahia University of Targoviste, 130004 Targoviste, Romania

³Research Laboratory in Innovation and Economic and Financial Analysis. Badji Mokhtar - Annaba University P.O Box.12, Annaba-23000 Algeria

⁴Laboratory of Agro-biotechnology and Nutrition in arid and semi-arid zones, Ibn Khaldoun University, Tiaret, Algeria

⁵Department of Biology, Faculty of Science-Université Med Boudiaf, M'Sila 28000

Abstract: *Hyoscyamus albus* L. (*Solanaceae*) is a medicinal plant traditionally used for its sedative and parasympatholytic properties and contains flavonoids, alkaloids, and tropane derivatives. This study evaluated the effects of aqueous extracts from the aerial parts of *H. albus*, collected in the M'sila region (Algeria), on biochemical, hormonal, and behavioral parameters in Wistar rats. Animals received 1 mL/day of extract (292 g/L) orally for seven consecutive days. Biochemical analyses revealed significant changes in blood glucose, cholesterol, triglycerides, urea, and creatinine, including hypoglycemia and reduced cholesterol levels in treated animals. Plasma adrenocorticotrophic hormone (ACTH) was also significantly modulated. Behavioral assessments using the open field, elevated plus maze, and forced swimming tests showed a clear reduction in anxiety-like behavior. These findings indicate that *H. albus* aqueous extract exerts both anxiolytic and metabolic effects, supporting its traditional use and highlighting potential therapeutic applications.

Keywords: *Hyoscyamus albus* L., *Solanaceae*, anxiolytic, behaviour, biochemistry.

INTRODUCTION

The introduction of insect pests into crops poses a significant challenge, reducing production yields and leading to substantial time and financial losses (Donatelli et al., 2017). Synthetic pesticides are commonly used to address this issue due to their rapid action and ease of application (Weinberger et al., 2009; Satpathy et al., 2020; Mena et al., 2022). However, the continued use of these chemicals can lead to various environmental problems (Aktar et al., 2009; Bolzonella et al., 2019), which may negatively impact human health and other living organisms (Mineau et al., 2005; Hua et al., 2012; Braak et al., 2018).

Additionally, the intensive use of synthetic pesticides over the last century has resulted in increased resistance among crop pests (Hawkins et al., 2019). Therefore, it is crucial to find more effective alternatives with lower environmental impact to manage pest populations (Furlan et al., 2015). Various agricultural alternatives to systemic pesticides were evaluated by Furlan et al. in 2021, including changes in agricultural techniques, landscape manipulation, the use of specialized organisms to control pests, and the establishment of traps (Furlan et al., 2021). One such alternative is the use of naturally occurring pesticides, such as plant extracts, which are more sustainable and environmentally friendly than synthetic commercial insecticides (Amoabeng et al., 2019).

The use of plants to protect crops from insect pests' dates back to ancient civilizations and continues to be widely practiced by farmers globally, particularly in regions where synthetic pesticides are less accessible

and in organic farming. These practices involve the use of plant materials such as extracts, companion plants, or harvested plants (Belmain et al., 2001; Grzywacz et al., 2014; Stevenson et al., 2016; Mobolade et al., 2019; Habbachi et al., 2020; Boublata et al., 2020; Boublata et al., 2021a; Boublata et al., 2021b; Habbachi et al., 2021; Saadane et al., 2021a; Saadane et al., 2021b; Bouzar et al., 2022a; Bouzar et al., 2022b).

Natural plant products have gained increasing importance in recent years as researchers investigate them as sources of new insecticides (Pavela, 2016). *Hyoscyamus* is a diverse genus within the *Solanaceae* family, comprising 20 species globally known as henbanes (Mohy-ud-dint et al., 2010). This genus is notable for its rich reservoir of tropane alkaloids, which have pharmacological effects such as antispasmodic, mydriatic, anticholinergic, and antiemetic properties (Herborane and Baxter, 1993). Among these species, *H. albus* is particularly renowned in Europe for its medicinal uses. Historically, it was used in rituals by oracles and soothsayers under various names like dragon plant, Zeus bean, and Apollo's plant (Ratsch, 2005). Its seeds were burned as incense, and its leaves were used as additives in wine. The inhalation of seed smoke and consumption of leaf-infused wine were believed to invoke the deity Apollo. Today, *H. albus* continues to be used in Moroccan incense rituals (Ratsch, 2005; Weinberger et al., 2009).

Studies on *H. albus* have revealed that its crude methanol leaf extract possesses analgesic and antipyretic properties (Benhouada and Yahiaï, 2014).

*Correspondence: Nour El Imène Boublata, Laboratory of Applied Neuroendocrinology. Badji Mokhtar - Annaba University P.O Box.12, Annaba-23000 Algeria, phone number: +213 (0) 540 90 91 01, email: nour-el-imene.boublata@univ-annaba.dz

Mahmood et al. (2001) identified a new tropane alkaloid, 2,3-dimethylnonacosane, along with known compounds such as scopolamine, atropine, hyoscyne, and hyoscyamine in the aerial parts of this plant. Additionally, tropane alkaloids hygrine and norhygrine have been reported in the roots and aerial parts of various *Hyoscyamus* species (Parr et al., 1990). The anticholinesterase activities of scopolamine and hyoscyamine isolated from different *Hyoscyamus* species have also been investigated (Schmeller et al., 1995).

This study aims to investigate the impact of the aqueous extract derived from the Algerian Sahara plant *H. albus* on Wistar rats, with a focus on assessing both the effectiveness of this extract and its potential neurobehavioral outcomes following administration.

MATERIALS AND METHOD

Animal

To conduct the tests, we used adult Wistar rats (*Rattus rattus*) obtained from the Pasteur Institute of Algiers (Algeria), each weighing between 180 and 200 grams. The rats were housed in plastic cages lined with sawdust, equipped with steel lids and water bottles, in the animal house of the University of Annaba's Faculty of Biology. They were fed a diet of corn sticks, barley, and vitamin supplements. The sawdust bedding was

replaced two to three times a week for all animal groups. The animals were kept under laboratory conditions with a temperature of 25°C, humidity levels between 70-80%, and a 12:12 hour light-dark cycle.

Hyoscyamus albus L.

H. albus, an endemic Saharan plant in the Solanaceae family, is commonly used by the people of southern Algeria. This hallucinogenic plant was used in ancient Greece for divinatory purposes and ritual fumigations (Schultes and Hofmann, 1993). Scopolamine, a plant alkaloid found in *H. albus*, causes anterograde amnesia and loss of willpower and was employed as a "truth serum" during World War II (Goullé et al., 2004). White henbane (*Hyoscyamus albus* L.) contains high levels of tropane alkaloids, such as atropine and scopolamine, making all henbane varieties toxic. However, in therapeutic doses, henbane is used as a parasymphatholytic and nervous sedative (Jouzier, 2005).

H. albus, also known as white henbane, is an annual or biennial plant used to produce hyoscyamine, a plant-derived anticholinergic medication (Tamari et al., 2014). This herbaceous plant grows 20-80 cm tall, has dense, sticky glandular hairs, emits a strong odor, and features an erect, slender stem.

A photograph of the plant is shown in Figure 1.



Fig. 1: *Hyoscyamus albus* L (Original photo).

Collection Site of plant material

The leaves of *H. albus* were collected in December 2020 from their natural habitat around the region of M'sila, Algeria. The plant was identified by Pr.

RABBS.K from the Department of Biology, Faculty of Sciences, Med Boudiaf University, M'Sila, Algeria (33°48'24" north latitude, 2°52'56" east longitude).

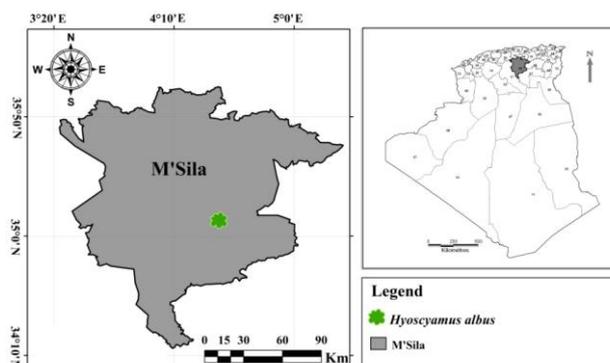


Fig. 2: Map of the M'sila region.

Preparation of the aqueous extract

After harvesting, the aerial parts of the plant were washed and dried at room temperature in our laboratory at the Department of Biology, Faculty of Sciences, University Badji Mokhtar Annaba, Algeria. The dried material was then ground into a powder using an electric grinder and sieved to obtain a homogeneous consistency.

To prepare the aqueous extract of *H. albus* leaves, 149 g of fresh leaves were boiled in 1 L of distilled water for 30 minutes on a hot plate. The resulting mixture was filtered using filter paper, and one liter of the filtrate was collected.

The extract was stored in dark-colored glass bottles at 4°C and used within 48 hours after preparation in order to avoid microbial contamination and degradation of active compounds.

Treatments

A total of forty rats were divided into two groups: a control group (20 individuals: 10 males and 10 females) and a treatment group (20 individuals: 10 males and 10 females). The treatment group was intoxicated by gavage with 1 ml of *H. albus* extract (292 g/l) for 7 consecutive days.

Rat's Behaviors in different anxiety-provoking situations

The behavioral tests conducted include the open field test (OP) (Crawley, 1999; Palanza, 2001; Karl et al., 2003; Prut and Belzung, 2003; Elizalde et al., 2008), the elevated plus maze (EPM) (Rodgers and Dalvi, 1997; Van Gaalen and Steckler, 2000; Karl et al., 2003; Walf and Frye, 2007; Elizalde et al., 2008), and the forced swim test (Porsolt et al., 1977; Detke et al., 1995; Karl et al., 2003; Elizalde et al., 2008; Alijanpour et al., 2019).

Behavioral Studies for Anxiety

The behavioral tests were conducted in the following order, with a 48-hour interval between each to avoid interference with subsequent results: open field (OF) test, elevated plus maze (EPM) test, and forced swim test. After a 20-minute acclimation period to the test room during the lighting phase, all tests were performed in a quiet room between 10:00 and 12:00. After each behavioral evaluation, the rats were returned to their breeding room.

Open field test (OFT)

This test is well-known for assessing the effects of tested chemicals on exploratory behavior and anxiety (Galdino et al., 2009; Asif et al., 2019). To prevent animals from escaping, the test was conducted in a square-shaped open area (70 cm x 70 cm) surrounded by a 40 cm high wall. The rats' capacity to explore the arena was measured for 5 minutes after they were individually introduced to the open field. The number of entries and the time spent in the central area were recorded, as a preference for the central region is an indicator of reduced anxiety.

Elevated plus maze test (EPM)

This is a common behavioral test used to assess the effects of test chemicals on anxiety (Barua et al., 2009; Leo and Pamplona, 2014; Asif et al., 2019). The test apparatus used was an elevated plus maze (110 cm x 10 cm) with both open and closed arms, elevated to a height of 50 cm. Individual rats were evaluated for 5 minutes while starting from the center of the maze facing an open arm. The time spent in the open and closed arms indicates the plant's anxiolytic potential, as increased preference for the exposed areas is considered indicative of lower anxiety in rodents (Foyet et al., 2012).

Behavioral Studies for Depression

Forced swimming test (FST)

The forced swim test (FST) was used to assess depressive behavior in rats (Porsolt et al., 1977). The apparatus consisted of an aquarium (54 cm x 38 cm x 40 cm) filled with water at 25°C to a height of 30 cm. At this depth, the rat cannot touch the bottom with its hind limbs and is therefore compelled to swim. The water temperature was maintained using an electric immersion heater before each session.

The FST was conducted in two sessions: a pre-test and a test, separated by a 24-hour interval. During the pre-test, the rat was placed in the water-filled aquarium for 15 minutes, where it could not escape, to induce a state resembling depression (depressed session). On the test day, the rat underwent a 5-minute session in the aquarium, during which time spent immobile, swimming, and climbing were recorded as indicators of depressive behavior among groups during behavioral tests.

Effect on certain biochemical parameters

Blood samples were collected from all experimental groups, including both untreated (control) and *H. albus*-treated animals. Plasma was separated by centrifugation at 3000 rpm for 10 minutes and immediately frozen, then stored at -20°C until analysis.

The concentration of ACTH in the plasma was determined using a radioimmunoassay kit according to the manufacturer's instructions (Raff et al., 2004).

In addition, biochemical parameters including blood glucose, total cholesterol, triglycerides, urea, and creatinine were measured using standard enzymatic methods with commercial kits (Manufacturer, Country), following the manufacturer's protocols.

Data Analysis

The study data were analyzed using descriptive statistics and comparative methods, specifically variance analysis, conducted using XLStat 2009 software (Addinsoft, New York, NY).

RESULTS & DISCUSSION

Rats' behaviors in different anxiety-provoking situations

Open Field Test

The anxiolytic and sedative effects of *Hyoscyamus albus* were evaluated using the open field test, which takes advantage of rats' natural aversion to open spaces and their exploratory behavior (Brown et al., 1999).

Male rats treated with the aqueous extract of *H. albus* spent significantly more time in the central area of the apparatus (21.21 ± 19.82 s) compared to the control group (1.53 ± 0.58 s), indicating a marked reduction in anxiety-like behavior (Table 1). In females rats, no significant differences were detected between treated and control groups for any of the measured parameters ($p > 0.05$), indicating an absence of anxiolytic or sedative effects under the experimental conditions used. This sex-dependent response suggests a differential sensitivity to *H. albus* extract between male and female rats.

These results are consistent with previous studies reporting similar anxiolytic effects using extracts of

Solanaceae and other medicinal plants. For instance, Abubakar et al. (2020) observed increased central area exploration in rats treated with fractions of the methanolic extract of *Solanum aethiopicum* fruit. Similarly, studies by Boublata et al. (2020, 2021) using aqueous and ethanolic extracts of *Cleome arabica* L. demonstrated significant differences in time spent in the central and peripheral areas, further supporting the anxiolytic potential of plant-derived compounds.

Overall, the increase in central area activity in treated rats confirms the anxiolytic and exploratory-promoting effects of *H. albus*, aligning with previous literature and validating its traditional use as a sedative agent.

Table 1.

H. albus effect on the behavior of rats (in open filed)

N=10	Time spent in the central area	Time spent in the peripheral area	Number of adjustments	Time of immobility
♂C	1.53±0.58	298.46±0.58	22.00±5.67	136.46±31.82
♂H.a	21.21±19.82	278.78±19.82	24.00±5.93	101.36±43.90
F_{obs}	1156.11	1156.13	1.09	1.90
p	<0.0001***	<0.0001***	0.92	0.49
♀C	1.00±0.46	299.00±0.46	22.14±3.58	103.92±24.51
♀H.a	0.84±0.42	299.15±0.42	26.14±3.82	77.75±34.63
F_{obs}	1.20	1.20	1.17	1.99
p	0.82	0.82	0.85	0.42

[C: Control; H.a: *H. albus*] [* significant*; ** highly significant; *** very highly significant]

Plus maze test

The elevated plus maze (EPM) is a widely used model to assess anxiolytic effects, based on the conflict between rodents' natural fear of elevated open spaces and their exploratory drive (Handley and Mithani, 1984; Dawson and Tricklebank, 1995). Anxiolytic agents are expected to increase entries into and time spent in the open arms (Lister, 1987).

In this study, treatment with *H. albus* significantly decreased the time spent in the closed arms in male rats (126.98 ± 34.99 s) compared to controls (227.26 ± 23.94 s), indicating reduced anxiety-like behavior (Table 2). These results are consistent with findings from Momin and Mohan (2012), who reported significant anxiolytic effects of the methanolic extract of *Solanum torvum* in mice.

Female rats treated with *H. albus* spent a similar amount of time in the center of the maze (221.94 ± 27.54 s) compared to controls (222.26 ± 27.93 s), suggesting that sex differences did not influence the anxiolytic effect under these conditions (Table 2). Similar observations were reported with acute exposure to *Solanum macrocarpon* leaf extracts (Giorgetti and Negri, 2011).

Overall, these findings support the anxiolytic potential of *H. albus*, aligning with results from previous studies on Solanaceae and other medicinal plants (Luciana et al., 2014; Katolkar et al., 2015; Yusuf et al., 2016; Caro et al., 2018; Abubakar et al., 2020; Ozioko et al., 2020; Boublata et al., 2020, 2021a, 2021b; Jhansi Lakshmi et al., 2021).

Table 2.

H. albus effect on the behavior of rats (in Elevated plus maze)

N=10	Time spent in closed arms	Time spent in open arms	Number of closed arms	Number of open arms
♂C	227.26±23.94	49.63±27.61	5.00±2.21	2.83±1.07
♂H.a	126.98±34.99	62.80±22.76	4.50±1.17	3.50±0.99
F_{obs}	2.36	1.47	3.55	1.18
p	0.04*	0.68	0.19	0.86
♀C	222.02±27.93	19.49±10.28	4.42±1.25	2.42±0.52
♀H.a	221.94±27.54	51.62±24.25	6.42±0.97	3.57±0.68
F_{obs}	1.02	5.56	1.65	1.68
p	0.94	0.05	0.55	0.58

[C: Control; H.a: *H. albus*] [* significant*; ** highly significant; *** very highly significant]

Forced swimming test

The forced swimming test is a widely used behavioral model to assess antidepressant-like activity

in rodents. When placed in an inescapable water-filled container, rats or mice eventually adopt an immobile posture, reflecting behavioral despair analogous to

depressive-like states (Porsolt et al., 1977; Kulkarni and Mehta, 1985).

In this study, oral administration of the aqueous extract of *H. albus* significantly reduced immobility time in male rats (111.92 ± 26.75 s) compared to controls (173.48 ± 0.61 s) (Table 3). Female rats also showed a similar reduction (75.10 ± 23.58 s vs. 117.16 ± 1.39 s in controls) (Table 3). Treated animals displayed increased swimming activity, further indicating antidepressant-like effects (Table 3).

These findings are consistent with previous studies on *Hyoscyamus niger*, which demonstrated reduced

immobility in the forced swim and tail suspension tests without affecting motor activity (de Patil et al., 2013). Comparable antidepressant-like effects have been reported with other medicinal plants, including *Datura fastuosa* (Abena et al., 2004) and *Cestrum nocturnum* (Paragati et al., 2016; Snafi, 2018; Firdaus et al., 2020; Jhansi Lakshmi et al., 2021), all showing decreased immobility in rodent models.

Overall, these results highlight the potential of *H. albus* in modulating depressive-like behaviors in rodents, supporting further investigation into its therapeutic applications for mood disorders.

Table 3.

H. albus effect on depressive state

N=10	Swimming time	Climbing time	Time of immobility
♂C	126.52±2.03	2.27±0.07	173.48±0.61
♂H.a	188.08±27.28	38.80±13.73	111.92 ±26.75
F_{obs}	300.19	41941.33	3194.61
p	0.007**	<0.0001***	<0.001**
♀C	126.52±0.83	2.47±0.14	117.16±1.39
♀H.a	157.28±21.08	67.52±6.97	75.10±23.58
F_{obs}	1104.68	4125.72	666.56
p	<0.0001***	<0.001	0.003**

[C: Control; H.a: *H. albus*] [* significant*; ** highly significant; *** very highly significant]

***H. albus* effect on biochemical parameters and the hormone ACTH:**

Oral administration of the aqueous extract of *Hyoscyamus albus* did not induce any visible signs of acute toxicity or mortality during the observation period. Biochemical analysis (Table 4) revealed no significant differences in urea and creatinine levels between treated and control rats of both sexes ($p > 0.05$), with urea values ranging from 0.30 ± 0.04 to 0.35 ± 0.01 g/L and creatinine levels between 7.02 ± 0.17 and 7.97 ± 0.19 mg/L. Similarly, triglyceride concentrations were not significantly affected by the treatment in either males (0.46 ± 0.23 vs. 0.37 ± 0.03 g/L) or females (0.44 ± 0.04 vs. 0.37 ± 0.03 g/L), indicating no alteration of lipid metabolism at this level (Table 4).

Treatment with *H. albus* significantly reduced cholesterol levels in both male and female rats, decreasing from 0.72 ± 0.06 to 0.49 ± 0.02 g/L in males ($p = 0.008$) and from 0.67 ± 0.09 to 0.41 ± 0.05 g/L in females ($p = 0.03$) (Table 4). In addition, a pronounced hypoglycemic effect was observed in female rats, with glycemia decreasing from 1.16 ± 0.02 to 0.78 ± 0.04 g/L ($p < 0.00001$), whereas no

significant change was detected in males (0.97 ± 0.03 vs. 1.07 ± 0.04 g/L; $p > 0.05$) (Table 4). These results support the potential anti-diabetic activity of *H. albus*, in agreement with previous studies (Mukherjee et al., 2006; Tanko et al., 2008; Dass et al., 2009; Benhouda et al., 2014; Choudhary et al., 2016; Yahia et al., 2016).

The active constituents of *H. albus*, including calystegines, have been reported to protect β -cells of the islets of Langerhans, promote β -cell regeneration, enhance insulin secretion, and improve lipid profiles in diabetic models (Bourebaba et al., 2016).

Furthermore, plasma ACTH levels were significantly reduced in treated animals, suggesting anti-stress effects. This is consistent with findings on withanolides from *Datura fastuosa*, which exhibit anxiolytic and anti-stress properties by preventing stress-induced adrenal cortisone depletion (Firdaus et al., 2020; Boublata et al., 2020, 2021a, 2021b). These results highlight the potential of *H. albus* as both a metabolic modulator and anti-stress agent, warranting further investigation of dosage, duration, and molecular mechanisms.

Table 4.

H. albus effect on biochemical parameters and the hormone ACTH

N=10	Glycemia (g/l)	Cholesterol (g/l)	Triglyceride (g/l)	Urea (g/l)	Creatinine (mg/l)	ACTH (pg/ml)
♂C	0.97±0.03	0.72±0.06	0.46±0.23	0.33±0.02	7.97±0.19	78.42±3.95
♂H.a	1.07±0.04	0.49±0.02	0.37±0.03	0.30±0.04	7.02±0.17	38.52±7.23
F_{obs}	1.48	3.48	0.82	0.55	1.31	6.93
P	0.71	0.008**	0.43	0.59	0.79	0.002**
♀C	1.16±0.02	0.67±0.09	0.44±0.04	0.35±0.01	7.71±0.21	23.13±0.49
♀H.a	0.78±0.04	0.41±0.05	0.37±0.03	0.33±0.02	7.02±0.17	5.70±1.72
F_{obs}	8.55	2.52	1.12	0.82	2.45	9.73
P	<0.00001***	0.03*	0.29	0.43	0.40	0.001**

[C: Control; H.a: *H. albus*] [* significant*; ** highly significant; *** very highly significant]

CONCLUSION

The present study demonstrates that the aqueous extract of *H. albus* exerts significant anxiolytic- and antidepressant-like effects in Wistar rats, as evidenced by the open field, elevated plus maze, and forced swimming tests. In addition, the extract significantly modulated biochemical parameters, including blood glucose and cholesterol levels, and reduced ACTH concentrations, indicating potential anti-diabetic and anti-stress properties. No signs of acute toxicity were observed, supporting the safety of the extract under the tested conditions.

These findings validate the traditional use of *H. albus* as a sedative and metabolic modulator and highlight its potential therapeutic applications. Further studies are warranted to isolate and characterize the active compounds, investigate their molecular mechanisms, and explore additional pharmacological and biological effects, including potential applications in pest control.

AUTHORS CONTRIBUTION

All authors equally contributed to this study. Nour El Imène BOUBLATA, Sarra HABBACHI, Alina Iuliana TABIRCA, Reda DJAOUAHDOU, Wafa HABBACHI, Khellaf REBBAS, Abedkrim TAHRAOUI, designed and carried out the experimental study and wrote the manuscript.

FUNDING

This research was not funded by any institution, industrial group, or any other party.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

Abena AA, Miguel LM, Mouanga A, Ouamba JM, Sianard DF, Thiebolt MH, Hondi-Assah TC, Diatwa M. Neuro-psychopharmacological effects of leaves and seeds extracts of *Datura fastuosa*. *Biotechnol.* 2004;3(2):109–113.

Abubakar AR, Sani IH, Malami S, Yaro AH. Neuropharmacological activity of various fractions obtained from *Solanum aethiopicum* (Linn.) fruit in mice. *J Basic Soc Pharm Res.* 2020;1(5):12–23.

Aktar MW, Sengupta D, Chowdhury A. Impact of pesticides use in agriculture: their benefits and hazards. *Interdiscip Toxicol.* 2009;2:1–12.

Alijanpour S, Khakpai F, Ebrahimi-Ghiri M, Zarrindast MR. Co-administration of low dose orexin and nitrenergic antagonists induces an antidepressant-like effect in mice. *Biomed Pharmacother.* 2019;109:589–594.

Amoabeng BW, Johnson AC, Gurr GM. Natural enemy enhancement and botanical insecticide source: a review of dual use companion plants. *Appl Entomol Zool.* 2019;54:1–19.

Asif HM, Hayee A, Aslam MR, Ahmad K, Hashmi AS. Dose-dependent antidepressant and anxiolytic effects of a traditional medicinal plant in animal models. *Dose-Response.* 2019;17(4):1–6.

Barua CC, Roy JD, Buragohain B, Barua AG, Borah P, Lahkar M. Anxiolytic effect of hydroethanolic extract of *Drymaria cordata* L. Willd. *Indian J Exp Biol.* 2009;47(12):969–973.

Belmain S, Stevenson P. Ethnobotanicals in Ghana: reviving and modernising age-old farmer practice. *Pestic Outlook.* 2001;12:233–238.

Benhouada A, Yahia M, Khadhraoui H, Benbia S, Benhouada D. Hypoglycemic activity of methanolic extract of *Hyoscyamus albus* L. leaves in streptozotocin-induced diabetic rats. *Nat Prod Chem Res.* 2014;2:5.

Benhouada A, Yahia M. Toxicity, analgesic and anti-pyretic activities of methanolic extract from *Hyoscyamus albus* leaves in albino rats. *Int J Pharm Pharm Sci.* 2014;6:121–127.

Bolzonella C, Zanella A, Sartori G, Jabiol B, Nicolini G, Zechmeister-Boltenstern S, et al. Humusica 2, Article 19: techno humus systems and global change – conservation agriculture and 4/1000 proposal. *Appl Soil Ecol.* 2019;122:271–296.

Boublata NEI, Bekhakheche M, Habbachi S, Saadane FZ, Bouzar A, Habbachi W, Rebbas K, Tahraoui A. Undesired effects of bioinsecticide molecules in Wistar rats. *J Bioresource Manag.* 2021;8(4):27–54.

Boublata NEI, Habbachi W, Habbachi S, Saadane FZ, Benhissen S, Tahraoui A. Effects of *Cleome arabica* aqueous extract in Wistar rat behavior, biochemical parameters and ACTH hormone. *Curr Trends Nat Sci.* 2020;9(18):202–209.

Boublata NEI, Saadane FZ, Habbachi S, Bouzar A, Habbachi W, Benhissen S. Effects of *Cleome arabica* ethanolic extract in Wistar rat behavior, biochemical parameters and ACTH hormone. *Bioscience Research.* 2021;18(2):1471–1479.

Bourebaba L, Saci S, Touguit D, Gali L, Terkmane S, Oukil N, Bedjou F. Evaluation of antidiabetic effect of total calystegines extracted from *Hyoscyamus albus*. *Biomed Pharmacother.* 2016;82:337–344.

Bouzar A, Samai I, Habbachi S, Rahat M, Boublata NEI, Saadane FZ, Habbachi W, Tahraoui A. Insecticidal effects of the spontaneous plant *Urtica dioica* L. (Urticaceae) on the mortality and behavior of *Drosophila melanogaster* (Diptera: Drosophilidae). *Asia Life Sci.* 2022a;12(6):1267–1280.

Bouzar A, Habbachi S, Samai I, Rahat M, Boublata NEI, Saadane FZ, Habbachi W, Benhissen S, Tahraoui A. *Nicotiana glauca* Graham (Solanaceae) bioactivity and toxic effects on mortality, feeding behavior and pupation choice of *Drosophila melanogaster* larvae (Diptera: Drosophilidae). *Journal of Bioresource Management.* 2022b;9(4):149–161.

Braak N, Neve R, Jones AK, Gibbs M, Breuker CJ. The effects of insecticides on butterflies – a review. *Environ Pollut.* 2018;242:507–518.

Brown RE, Corey SC, Moore AK. Differences in measures of exploration and fear in MHC-congenic C57BL/6J and B6-H-2k mice. *Behav Genet.* 1999;29(4):263–271.

- Caro DC, Rivera-de Ocampo Y, Franco LA, Salas RD. Pharmacological evaluation of *Mentha spicata* L. and *Plantago major* L., medicinal plants used to treat anxiety and insomnia in Colombian Caribbean Coast. *Evid Based Complement Alternat Med.* 2018;2018:1592151.
- Choudhary P, Khandelwal V, Bhatia AK, Goel A, Sharma R, Goel R. Efficacy of aqueous extract of *Solanum xanthocarpum* on hematological and biochemical parameters of Wistar albino rat. *Adv Biores.* 2016;7(3):155–161.
- Crawley JN. Behavioral phenotyping of transgenic and knockout mice: experimental design and evaluation of general health, sensory functions, motor abilities, and specific behavioral tests. *Brain Res.* 1999;835(1):18–26.
- Dass A, Palaniswamy M, Angayarkanni J, Dhanabalan R. Antidiabetic activity of water extract of *Solanum trilobatum* (Linn.) in alloxan-induced diabetic rats. *Afr J Biotechnol.* 2009;8(20):5562–5564.
- Dawson GR, Tricklebank MD. Use of the elevated plus maze in the search for novel anxiolytic agents. *Trends Pharmacol Sci.* 1995;16(2):33–36.
- Detke MJ, Rickels M, Lucki I. Active behaviors in the rat forced swimming test differentially produced by serotonergic and noradrenergic antidepressants. *Psychopharmacology.* 1995;121:66–72.
- Donatelli, M., Magarey, R.D., Bregaglio, S., Willocquet, L., Whish, J.P.M., & Savary, S. (2017). *Modelling the impacts of pests and diseases on agricultural systems.* *Agricultural Systems*, 155, 213–224. <https://doi.org/10.1016/j.agsy.2017.01.019>
- Elizalde N, Gil-Bea FJ, Ramirez MJ, et al. Long-lasting behavioral effects and recognition memory deficit induced by chronic mild stress in mice: effect of antidepressant treatment. *Psychopharmacology.* 2008;199(1):1–14.
- Firdaus N, Viqar U, Kazmi MH. Potential and pharmacological actions of *Datura metel* L.: as a deadly poison and as a drug: an overview. *Int J Pharm Sci Res.* 2020;11(7):3123–3137.
- Foyet HS, Tsala De, Bouba AA, Hritcu L. Anxiolytic and antidepressant-like effects of the aqueous extract of *Alafia multiflora* stem bark in rodents. *Adv Pharmacol Sci.* 2012;2012:912041.
- Furlan L, Kreutzweiser D. Alternatives to neonicotinoid insecticides for pest control: case studies in agriculture and forestry. *Environ Sci Pollut Res.* 2015;22:135–147.
- Furlan L, Pozzebon A, Duso C, Simon-Delso N, Sánchez-Bayo F, Marchand PA, Codato F, Lexmond MB, Bonmatin JM. An update of the worldwide integrated assessment on systemic insecticides. Part 3: alternatives to systemic insecticides. *Environ Sci Pollut Res.* 2021;28:11798–11820.
- Galdino PM, Nascimento MVM, Sampaio BL, Ferreira RN, Paula Jr, Costa EA. Antidepressant-like effect of *Lafoensia pacari* A. St-Hil. ethanolic extract and fractions in mice. *J Ethnopharmacol.* 2009;124(3):581–585.
- Giorgetti M, Negri G. Plants from Solanaceae family with possible anxiolytic effect reported in 19th century's Brazilian medical journal. *Rev Bras Farmacogn.* 2011;21(4): 772–780
- Goullé JP, Pépin G, Dumestre TV, Lacroix C. Botany, chemistry and toxicology of hallucinogenic Solanaceae: belladonna, datura, henbane, mandrake. *Ann Tox Anal.* 2004;22:22–35.
- Grzywacz D, Stevenson PC, Mushobozi WL, Belmain S, Wilson K. The use of indigenous ecological resources for pest control in Africa. *Food Sec.* 2014;6:71–86.
- Habbachi S, Boubalta NEI, Benhissen S, Habbachi W, Rebbas K, Tahraoui A. Evaluation of *Cleome arabica* L. toxicity: effects on mortality and sexual behaviour of *Drosophila melanogaster* (Diptera: Drosophilidae). *Curr Trends Nat Sci.* 2020;9(17):210–217.
- Habbachi S, Boublata NEI, Saadane FZ, Bouzar A, Habbachi W, Benhissen S, Rebbas K, Tahraoui A. Effects of *Hyoscyamus albus* aqueous extracts on mortality, sexual behavior and oviposition of *Drosophila melanogaster* (Diptera: Drosophilidae). *Studia Univ Vasile Goldis Life Sci Ser.* 2021;31(3):137–143.
- Handley SL, Mithani S. Effects of α -adrenoceptor agonists and antagonists in a maze-exploration model of “fear”-motivated behaviour. *Naunyn Schmiedebergs Arch Pharmacol.* 1984;327(1):1–5.
- Harborne JB, Baxter H. *Phytochemical Dictionary: Handbook of Bioactive Compounds from Plants.* London: Taylor & Francis; 1993.
- Hawkins NJ, Bass C, Dixon A, Neve P. The evolutionary origins of pesticide resistance. *Biol Rev Camb Philos Soc.* 2019;94:135–155.
- Hua J, Relyea RA. East coast vs west coast: effects of an insecticide in communities containing different amphibian assemblages. *Freshw Sci.* 2012;31:787–799.
- Jhansi Lakshmi V, Santosh Kumar R, Amulya RK, Srinu P. Review on pharmacological effects of night blooming jasmine (*Cestrum nocturnum*). *J Global Trends Pharm Sci.* 2021;12(2):9396–9402.
- Jouzier E. Medicinal Solanaceae and philately. *Plante Sci.* 2005;144:311–332.
- Karl T, Pabst R, von Horsten S. Behavioral phenotyping of mice in pharmacological and toxicological research. *Exp Toxicol Pathol.* 2003;55:69–83.
- Katolkar P, Bhuskute S, Duragkar N. Evaluation of antianxiety and antidepressant activity of *Cestrum nocturnum* leaves. *Int J Res Biosci Agric Technol.* 2015;2(3):232–236. doi:10.29369/ijrbat.2015.03.ii.0067:
- Kulkarni SK, Mehta AK. Purine nucleoside-mediated immobility in mice: reversal by antidepressants. *Psychopharmacology.* 1985;85:460–463.
- Lister RG. The use of an elevated plus-maze to measure anxiety in the mouse. *Psychopharmacol.* 1987;92:180–185.
- Luciana M. Leo, Suellen Almeida-Corrêa, Claudio A. Canetti, Olavo B. Amaral, Fernando A.

- Bozza. Age-Dependent Relevance of Endogenous 5-Lipoxygenase Derivatives in Anxiety-Like Behavior in Mice. *PLoS ONE*. 2014;9(7):e103943. <https://doi.org/10.1371/journal.pone.0103949>
- Leo LM, Pamplona FA. Elevated Plus Maze Test to Assess Anxiety-like Behavior in the Mouse. *Bio-protocol*. 2014;4(16):e1211. doi:10.21769/BioProtoc.1211
- Mahmood U, Yogendra S, Raghunath S, Thakur R. 2,3-Dimethyl nonacosane and tropane alkaloids from *Hyoscyamus albus*. *Phytochemistry*. 2001;24:1618–1619.
- Mena TT, Coa TH, Phiena HH, Nguyen YDH, Tuan LA Pham, Kamei BK, Binh TD. Evaluation of the insecticidal activity of *Solanum mammosum* (L.) fruit extract against *Drosophila melanogaster*. *J Anim Behav Biometeorol*. 2022;10:2218.
- Mineau P, Downes CM, Kirk DA, Bayne E, Csizy M. Patterns of bird species abundance in relation to granular insecticide use in the Canadian prairies. *Ecoscience*. 2005;12:267–278.
- Momin R, Mohan M. Involvement of central noradrenaline, serotonin and dopamine system in the antidepressant activity of fruits of *Solanum torvum* (Solanaceae). *Natural Product Research*. 2012;26(5):416422. <https://doi.org/10.1080/14786419.2010.49507>
- Mobolade AJ, Bunindro N, Sahoo D, Rajashekar Y. Traditional methods of food grains preservation and storage in Nigeria and India. *Ann Agric Sci*. 2019;64:196–205.
- Mohy-ud-Dint A, Khan Z, Ahmad M, Kashmiri MA. Chemotaxonomic value of alkaloids in *Solanum nigrum* complex. *Pak J Bot*. 2010;42:653–660.
- Mukherjee P, Maiti K, Mukherjee K, Houghton PJ. Leads from Indian medicinal plants with hypoglycemic potentials. *J Ethnopharmacol*. 2006;106:1–28.
- Ozioko US, Mba CE, Ozor II, Egwuatu D. Anxiolytic and curative effect of *Solanum macrocarpon* leaves extract on acetaminophen-induced brain injury in adult Wistar rats. *J Pharmacogn Phytochem*. 2020;9(3):205–212.
- Palanza P. Animal models of anxiety and depression: how are females different? *Neurosci Biobehav Rev*. 2001;25:219–233.
- Parr AJ, Payne J, Eagles J, Chapman BT, Robins RJ, Rhodes MJC. Variation in tropane alkaloids accumulation within the Solanaceae and strategies for its exploitation. *Phytochemistry*. 1990;29:2545–2550.
- Patil AD, Patil AY, Rajee AA. Antidepressant-like property of *Hyoscyamus niger* Linn. in mouse model of depression. *Innov Pharm Pharmacother*. 2013;1(2):60–69.
- Pavela R. History, presence and perspective of using plant extracts as commercial botanical insecticides and farm products for protection against insects – a review. *Plant Protect Sci*. 2016;52:229–241.
- Porsolt RD, Le Pichon M, Jalfre M. Depression: a new animal model sensitive to antidepressant treatments. *Nature*. 1977;266:730–732.
- Pragati R, Dipali R, Pallavi D, Gouri S. Extraction and antimicrobial activity of *Cestrum nocturnum*. *IJR*. 2016;6(5):739–741.
- Prut L, Belzung C. The open field as a paradigm to measure the effects of drugs on anxiety-like behaviors: a review. *Eur J Pharm*. 2003;463(1-3):3–33.
- Raff H, Lee JJ, Widmaier EP, Oaks MK, England WC. Basal and adreno-corticotropin stimulated corticosterone in neonatal rats exposed to hypoxia from birth: modulation by chemical sympathectomy. *Endocrinology*. 2004;145:79–86.
- Ratsch C. The encyclopedia of psychoactive plants: ethno pharmacology and its applications. Inner Traditions/Bear & Co; 2005.
- Rodgers RJ, Dalvi A. Anxiety, defence and the elevated plus-maze. *Neurosci Biobehav Rev*. 1997;21(6):801–810.
- Saadane FZ, Boublata NEI, Habbachi S, Bouzar A, Habbachi W, Slimani A, Tahraoui A. Valorisation of the effects of bioactive compounds of the ethanolic extract of *Ramalina farinacea* (Ramalinaceae) on the development, eating and pupation behaviour of *Drosophila melanogaster* (Diptera: Drosophilidae). *J Bioresource Manag*. 2021; 2021;8(4):113–120. doi:10.35691/JBM.1202.0208
- Saadane FZ, Habbachi W, Habbachi S, Boublata NEI, Slimani A, Tahraoui A. Toxic effects of *Drimia maritima* (Asparagaceae) ethanolic extracts on the mortality, development, sexual behaviour and oviposition behaviour of *Drosophila melanogaster* (Diptera: Drosophilidae). *J Anim Behav Biometeorol*. 2021;9(1):2102. doi:10.31893/jabb.21002.
- Satpathy S, Gotyal BS, Babu VR. Role of novel insecticides in crop protection and their selectivity to natural enemies: a review. *J Environ Biol*. 2020;41:149–160.
- Schmeller T, Sporer F, Sauerwein M, Wink M. Binding of tropane alkaloids to nicotinic and muscarinic acetylcholine receptors. *Pharmazie*. 1995;50:493–495.
- Schultes RE, Hofmann A. *Plants of the Gods: Les plantes hallucinogènes, botanique et ethnologie*. Éd Du Léopard; 1993.
- Snafi AE. Therapeutic importance of *Hyoscyamus* species grown in Iraq (*Hyoscyamus albus*, *Niger* and *Reticulatus*) – a review. *IOSR J Pharm*. 2018;8(6):18–32.
- Stevenson PC, Belmain SR. Pesticidal plants in African agriculture: local uses and global perspectives. *Outlooks Pest Manag*. 2016;27:226–230.
- Tamari N, Mine A, Sako A, Tamagawa S, Tabira Y, Kitamura Y. Possible application of the medicinal plant *Hyoscyamus albus* in phytoremediation: excess copper compensates for iron deficiency, depending on the light conditions. *Am J Plant Sci*. 2014;5:3812–3822.

- Tanko Y, Yerima M, Mahdi MA, Yaro AH, Musa KY, Mohammed A. Hypoglycemic activity of methanolic *Adansonia digitata* extract on blood glucose levels of streptozotocin-induced diabetic Wistar rats. *Int J Appl Res Nat Prod.* 2008;1:32–36.
- Van Gaalen MM, Steckler T. Behavioural analysis of four mouse strains in an anxiety test battery. *Behav Brain Res.* 2000;115(1):95–106.
- Walf A, Frye C. The use of the elevated plus maze as an assay of anxiety-related behavior in rodents. *Nat Protoc.* 2007;2:322–328.
- Weinberger K, Srinivasan R. Farmers' management of cabbage and cauliflower pests in India and their approaches to crop protection. *J Asia Pac Entomol.* 2009;12:253–259.
- Yusuf MB, Bello B, Jaafaru IJ. Anxiolytic effect of aqueous root extract of *Citrus aurantium* in Wistar albino rats. *J Adv Med Pharm Sci.* 2016;9(2):1–9.
- Yahia M, Yahia M, Benhouda A, Chafaa N, Hachani K, Djahida B, Boutaghrine S. Hepatoprotective effect of *Hyoscyamus albus* leaves on carbon tetrachloride-induced acute hepatotoxicity in rats. *European Journal of Biotechnology and Bioscience.* 2016;4(11):8–12