World Journal of Biology Pharmacy and Health Sciences
eISSN: 2582-5542
Cross Ref DOI: 10.30574/wjbphs
Journal homepage: https://wjbphs.com/

(RESEARCH ARTICLE)

Check for updates

Anxiolytic effects of aqueous extract of *Hiptis spicigera* Lam (Lamiaceae) in mice on chronic immobilisation and physical activity test

Omam Omam Jean Pierre [1,*], Hamadou André [2], Mbouh Samuel [2], Kinyock MC Jesus [3], Ze Minkoulou Mireille Delphine [4], Zoa Kingoumbock Christine Monique [1], Etame Loé Gisèle [5] and Ngo Bum Elisabeth [6,7]

[1] *Department of Biological Sciences, High Teacher Training College University of Yaounde I, Yaoundé, Cameroon.*
[2] *National Institute of Youth and Sports, University of Yaoundé I, Yaoundé, Cameroon.*
[3] *Department of Chemistry, High Teacher Training College University of Yaounde I, Yaoundé, Cameroon.*
[4] *Department of Animal Biology and Physiology, Faculty of Sciences University of Yaounde I, Yaoundé, Cameroon.*
[5] *Faculty of Medicine and Pharmaceutical Sciences, University of Douala, Douala, Cameroon.*
[6] *Department of Biological Sciences, University of Maroua, Maroua, Cameroon.*
[7] *Department of Biological Sciences, Faculty of Sciences University of Ngaoundere, Ngaoundere, Cameroon.*

World Journal of Biology Pharmacy and Health Sciences, 2023, 14(02), 296–305

Publication history: Received on 10 February 2023; revised on 25 May 2023; accepted on 27 May 2023

Article DOI: https://doi.org/10.30574/wjbphs.2023.14.2.0215

Abstract

The Anxiety disorders are considering as serious problems in our society life like family environment, school and work functioning. They increase the risk of other psychiatric disorders pathologies, including depression, drug and alcohol abuse and the incidence of suicide. For ameliorate this situation, anxiolytic and hypnotic substances like benzodiazepines, barbiturates and their relatives compounds are very regularly prescribed. However, the frequent use of tranquillizers and psychotropic compounds involve the variety in autonomic, endocrine, allergic, and neurological side effects. So, anxiety disorders are reveled as the world health problem. This study aims at assessing the combined of anxiolytic effect of the decoction of dried *Hypstis spicigera* leaves and aerobic physical activity in white mice. For this purpose, the Elevated Plus Maze test (EPM) and the Open Field (OF) test were used. Thirty male and female mice of approximately eight weeks of age and 23 g body weight were used. They were divided into seven groups of five animals each. The first one normal group received distilled water (per os), a negative control group received distilled water by oral way and underwent anxiety induction and a positive control group received diazepam intraperitoneally and underwent anxiety induction and physical activity. Then, four groups received different concentration of decoction of *Hypstis spicigera* by oral way, followed by chronic immobilisation stress and physical activity. Thus, the results of the EPM showed that chronic stress caused a significant decrease ($p<0.001$) of the percentage of time spent in the open arms from 42.93% in the normal control mice to 14.79% in the negative control group. Similarly, *Hyptis spicigera* lam decoction and physical activity induced a significant ($p<0.001$) increase of this percentage from 14.79% in the negative control group to 82.52% of the 225 mg/kg dose group. The diazepam also induced a significant ($p<0.001$) increase of this percentage. Those from the open field showed that chronic stress caused a significant ($p<0.001$) decrease in the amount of time spent in the centre from 4.8±0.83 s in the normal control mice to 1.2±0.44 s in the negative control group. Similarly, *Hyptis spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) increase of time spent from 1.2±0.44 s in the 225 mg/kg dose group in the negative control group to 15±1s in the 225mg/kg dose group. Diazepam also induced a significant ($p<0.001$) increase of this time. *Hyptis spicigera* Lam decoction and physical activity have anxiolytic activity. Reason is why this plant is used more in traditional medicine.

Keywords: *Hiptis spicigera*; Decoction; Anxiolytic; Stress; Physical Activity

* Corresponding author: Jean Pierre Omam Omam

1. Introduction

Anxiety is a physiological response of the body upcoming the stress. Although, it is a natural adaptive reaction and it can become pathological when the response becomes uncontrolled, excessive and persistent: these are the anxiety disorders [1]. Anxiety disorders, as defined in the Diagnostic and Statistical Manual of Mental Disorder [2], are the most common group of psychiatric disorders. Worldwide more than 260 million people have anxiety disorders (WHO 2017[3]). They most often begin in adolescence or young adults. Certain risk factors are associated with the pathology. The most important are the presence of a family history or personal history of anxiety or marked shyness during childhood or adolescence. Experiencing stressful or traumatic events, such as abuse, can also precipitate an anxiety disorder. Finally, the presence of a psychiatric disorder has been identified as a factor that increases the risk of developing anxiety disorders in our environment [4]. The anxiety disorders lead problems in social, family, school and occupational functioning. They increase the risk of other psychiatric disorders, including depression, substance abuse and the incidence of suicide. [5]. They can cause a great burden on patients and their family members, and could have a significant costs to the society live for increasing health care use and decreasing work productivity (Canadian Psychiatric Association 2006). Physical activity can also be an alternative treatment for anxiety. Physical activity is defined as any bodily movement produced by the muscle that requires the expenditure of energy. It includes both activities of daily living and leisure activities [6]. According to Steptoe *et al.* 1993, Brunner et *al. 2021* [7, 8]. Engaging in physical activity allows for better stress management even without correlation to improved physical fitness. These results suggest that, the increased ability to manage stress depends to the factors other than physiological factors [9]. Managing anxiety to prevent it from undermining performance is an issue of great interest in sport. Most athletes, coaches and psychologists claim that the ability to manage anxiety before, during and after competitions is a critical element of sports performance. This management is achieved through psychological methods and the uses of natural substances [10]. For the management of athletes, anxiolytic and hypnotic benzodiazepines and their relatives are very regularly prescribed. In 2015, 13.4% of the population would have consumed at least one benzodiazepine, whatever the indication, of which 99.6% were anxiolytic and sedative benzodiazepines [1]. However, prolonged use of tranquilizers and psychotropic drugs leads to addiction and a variety of endocrine and neurological side effects. In addition, these agents mainly relieve symptoms and offer a palliative option of a temporary nature [11, 12]. In Africa, these substances are not always available to all populations or to their financial purse. Many opportunities can be offer to solve this problem like some of our research has turned to phytotherapy [13]. This is the reason why *Hyptis specigera* Lam was chosen. The plants of *Hyptis* genus belong to Lamiaceae family. They have many virtues and uses, especially for medicinal and culinary purposes. Among the plants of this family, the species *Hyptis specigera* lam has the plural medicinal virtues [14]. It also has anxiolytic properties [15]. Several studies have demonstrated the anxiolytic effects of plants and physical activity individually, but few are those that have been interested in their combined effects. The aim of this work is to evaluate both the combined anxiolytic effects of the decoction of dried leaves of *Hyptis specigera* Lam and physical activity in white mice *Mus Musculus* Swiss

2. Material and methods

2.1. Material

2.1.1. Plant Material

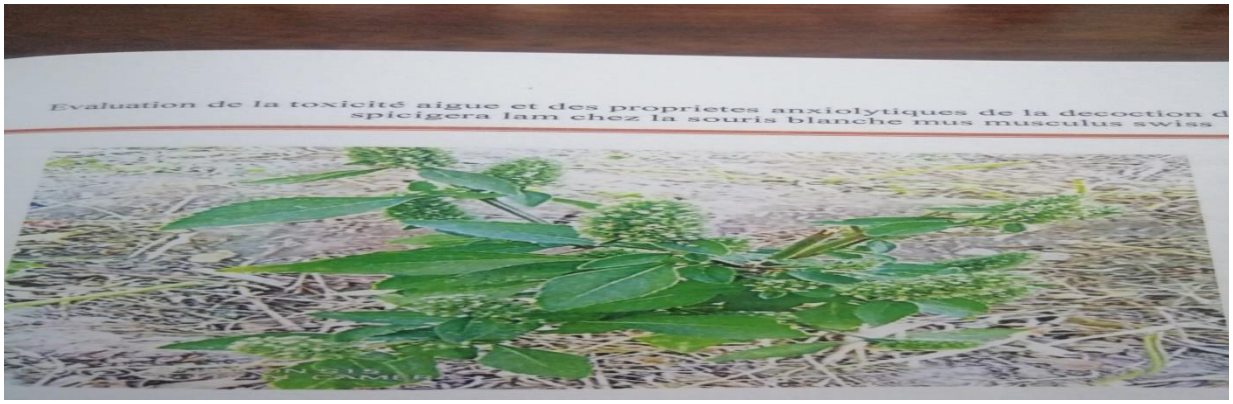


Figure 1 *Hyptis spicigera* Lam (Omam, 2023)

The leaves of *Hyptis spicigera* Lam. were collected in the far north of Cameroon, precisely in the Diamare Division, Maroua district and Zokok Laddeo village. The identification of the plant was carried out at the national herbarium of Cameroon in Yaounde. Sample identified in comparison with Letonzey's material R 10908 of the specimen of the Herbarium collection n° 28063 SRF/Cam.

2.1.2. Animals

Male and female *Mus musculus* Swiss mice with no previous experiments were used in our study. The weight of these mice averaged approximately 19 g and above. These mice were collected from the animal house of the Department of Animal Biology of the Faculty of Science of the University of Yaoundé 1 and were transported to the laboratory of physiology of animal organisms of the Higher Teacher Training College of the same University. There, they had a period of acclimatization to the conditions of laboratory environment. They had also free access to tap water and food. Their cages were cleaned every 2 days. The temperature in the animal experimentation room was 24°C. The lighting was artificial and the nycthemera respected with sequences of 12h of lighting and 12h of darkness. Concerning the determination of the anxiolytic activity, we used randomly male and female mice of 9 weeks of age.

2.1.3. Chemical

The DZP retained for the preparation of used solutions was contained in ampoules of 10 mg/2mL, which involved a concentration of 5 mg/ml under the conditions where the administration volume was fixed at 10 ml/kg.

2.2. Methods

2.2.1. Preparation of decoction

Fresh leaves of *Hyptis spicigera* Lam were washed with tap water, cut and dried in the shade. A decoction of these leaves was prepared by introducing 5 g of plant powder into a beaker containing 50 ml of distilled water. After the beaker closed, the mixture was boiled on a hot plate for about 20 minutes. Then, the mixture was cooled, it was filtered through a number 3 Wattman paper to recover a liquid called stock solution. This is the solution to be administered to the animals.

2.2.2. Preparation of diazepam

For the preparation of the different concentrations of DZP that were used in the chronic behavioural tests, knowing that the final concentration needed was 0.2 mg/ml, a volume of DZP of 1 ml was taken and introduced into the beaker, completed with distilled water to 25 ml for a dose of 2 mg/kg. This solution was prepared for all chronic stress tests. Concerning the preparation of the concentration of DZP which was used to OF test, knowing that the final concentration needed is 0.3 mg/ml, it was taken a volume of DZP of 1 ml and introduced in the beaker then, supplemented with distilled water to 16,6 ml for a dose of 3 mg/kg.

2.2.3. Experiment Procedure: Chronic Immobilisation Stress Test

Chronic anxiety was induced in the animals by immobilising them in a narrow tube each day for 2 hours for 10 consecutive days. The mice were divided into 7 groups of 5 mice each. Group 1, also called the normal group, received distilled water; group 2 (negative control) received distilled water and done stress induction and physical activity; group 3 received the plant decoction at 22 mg/kg and done stress induction and physical activity ; group 4 received the plant at 56 mg/kg, done stress induction and physical activity; group 5 received the plant decoction at 112 mg/kg done stress induction and physical activity, group 6 received the plant decoction at 225 mg/kg done stress induction and physical activity; and finally group 7 called the positive group received diazepam and done stress induction and physical activity. After that, animals have trained to show performances in the physical training programme. The swimming programme lasted 10 days. The training frequency was one session per day. The duration of the sessions varied from 5 to 12 minutes. The mice swam individually and continuously until the time limit was reached, then were taken out of the pool, dried with a suitable towel and returned to their cage. The water used for training was immediately replaced. The swimming was done between 3 p.m. and 6 p.m. The animals had free access to food and water after the day experiment. After these different treatments, the mice were placed one after the other on the elevated plus maze test and the open field test. [16].

2.2.4. Elevated Plus Maze Test (EPM)

According to Elhage in 2012 [17], the EPM test is a cross-shaped device raised 65 cm above the ground. It consists of four arms (L = 50 cm × W = 10 cm) facing each other; two of these arms are closed by 49 cm high sides while the other two are open. The arms are connected with a central platform (10 cm × 10 cm). This device is topped by a video camera

to record the animal's behaviour during the test. The mouse is placed in the centre of the device, facing an open arm, and left free to explore it for 5 min. The floor was cleaned with 10% ethanol between each mouse to avoid any odour that might affect the behaviour of the next animal. Several parameters were measured by analysis of the video recordings: the number of entries and the time spent in the open arms (OA) and closed arms (CA). An entry is counted when the animal's four legs cross the threshold of the arm. These data are used to calculate the time spent in the central platform, considered as an indicator of hesitation between approaching and avoiding the most anxiety-provoking arms, the total number of entries in all the arms, considered as a good indicator of the animal's locomotor activity, as well as the percentage of time spent in the open arms (OA) (time spent in OA*100/time OA + time CA) considered as reflecting the animal's anxiety state [17].

2.2.5. Open Field Test

The Open Field (OF) is a square enclosure with eleveted edges, illumited at the centre, which does not allow the animal inside to escape or hide. The exploration surface is divided into 17 tiles: 16 tiles dividing the interior surface of the experimental paradig and one central tile. The dimensions of the OF were 40 cm square and 19 cm high [18].The OF test is commonly used to assess locomotor activity, exploration and emotional reactivity in rodents. The mice were evenly divided into six groups of five animals each. These animals were treated with distilled water for the negative control, with different doses of *Hiptis spicigera* decoction (213 mg/kg; 106 mg/kg; 53 mg/kg; 21mg/kg; p.o.) for the test groups and diazepam (3 mg/kg; i.p.) for the positive control. After administration of the different substances, the mice were returned to their original cages to reduce neophobic responses due to the experimental environment [19]. One hour after the administration of the different substances to the mice, they were placed one after the other in the centre of the experimental paradigm. The behaviour of each mouse was observed and noted for a period of 5 minutes. Among parameters recorded, there were the number of crossings and the time spent in Centre. Also the number of crossings, groomings. After 5 minutes of observation, the mouse was returned to its original cage and the experimental device was cleaned with ethyl alcohol (70°C).

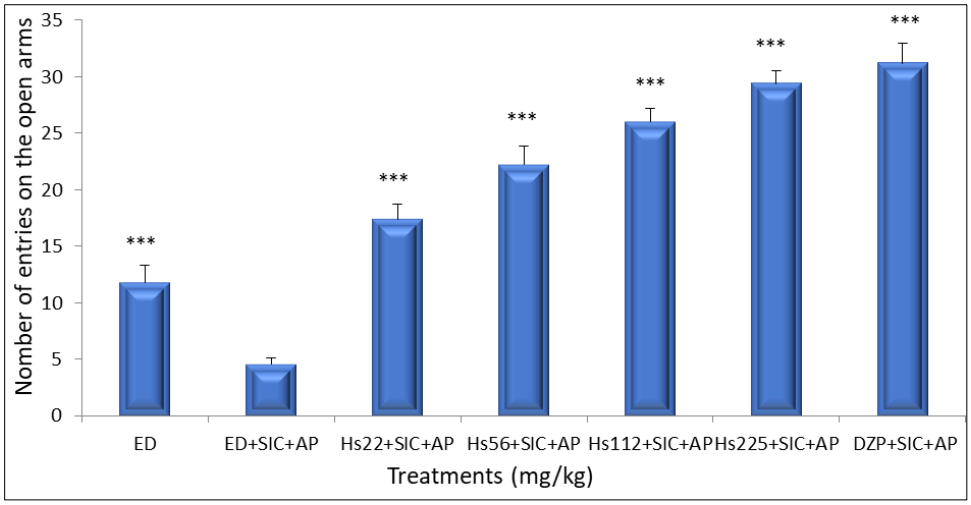
2.2.6. Statistical Analysis

Values were compared using the analysis of variance (ANOVA) test and where differences prevailed, Tukey's comparison tests (HSD) were used to separate them. From $p \leq 0.05$, the values were considered to be different, thus significant.

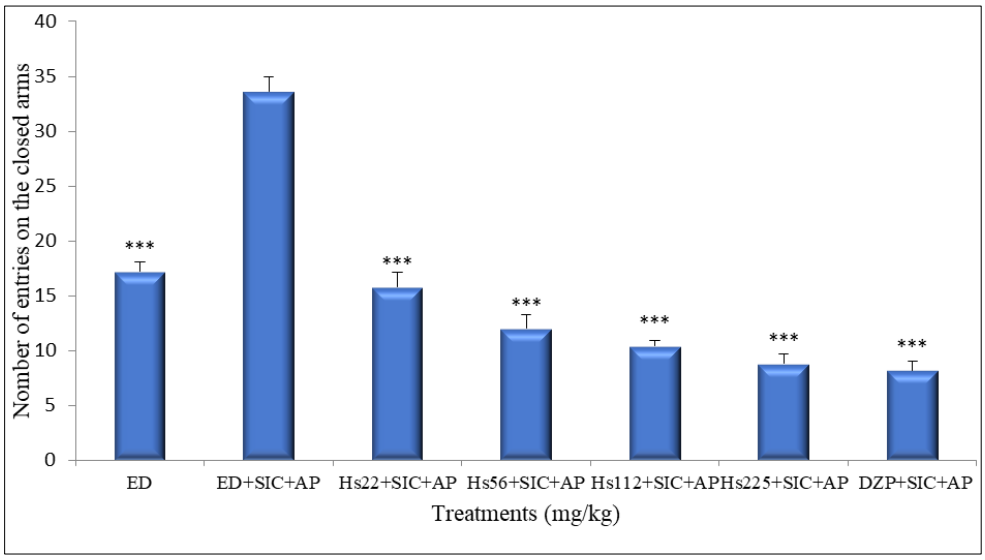
3. Results

3.1. Anxiolytic Effects of *Hyptis spicigera* Lam decoction and physical activity on the Elevated Plus Maze

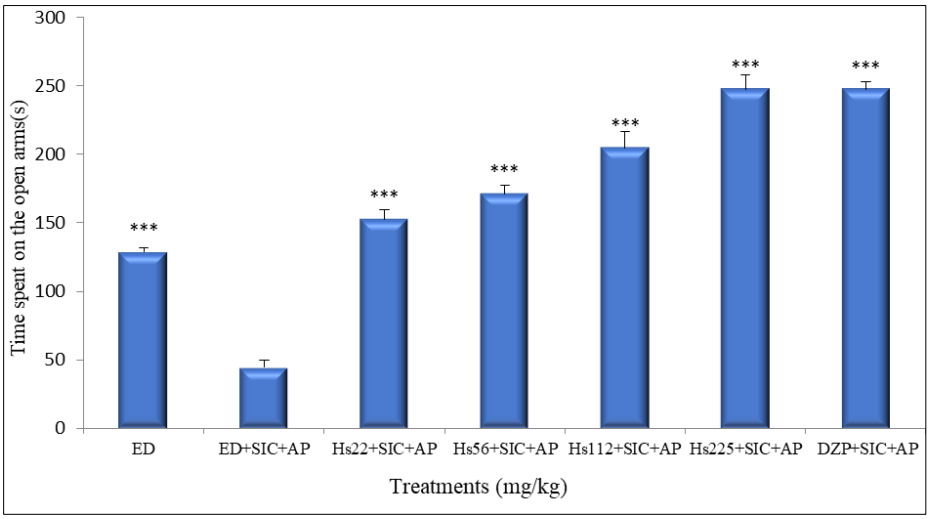
Fig2A showed that chronic stress (CIS) induced a significant ($p<0.001$) decrease of the number of open arm entries of mice in the normal group from 11.8±1.48 to 4.6 ±0.54 of mice in the negative control group. *Hiptis spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) increase of this number from 4.6±0.54 of mice in the negative control group to 29.4±1.14 of the Hs225mg/kg group. The diazepam also induced a significant ($p<0.001$) increase of this number. Conversely, fig2B showed that, the SIC induced a significant ($p<0.001$) increase of the number of entries into closed arms of mice in the normal group from 17.20 ± 0.84 to 33.60 ± 1.34 of mice in the negative control group. *H. spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) decrease from 33.60 ± 1.34 of the mice of the negative control group to 8.80 ± 0.84. The fig2C showed that CIS induced a significant ($p<0.001$) decrease of the time spent in the open arms of the normal group mice from 128.8 ± 3.27 s to 44.40 ± 5.18 s of the negative control group mice. *H. spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) increase of this time from 44.40 ± 5.18 s of the mice in the negative control group to 247.60 ± 10.78 s. In contrast, the fig2D showed that, the CIS induced a significant ($p<0.001$) increase of the time spent in the closed arms of the normal group mice from 171.20 ± 3.27 s to 247.60 ± 12.60 s of negative control group mice. *H. spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) decrease from 247.60 ± 12.60 s of the negative control group mice to 52.40 ± 10.78 s. On the other hand, the fig2E showed that, the CIS induced a significant ($p<0.001$) decrease of the percentage of time spent in the open arms of the normal group mice from 42.92 to 14.77% of the negative control group mice. *H. spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) increase of this precentage from 14.77% of mice in the negative control group to 82.53%. Also, fig2F showed that SIC induced a significant ($p<0.001$) decrease of the number of head dipping of mice in the normal group from 7 ± 1.22 to 2.60 ± 0.55 of mice in the negative control group. *H. spicigera* Lam decoction and physical activity induced a significant ($p<0.001$) increase from 2.60 ± 0.55 of the mice in the negative control group to 38 ± 1.00. The diazepam also induced a significant ($p<0.001$) increase of this number.


Nomber of entries on the open arms
35
30
25
20
15
10
5
0
***
ED
ED+SIC+AP
Hs22+SIC+AP
Hs56+SIC+AP
Hs112+SIC+AP
Hs225+SIC+AP
DZP+SIC+AP
Treatments (mg/kg)

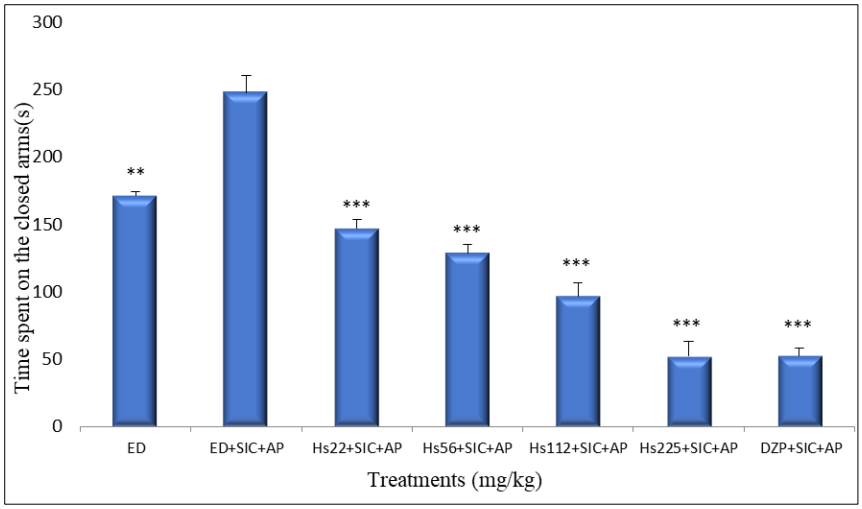
A


Nomber of entries on the closed arms
40
35
30
25
20
15
10
5
0
***
ED
ED+SIC+AP
Hs22+SIC+AP
Hs56+SIC+AP
Hs112+SIC+AP
Hs225+SIC+AP
DZP+SIC+AP
Treatments (mg/kg)

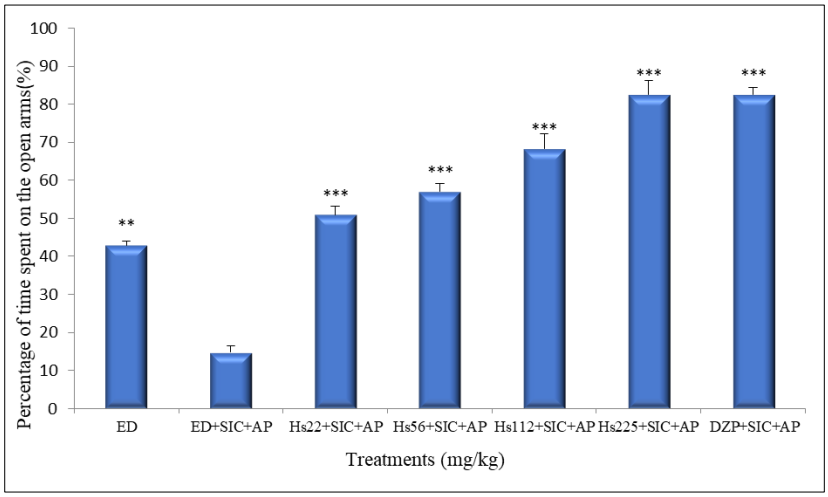
B


Time spent on the open arms(s)
300
250
200
150
100
50
0
***
ED
ED+SIC+AP
Hs22+SIC+AP
Hs56+SIC+AP
Hs112+SIC+AP
Hs225+SIC+AP
DZP+SIC+AP
Treatments (mg/kg)

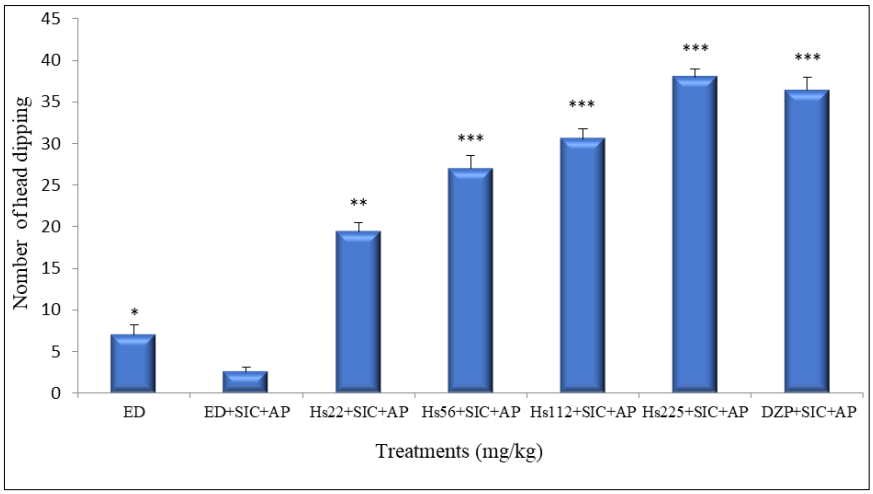
C



D



E

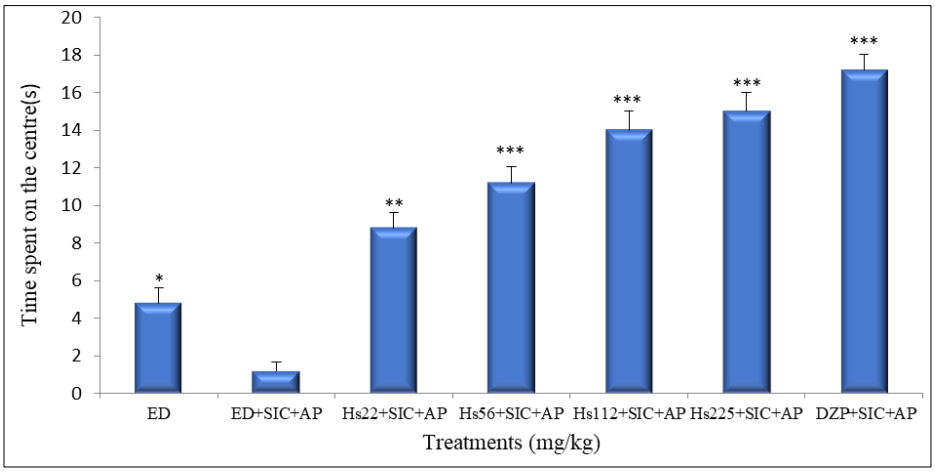


F

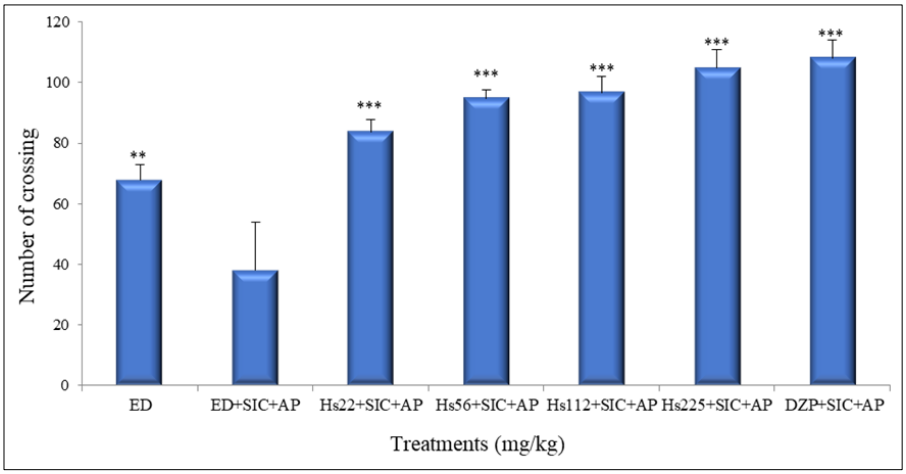
Figure 2 Anxiolytic effects and physical activity of *Hiptis spicigera* Lam decoction on EPM. A (on the number of entries on the open arms); B (on the number of entries on the closed arms); C (on the time spent on the open arms); D (on the time spent on the closed arms); E (Percentage of time spent on the open arms); F (Number of head dipping). Each bar represents the parameters of the EPM, n = 5. *p<0.05, **p<0.01, ***p<0.001; significant difference from negative control; ED: distilled water; SIC: chronic immobilisation stress, PA: physical activity, Hs22, Hs56, Hs112 and Hs225: *different doses of Hiptis spicigera*; DZP: diazepam.

3.2. Anxiolytic Effects of *Hyptis spicigera* Lam Decoction and Physical Activity on the Open Field Paradigm

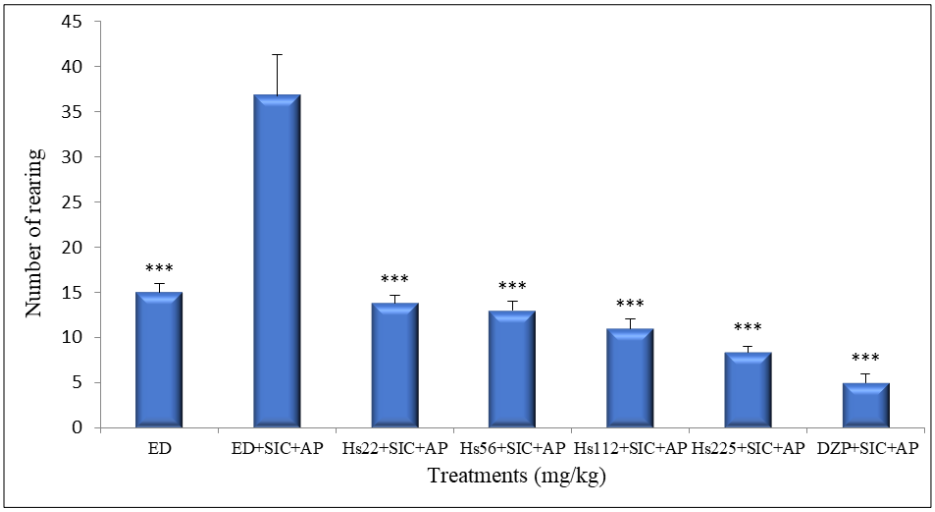
The fig3A showed that, the CIS induced a significant (p<0.001) decrease of the time spent in the center of the normal group mice from 4.8 ± 0.84 s to 1.2 ± 0.45 s of the negative control group mice. *H. spicigera* Lam decoction and physical activity induced a significant (p<0.001) increase from 1.2 ± 0.45 s of mice in the negative control group to 15 ± 1.00 s. The diazepam also induced a significant (p<0.001) increase of this time. Conversely, the fig3B induced a significant (p<0.001) decrease of the number of crossing of mice in the normal group from 67.8 ± 5.26 to 38 ± 16.02 of mice in the negative control group. The decoction of *H. spicigera* Lam and physical activity induced a significant (p<0.001) increase from 38 ± 16.02 of mice of the negative control group to 104.8 ± 6.10. Also, the fig3C showed that, the SIC induced a significant (p<0.001) increase of the number of rearng from 15 ± 1.00 to 36 ± 4.55 of the mice in the negative control group. *H. spicigera* Lam decoction and physical activity induced a significant (p<0.001) decrease of this number from 36 ± 4.55 of mice in the negative control group to 8.4 ± 0.55. Finally, the fig3D showed that, the SIC induced a significant (p<0.001) increase of mass of stools from 1.6 ± ± 0.55 to 5.2 ± 0.84 mg of mice in the negative control group. *H. spicigera* Lam decoction and physical activity induced a significant (p<0.001) decrease from 36 ± 4.55 of mice in the negative control group to 0.4 ± 0.55. The diazepam also induced a significant (p<0.001) increase of this mass of stools.



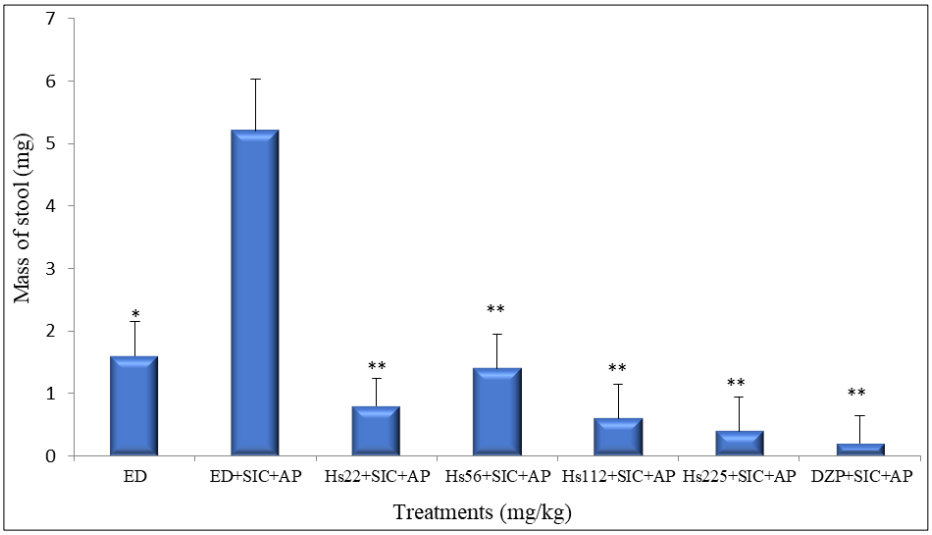
A



B



C



D

Figure 3 A (on the time spent on the centre); B (on the number of crossing); C (on the number of rearing); D (on the mass of stools). Each bar represents the parameters of the OF, n = 5. *p<0.05, **p<0.01, ***p<0.001; significant difference from negative control; ED: distilled water; SIC: chronic immobilisation stress, PA: physical activity, Hs22, Hs56, Hs112 and Hs225: *different doses of Hiptis spicigera*; DZP: diazepam.

4. Discussion

The elevated plus maze test is the best test for assessing the anxiolytic effects of substances. The behavioral analysis of mice treated with different doses of the extract and diazepam (3 mg/kg) combined with physical activity revealed an increase of the number of head dipping, the number of entries, the time spent and the percentage of time spent in the open arms, while a decrease of the number of entries and the percentage of time spent in the closed arms was observed. This result are explained that, the mouse has decreased his fear and has reduced also his anxiety [20,21]. All these results show that the decoction of *Hyptis spicigera* Lam and physical activity would possess the properties through certain hypotheses that explain the action mechanism [22, 23]. The GABAergic pathway could be involved through the GABA-A receptor complex where the decoction associated with physical activity could be carried by the benzodiazepine or barbturic site and of course the other receptor sites of this complex. These results are in agreement with those of Grundman or Venault [24, 25] whose thought that the fixation of the compounds of the plant decoction at this level would cause the increase of the duration of the opening of the chloride channels thus leading to the membrane hyperpolarization due to an increasingly important flow of chloride ions in the cell. Our results therefore showed that the combined effect of *Hyptis spicigera* Lam and physical activity would further provoke this increase. Subsequently, the anxiolytic effects observed could also be caused, according to the chronic immobilization test, by the stimulation of the

hypothalamo-hypphys-adrenal (HPA) axis causing the production of biogenic amines that promote the release of glucocorticoids responsible for the anxiety state [26]. However, in view of the results, the combined effect of the plant and physical activity would have prevented or reduced the production of corticosterone, which is a marker of anxiety via the serotonergic pathway [27,28]. Also, the OF test, is commonly used to assess locomotor activity, exploration level and emotional reactivity in rodents [29,30]. This situation also allows highlighting the anxiolytic or anxiogenic properties of pharmacological compounds. The results obtained in the OF test showed that the decoction of *Hyptis spicigera* lam and physical activity showed an increase of the time spent in the center and the number of crossing. . The increase of these parameters is a manifestation of the increase of locomotor activity and exploration level of the mouse [20,21]. In the open field test, this increase of activity in the rodent indicates a reduction of anxiety [21]. In contrast, a decrease of the number of rearing and the mass of stools was observed. The reduction of these parameters reflects the reduction of anxiety in these mice by the solicitation once again of the Gaba receptor complex via benzodiazepine or barbiturate sites, either by the hypothalamic-hypphys adrenal (HPA) axis or to a lesser extent the NMDA receptors of the Kainate

5. Conclusion

There is realized finally that, the plant decoction possessed the anxiolytic effects and physical activity in mice with the CIS test both EPM and OF paradigms. Because we observed a significant increase of some classical behavioral parameters such as the number of entries and the time spent in the open arms and their percentages. As well as a significant decrease of the number of entries and the time spent in the closed arms of these mice was observed. All of these results showed that the decoction associated with physical activity significantly improves the state of mice previously judged anxious. Better still, it also improved the physical performance of these animals. Knowing that *Hyptis spicigera* lam is used empirically, it is desirable that this plant in association with physical activity be advised to sedentary, elderly and even young people who are overweight.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare that there is no conflict of interest. No thanks.

References

[1] Gabarrell-Pascuet A, Garcia-Mieres H, Giné-Vazquez L, Moneta MV, Domènech-Abella J. (2023). The Association of Social Support and Loneliness with Symptoms of Depression, Anxiety, and Posttraumatic Stress during the COVID-19 Pandemic: A Meta-Analysis. International Journal of Environmental Research and Public Health. 20: 1-25.

[2] American Psychiatric Association. Diagnostic and statistical manuel of mental disorders (2000). 4TH ed. Text Rev.Washington DC: American Psychiatric Association

[3] Necho M, Tsehay M, Birkie M, Biset G, Tadesse E. (2021). Prevalence of Anxiety, Depression and Psychological distress among the general population during the COVID-19 pandemic a systematic review and meta-analysis. International Journal of Social Psychiatry. 67(7): 892-906.

[4] Canada psychiatrisants association (2006).Clinical practical guide. Curing of anxious disorders. Can J Psychiatry; 51 (suppl.2): IS-96

[5] Pelissolo A. (2012). Anxious and nevrotic disorders. EMC of Psychiatry, S1634-6939(12)4804-1

[6] Gramond J. (2018). What is interest of sport for take care of anxious disorders of depressive patients? PhD, University of Limoges, France

[7] Steptoe A. Kearsley N,Waltyers N, (1993). Cardiovascular during mental stress following vigorous exercise in sportsman and inactive men.Psychophysiology vol.30p. 245-252.

[8] Brunner E,Trippolini M,Dauru A,Egloft N,Schleifer R,Vancampfort D (2021). Physic activity against the depression and anxiety forum Med Swizland doi:10.4414/fms.08672

[9] Taylor A, (2000). Physical activity,anxiety and stress in:Biddle S,Fox K, Boutcher S editors, Physical activity and psychological well-being, London: routledge p 10-45

[10] Omam Omam, JP. Study of the anxiolytic properties of the decoction of dried barks of Parkia biglobosa (Minmosacaceae) in the white mouse Mus musculuss Swiss (Muridae) (Doctoral thesis/Ph.D in pharmacology of medicinal plants). University of Ngaoundéré, Cameroon; 2018.

[11] Handa SS, (1995). Plants and plant products for mental health. In: Koslow SH,Murthy RS,Coelho GV Eds. Decade of the brain, US Department of health and human Services,pp.163-171

[12] Thakur VD, Mengi SA, (2005). Neuropharmacological profile of Eclipta alba (Linn.) Hassk. Journal of Ethnopharmacology 102 23-31

[13] Nanga L D,Ayissi M ,Moto OFC,DIMO T, Ngo Bum E, (2019). Anxiolytic properties of Garcinia lucida Vasque (Clusiaceae) in mice and its possible action mechanisms. The Pharmaceutical and chemical Journal 6(5):58-72

[14] Roli RS, Igor DSC, Stella CJ, Glauce RP, Marcos JS, Adair RSS, Felipe MDF. (2017). Oral treatment with essential oil of Hyptis spicigera Lam. (Lamiaceae) reduces acute pain and inflammation in mice: Potential interactions with transient receptor potential (TRP) ion channels. Journal of Ethnopharmacology. 200: 8-15.

[15] Dang Bouba K (2021). Evaluation of the acute toxicity and anxiolytic properties of the decoction of Hiptis spicigera lam leaves in the white mouse Mus musculus Swiss. Thesis of Pharmacy at the University of Douala, Faculty of Medicine and Pharmaceutical Sciences, Cameroon.

[16] Omam OJP, Hamadou A, Mbouh S, Koube J, Dang B K, Ze MMD, Moto O FC,Ngo Bum E(2022). Anxiolytic and Antioxydant Effects of Aqueous Extract of Hiptis spicigera Lam in Mice Exposed to Classical Paradigms and Chronic Immobilisation Test. Clinical Neurology and Neuroscience.Vol.6,No.4, pp.50-61.doi: 10.11648/j.cnn.20220604.11

[17] Omam OJP, Samuel Mbouh, Mbouh S, Kandeda AK, Koube J, MoussaDM, Ze MMD, Bidingha à G R, Mbom A, Moto OCF, Ngo Bum E. Anxiety Curing Evaluation with theAqueous Extract of Securidaca longepedunculata (Polygalaceae) Decoction in Mice on the Stress Paradigm Tests. American Journal ofPsychiatry and Neuroscience. Vol. 11, No. 1, 2023, pp. 1-12. doi: 10.11648/j.ajpn.202311014.11

[18] Simon Pale, Sidiki Neteydji, Germain Sotoing Taiwe, Nadège kouemou Emegan, Elisabeth Ngo Bum. (2021). Anticonvulsant effects of cymbopogon giganteus extracts with possible Effects on fully kindled seizures and anxiety in experimental rodent model of mesio-temporal Epilepsy induced by pilocarpine. Journal of Ethnopharmacology. 286(2): 114863.

[19] J. P. O. Omam, R. E. A. Mbomo, A. K. Kavaye, M. D. Z. Minkoulou, S. J. N. Kameni, F. C. M. Okomolo, E. N. Bum,(2017): GABA-A Receptor Complex in the Anxiolytic Properties of Parkia biglobosa in Mice. International Journal of Brain and Cognitive Sciences . 6 (2): 26-3

[20] Ngo Bum E, Taiwe GS, Moto FCO, Ngoupaye GT, Nkantchoua GCN, Pelanken MM, Rakotonirina SV, Rakotonirina A. (2009a). Anticonvulsant, anxiolytic and sedative properties of Nauclea latifolia Smith roots in mice. Epilepsy Behaviour. 15: 434-440.

[21] Njapdounke JS, Moto FC, Taiwe SG, Neteydji S, Pale S, Ayissi RE, Ngo Bum E. (2016). Anxiolytic like properties of Hallea ciliata in mice chez la souris. African Journal of Traditional Complementary and Alternative Medicine Journal of traditional complementary and alternative medicine. 13 (4): 1-7.

[22] Mozhui K ,Karlsson RM, Kash TL,Ihne J,Norcross M, Patel S,Farrell MR (2010). Strain differences in stress responsivity are associated with divergent amygdala gene expression and glutamate-mediated neuronal excitability. J of neuroscience 30: 357-367.

[23] Ayissi Mbomo RE,Ngo Bum E, Tene Tadoum SB (2019). Effects of Aqueous Extract of Garcinia lucida on chronic stress Induced Depression-like Behaviours in Mice. International Journal of Sciences and Reasearch Methodology. Vol:13, Issue: 4

[24] Grundman O.,Nakajima JI,Sea s.Butterweek V. (2007). Anti-anxiety effect of Apocynum venetum L. in the elevated plus maze test. Journal of Ethnopharmacology,110: 406-11

[25] Venault, P. and Chapouthier, G., (2007).. From the Behavioral Pharmacology of Beta-Carbolines to Seizures, Anxiety, and Memory.Life Sci. 39:1093–1100.

[26] Omam Omam, JP. Étude des propriétés anxiolytiques de la décoction des écorces séchées de Parkia biglobosa (Minmosacaceae) chez la souris blanche Mus musculuss Swiss (Muridae) (Thèse de doctorat/Ph.D en pharmacologie des plantes médicinales). Université de Ngaoundéré, Cameroun; 2018.

[27] Guezemec C.,Abdelmalki A.,Serrurier B.,Merino D.,Bigarg X.,Berthelot M.. (1998) Effects of prolonged exercise on brain ammonia and amino acids. International Journal of sports Medicine, vol 19,p. 323-327

[28] Medadom,when the sport become the mental support(2021)

[29] Bronikowski, A.M., Carter, P.A., Swallow, J.G., Girard, I.A., Rhodes, J.S., Garland, T.J., (2001). Open-field Behavior of

[30] Seillier, A., (2003). Livret des techniques, IFR des Neuroscience de Strasbourg, Université Louis Pasteur, laboratoire de Neurosciences comportementales et cognitives. Edition Tournier B., et Revel F., 246, 52 – 89.