

# Preliminary evaluation of the larvicidal activity and phytochemical study of aqueous extracts of eleven plants of the Niger biodiversity on *Anopheles* L2 larvae.

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## Abstract

Malaria is a preventable and curable disease. To prevent this disease, the National Malaria Control Program (PNLP) has distributed insecticide-treated mosquito nets and seasonal chemoprevention in recent years. However, we are witnessing the development of resistance of the mosquito vectors and of the plasmodium causal agent of the disease towards the active substances used. The objective of our work is to contribute to the research of new active substances effective in the control of malaria mosquito vectors. The larvicidal activity and the phytochemical composition of eleven plants of the Niger biodiversity have been studied on *Anopheles* stage 2 larvae. The methodology used was aqueous extraction from plant powders and qualitative identification of the main families of secondary metabolites. The toxicity of the extracts was tested in the laboratory according to the WHO protocol. The dead larvae were collected after 24 and 48 hours. The phytochemical analysis revealed the presence of the main families of secondary metabolites such as alkaloids, flavonoids, poly terpenes sterols, quinones, tannins and phenolic compounds. Larvicidal tests revealed that *Balanites aegyptiaca*, *Anacardium occidentale* and *Khaya senegale* induced 100% larval mortality after 24 hours. *Annona senegalensis*, *Tamarindus indica*, *Sclerocarya birea* and *Carica papaya* induced 100% mortality after 48h. Moreover, the analysis of the regression lines showed that the LD50 obtained with *Balanites aegyptiaca* is 1.02 mg/l with *Anacardium occidentale* it is 1.23 mg/l while it is 1.62 mg/l with *Khaya senegale* after 24h. These low values of LD50 lead us to propose the aqueous extracts of *Balanites aegyptiaca*, *Anacardium occidentale* and *Khaya senegale* in larviciding campaigns against mosquitoes.

**Keywords:** larvicide, mosquito, secondary metabolite, phytochemical, mosquito control

## 1-INTRODUCTION

Mosquitoes (Diptera, Culicidae) are a family of insects of considerable public health importance. In Africa, they are a nuisance and can also represent a public health risk. They are vectors of numerous arboviruses, protozoa and metazoa, agents of diseases including malaria (Messai, 2010; Robert, 2012). Malaria is a potentially fatal parasitic disease caused by a protozoan of the genus *Plasmodium* (WHO, 2014). Malaria is endemic in Africa and represents a major public health problem (Sylla *et al.* 2018). According to the annual report of the World Health Organization, Africa is the most affected continent. Ninety-five percent (95%) of malaria cases reported in 2020 were in the African region (OMS, 2021). In this region, it is not only a health disaster; it also represents a major obstacle to the economic and social development of the continent (Sani, 2013). Vector control, through the use of insecticides directed against mosquitoes, has contributed to the reduction of malaria incidence worldwide. It is also the main means of preventing and reducing malaria transmission (Rubert *et al.* 2016). In mosquito control campaigns, the active ingredients of insecticides used belong to the organophosphates, pyrethroids and synthetic carbamates. These preparations, although

they have proven to be very effective on culicid mosquitoes, have several disadvantages. Indeed, the significant accumulation of active ingredients in treated aquatic and terrestrial ecosystems is a pollution problem. In addition, the active substances of the products used have a broad spectrum of action and do not spare non-target organisms (Barbouche *et al.* 2001; Akono *et al.* 2017). If the coverage by vector control interventions is high enough in a given area, the whole community could be protected (WHO, 2018). Despite numerous efforts by the scientific community to reduce the prevalence of malaria to its lowest level, there has been a resurgence of this disease for several decades. This could be linked to the approximate application of the current preventive and curative methods, to economic problems (high costs of antimalarials and insecticides) and especially to the resistance of the plasmodium and the vector to antimalarials and synthetic insecticides respectively (Biboga *et al.* 2007; Akono *et al.* 2012; Mahaman *et al.* 2021). To contribute to sustainable environmental management, new methods of mosquito control are further encouraged. Natural substances that have a wide spectrum of action in pharmacology could be used in vector control. The use of plant extracts as insecticides has been known for a long time, as pyrethrum, nicotine and rotenone are already known as insect control agents. In some parts of Africa, tobacco leaves mixed with water were used to control mosquitoes (Crobey *et al.* 1966; Aouny *et al.* 2006). In Niger, studies on the insecticidal activity of plant extracts against mosquito larvae are very limited. Indeed, the work of (Yolidjé *et al.* 2016) on Anopheles larvae has provided promising results in the context of vector control. The objective of our study is to contribute to the search for new plant molecules with insecticidal effects. To this end, an inventory of antimalarial plants was drawn up, the larvicidal activity and the phytochemical composition of these plants in the biodiversity of Niger were determined.

## **2. MATERIALS AND METHOD**

### **2.1 Study area**

The city of Niamey is located at 13°31 north latitude and 2°6 east longitude. With an area of 255 km<sup>2</sup>, it is built on two plateaus overlooking the Niger River, at an altitude of 218 m. The city of Niamey is divided in two by the Niger River. The most important part is on the left bank, where the Gounti-Yéna valley is located. On the right bank, also known as Harobanda, the vast majority of the city's rice fields are located. The city of Niamey is subdivided into 5 communal districts. Niamey I, II, III, and IV are located on the left bank of the river, and Niamey V, the area of this study, is located on the right bank, which lies on a plain with an average altitude of 185 meters. There are flood zones below 182 m. The marshy areas made up of clay soils are often developed by hydro-agricultural projects, as is the case in Saguia and Kirkissoye Mamadou *et al.* 2008).

### **2.2 Animal material**

The animal material used for the toxicological study is composed of Anopheles larvae. These larvae were collected in rice fields located in Saguia in the Niamey V district. They were collected using a fine mesh strainer and a transparent bucket containing water from the site. In the laboratory, the larvae were transferred to transparent plastic containers containing water from the bed and left to rest for 24 hours before testing. The larvae are fed with cat food.

### **2.3 Choice of plants**

The plants used belong to the biodiversity of Niger. They were inventoried following an ethnobotanical survey carried out among traditional practitioners and their traditional uses by the local population in the fight against malaria (Table 1). This survey was conducted in the localities of Tillabéry and Niamey.

**Table 1: Anti-malarial plants surveyed**

Plant	Local name	Family name
<i>Annona senegalensis</i>	Moufa, gwadda	annonaceae
<i>Anacardium occidentale</i>	kade	anacardiaceae
<i>Bauhinia rufescens Lam</i>	Namari, dirga	caesalpiniaceae
<i>Carica papaya</i>	Papaye gna	caricaceae
<i>Balanites aegyptiaca (L.) Del.</i>	Garbey, adua	zygophylliaceae
<i>Grewia villosa</i>	say bombera	tiliaceae
<i>Khaya senegalensis (Desv.) A. Juss.</i>	farey	meliaceae
<i>Manguifera indica L.</i>	mangu gna	annacardiaceae
<i>Manihot esculenta crantz</i>	rogo	
<i>Sclerocarya birrea (A. Rich) Hochst</i>	diney	anacardiaceae
<i>Tamarindus indica L.</i>	bossey gna	cesalpiniaceae

## 2.4 Plant harvesting and aqueous extractions

### 2.4.1 Collection of the plants

The leaves of the plants were collected in the forest camp "Amoul Kinni" of Niamey and on the territory of the Abdou Moumouni University of Niamey.

### 2.4.2 Identification of the plants

The specimens were identified at the Biology Department of the Faculty of Science and Technology of the Abdou Moumouni University of Niamey.

### 2.4.3 Drying of plants

The harvested plant material (leaves) was dried on mulches in the entomology laboratory of the Abdou Moumouni University of Niamey for 48h to 96h.

### 2.4.4 Grinding of plants

The plant leaves were ground into powder using an electric grinder with an integrated 1mm diameter sieve, until they were reduced to powder at the Institut National de Recherche Agronomique (INRAN) photo 1.



Photo 1: Electric grinder

## 2.5 Preparation of aqueous extracts (decoction, filtration and evaporation)

After grinding the plants, 25 g of powder of each plant was introduced into a flask of 500 milliliters capacity containing 250 milliliters of distilled water and boiled for one hour photo 2. After cooling, the mixture obtained was filtered using cotton and a fine mesh sieve photo 3. The operation was repeated three times. The recovered filtrate is brought to the sand bath set

at 40°C until total evaporation of water photo 4. The product obtained is weighed and introduced into a bottle.



Photo 2: decoction in 3: filtration balloon heater Photo



Photo 4: sand bath



### 2.5.1 Determination of the yield

The yield of extraction is expressed as a percentage of the mass of plant powder used according to the formula:

$$\text{Yield} = \frac{\text{mass of extracted}}{\text{mass of initial powder}} \times 100$$

### 2.5.2 Preparation of solutions for toxicity tests

Stock solution of the aqueous extracts were obtained by dissolving 5,4 g of dry extract in 540 milliliters of distilled water. A few preliminary tests allowed us to select a range of concentrations for the test solutions. The water of the deposit was used for the preparation of the test solutions of the aqueous extracts.

The stock solution is prepared in such a way that all volumes to be determined are present. To obtain the initial volume for each final concentration, we proceeded by the following method:

$$C_i V_i = C_f V_f \rightarrow V_i = C_f V_f / C_i$$

Let an initial concentration  $C = 10 \text{ g/l}$  and the final volume,  $V_f = 100 \text{ ml}$ .

### 2.5.3 Phytochemical screening

Phytochemical screening was performed using standard characterization methods described by Ciulei in 1982; Wagner and Bladt in 1996 with some modifications. These reactions are based on precipitation phenomena or staining by specific reagents.

These detection tests were performed on the following families of chemical compounds: sterol terpenes, saponosides, flavonoids, tannins, phenolic compounds and alkaloids.

- Sterols and polyterpenes were sought by the Liebermann reaction. Five (5) ml of each extract was evaporated on sand bath. The residue was dissolved hot in 1 ml of acetic anhydride; 0.5 ml of concentrated sulfuric acid was added to the triturate. The appearance at interphase of a purple or violet ring, turning blue and then green, indicated a positive reaction.
- The ferric chloride ( $\text{FeCl}_3$ ) reaction was used to characterize the polyphenols. To 2 ml of each extract, one drop of 2% alcoholic ferric chloride solution was added. The appearance of a more or less dark blue-black or green coloration was the sign of the presence of polyphenols.
- Flavonoids were sought by the cyanidin reaction. Two (2) ml of each extract was evaporated and the residue was taken up in 5 ml of 2-fold diluted hydrochloric alcohol. On addition of 2 to 3 chips of magnesium, there is a heat release and then a pink-orange or purplish coloration. The addition of 3 drops of isoamyl alcohol intensified this coloration which confirms the presence of flavonoids
- Catechic tannins were tested using Stiasny's reagent. Five (5) ml of each extract was evaporated to dryness. After addition of 3 ml of Stiasny's reagent to the residue, the mixture was heated in a water bath at 80°C for 30 min. The appearance of a coarse flake precipitate



indicates the presence of catechic tannins. For gallic tannins, the previous solution was first filtered; the filtrate was collected and saturated with sodium acetate. Three (3) drops of FeCl<sub>3</sub> were added; the appearance of an intense blue-black coloration indicates the presence of gallic tannins.

- Quinonic substances were sought using Bornstraëgen's reagent. Two (2) ml of each extract was evaporated to dryness. The residue was triturated in 5 ml of 1/5 hydrochloric acid. The triturate was poured into a test tube. The triturate was then heated in a water bath for 30 min. After cooling, it is extracted with 20 ml of chloroform. Ammonia diluted 2 times (0.5 ml) was added to the chloroform solution. A red or purple coloration indicates the presence of quinones.
- Alkaloids were characterized using Burchard (iodine-iodide reagent) and Dragendorff (potassium iodo-bismuthate reagent) reagents. Six (6) ml of each extract was evaporated to dryness. The residue was taken up with 6 ml of 60° alcohol. Two (2) drops of Dragendorff's reagent were added to the alcoholic solution, the appearance of a precipitate or an orange coloration indicates the presence of alkaloids. Two (2) drops of Burchard's reagent were added to the alcoholic solution, the appearance of a precipitate of reddish-brown coloration indicates a positive reaction, the presence of alkaloids.
- For the saponoside test, 10 ml of the total aqueous extract was poured into a test tube. The well-sealed tube was shaken for 1 min and then left to stand for 15 min. A persistent foam height of more than 1 cm indicates the presence of saponosides.

### 3. Toxicity tests

The biological tests were performed on mosquito larvae, according to the WHO (World Health Organization) standardized test explained by [18] and [19] with some modifications. 25 Anopheles larvae were collected with a Pasteur pipette and introduced into Petri dishes containing 100 ml of the test solution. The dose ranges retained after the preliminary tests are: 4 g/l; 3.5 g/l; 3 g/l; 2.5 g/l; 2 g/l; 1.5 g/l; 1 g/l and 0.5 g/l, obtained from successive dilutions of the stock solution in the water of the lodge.

A negative control containing only the water of the deposit was constituted under the same conditions. For each test concentration and the control, the experiment was repeated 3 times. The counting of the larvae was carried out during 24h and 48h, of exposure to the various extracts photo 5.



Photo 5: Sensitivity test on Anopheles gambiae larvae

#### 3.1 Statistical analysis

In order to understand and model the results obtained were subjected to different statistical analyses. The percentage of mortality was calculated using the formula:

$$\text{percent mortality} = \frac{\text{number of dead larvae}}{\text{number of introduced larvae}} \times 100$$

When the mortality rate recorded in the controls is higher than 20%, the test is invalid. If the mortality of the controls is greater than 5% and less than 20%, the observed mortality is corrected by the Abbott (1925) formula:

$$\text{percentage mortality} = \frac{\text{mortality of treated larvae} - \text{mortality of controls}}{100 - \text{mortality of controls}} \times 100$$

If the mortality at the control level is less than 5% it can be accepted and does not require correction.

For the determination of LD50, the method used is the log-probit method of determining the dose corresponding to a given proportion carried out on the basis of linear regression on the Excel software.

The mortality means were processed and compared at the threshold of P = 0.05. Statistical analysis was performed using R software version 4.0.3.

## 4. RESULTS AND DISCUSSION

### 4.1 Yields, aspects and colors of aqueous extracts

In this study, 25 grams of leaf powder from each of the eleven plants were used (Table 2). After decoction, the color, appearance and yields obtained varied from one plant to another. The best yields were obtained from the extracts of *Annona senegalensis* 51.92% with a pasty aspect and brown color. On the other hand *Moniho esculentus* with the same characteristics, provided a yield of 31.84%. *Balanites aegyptiaca* and *Anacardium occidentale* provided yields of light-yellow color 38.64% pasty appearance and 9.2% powdery appearance respectively. Yields of powdery appearance and red-orange color were obtained with *Khaya senegalensis* 38.56% and *Bauhinia rufescens* 23.88%. *Sclerocarya birea* gave a yield of 34.76% with yellowish-red color and powdery appearance. Of powdery appearance *Carica papaya* has a yield of 30.72% with an olive-green color while *Tamarindus indicat* of the same appearance has a yield of 33.48%. The differences observed may be due to the nature of the constituents which differ from one plant to another and also to the living environment of these different plants. Aqueous extraction is a method which allows to obtain the majority of the large families of chemical compounds constituting a plant with relatively high yields. At this level our results are identical to those obtained in Niger Yolidjé et al. (2019) and Mali (Brehima, 2008). Several factors such as climate, ecology, extraction methods and storage conditions could explain the difference in yields obtained. This hypothesis is confirmed by several authors (Akono et al. 2012; Bauhini et al. 2021; Adini et al. 2018; Boudjema et al. 2021) who worked on the same plant: *Melissa officinalis* L. which provided different yields. Similarly, the appearance and color of the extract depend on the nature of the constituent minerals of the plant. Our results are identical to those obtained in Algeria (Eddine, 2018) and Niger (Yolidjé, 2019) on extracts of aerial parts of plants.

**Table 2: yields and aspects of aqueous extracts**

Plants	Yields %	Colour	Appearance
<i>Anacardium occidentale</i>	29,2	Bright yellow	Powder
<i>Annona senegalensis</i>	51,92	Brown	Pasty
<i>Balanites aegyptiaca</i> (L.) Del.	38,64	Bright yellow	Pasty
<i>Bauhinia rufescens</i>	23,88	Orange red	Powder
<i>Khaya senegalensis</i> (Desv.) A. Juss.	38,56	Orange red	Powder
<i>Carica papaya</i>	30,72	Olive green	Powder
<i>Grewia bicolor</i>	25,72	Red	Pasty
<i>Manguifera indica</i>	29	Red	Pasty

<i>Moniho esculentus</i>	31,84	Brown	Pasty
<i>Sclerocariya birea</i>	34,76	Yellowish red	Pasty
<i>Tamarindus indica</i>	33,48	Purple	Powder

#### 4.2 Phytochemical screening

The analysis of table 3 shows that polyphenols, flavonoids and sterols poly terpenes are present in the tested plants, that is 100% of the plants. Saponosides and quinones are present in 81.82% of the plants or 9 plants. Alkaloids and gall tannins are present in 63.63% of plants or 6 plants. As for catechic tannins, they are present in 54.54% of plants or 6 plants. These compounds constitute secondary metabolites and are found in all plants. Similar results have been reported by several authors (Al-snaif, 2015; Esseh *et al.* 2018; Kallo *et al.* 2018). The presence of alkaloids was also detected in the leaves of *Balanites aegyptiaca* (Hassane *et al.* 2020; Togolas *et al.* 2019). These substances have been detected in many plants such as *Khaya senegalensis*. Moreover, the therapeutic activity of a plant could be linked to the secondary metabolites it contains. The presence and quantity of these natural substances vary from one plant to another but also from the method of extraction (N’guessan *et al.* 2009). Indeed, the pharmacological utility of the plant extracts used in the present study has been reported by several authors. Thus, alkaloids are recognized for their antimalarial and antibacterial properties (N’guessan *et al.* 2009); insecticides (Arbonnier, 2002; Bruneton, 2014; Sané *et al.* 2018); antifungal (Romain, 2014). In addition, some authors (Badiaga, 2011; Hoekou *et al.* 2012; Mabika *et al.* 2013) have reported dyeing properties linked to the alkaloids constituting the secondary metabolites of plants. Polyphenols, flavonoids and polyterpenes sterols were identified in all our extracts. Obtaining a mortality rate of 100% after 48 days, we believe that these secondary metabolites have larvicidal properties. At this level our results are close to those of several authors (Raymond *et al.* 2013; Konno, 2011; Munôz *et al.* 2013) who have reported insecticidal properties of sterol terpenes. On the other hand (Soro *et al.* 2010; Zongo *et al.* 2011) have shown fungicidal and antimicrobial activities of terpenes and phenolic compounds. Inhibitory properties on the growth of many microorganisms including bacteria and fungi have been reported on tannins and flavonoids (Sepulveda *et al.* 2011). Polyphenols, flavonoids and terpenes are recognized for their antipyretic, analgesic and anti-inflammatory properties (Traoré *et al.* 2019). In addition, some authors (Pratp *et al.* Singh, 2013; Ghribia *et al.* 2014; Ahmed *et al.* 2014) have reported the inhibitory properties of AChE activity of phenolic compounds and flavonoids.

Catechic and gall tannins were totally absent in the extract of *Moniho esculentus* and *Sclerocariya birea*. These plants did not induce 100% mortality even after 48h. On the other hand, in the extract of *Balanites aegyptiaca* and *Khaya senegalensis*, we note the presence of gall tannin and catechic tannin respectively with 100% mortality after 24 hours of exposure of the larvae to the extracts. These results may explain the larvicidal activity of tannins in the catechic or gallic form. Identical results have been reported (Raymond *et al.* 2011) on the direct toxic effects of tannins on certain insect species and on the other hand (Vanderborre *et al.* 2011) have shown that tannins have an influence on the growth, development and fecundity of several insects. According to our results, quinones and saponosides may not have a direct effect on larval mortality. Indeed, the presence or absence of these secondary metabolites does not allow us to conclude objectively on their larvicidal effect. Thus, *Balanites aegyptiaca* induced 100% larval mortality within 24 hours while *Grewiya bicolor* did not induce 100% larval mortality, even after 48 hours of exposure to the extracts. On the other hand, in the absence of saponosides, *Anacardium occidentale* induced 100% mortality, 24 hours after exposure of the larvae to the extracts. *Carica papaya* induced 100% mortality of the larvae after 48h. In addition, antifungal activities of quinones have been reported (Diame, 2010).

**Table 3: Phytochemical screening of aqueous plant extracts**

Plantes	Polyphénols	Flavonoïdes	Stérols polyterpènes	Saponosides	Tanins		Quinones	Alcaloïdes
					Catéchiq ues	Galliqu es		
<i>Anacardium occidentale</i>	+	+	+	-	+	+	+	-
<i>Annona senalansis</i>	+	+	+	+	+	-	+	-
<i>Balanites aegyptiaca</i>	+	+	+	+	+	-	-	-
<i>Bauhinia rufescens</i>	+	+	+	+	+	-	+	+
<i>Carica papaya</i>	+	+	+	-	+	+	+	-
<i>Khaya senegalensis</i>	+	+	+	+	-	+	+	+
<i>Grewiya bicolor</i>	+	+	+	+	-	-	-	+
<i>Manguifera inica</i>	+	+	+	+	-	+	+	+
<i>Moniho esculentus</i>	+	+	+	+	-	-	+	+
<i>Sclerocariya birea</i>	+	+	+	+	+	+	+	+
<i>Tamarindus indica</i>	+	+	+	+	-	+	+	+

+ : certain presence, - : absence

#### 4.3 Larvicidal activity of aqueous extracts

After exposing L2 Anopheles larvae to different concentrations of aqueous extracts, a variation in mortality rate was observed. These mortalities vary according to time and concentration. Thus, after 24 h, *Balanites aegyptiaca*, *Khaya senegalensis* and *Anacardium occidentale* induced 100% mortality from doses that are 25 and 35 ppm respectively (Figure 1a). *Carica papaya*, *Moniho esculentus* and *sclerocariya birea* induced 98%, 95% and 68% mortality, respectively, while *Grewiya bicolor*, *Manguiefera indica* and *Bauhinia rufescens* induced mortalities below 50%. After 48 h, an increase in the mortality rate was observed for all plants and all concentrations. All plants induced 100% mortality except *Grewiya bicolor*, *Moniho esculentus*, *Bauhinia rufescens* and *Maguiefera indica* which induced respectively 93%; 97%; 80% and 87% mortality. From these results, a first classification of the toxic efficiency of the tested extracts towards Anopheles L2 larvae is highlighted. Thus, the most toxic extracts are those of *Balanites aegyptiaca*, *Caya senegalensis* and *Anacardium occidentale* leaves. These results show that there is a dose-effect relationship. The higher the concentration, the higher the mortality rate. These results also show that secondary plant metabolites can be used to control mosquito larvae that carry diseases. Similar results have been reported by several authors (Ebe *et al.* 2015; Abdullahi *et al.* 2011; Yolidjé *et al.* 2019) who have shown the efficacy of different plant extracts on mosquito larvae.



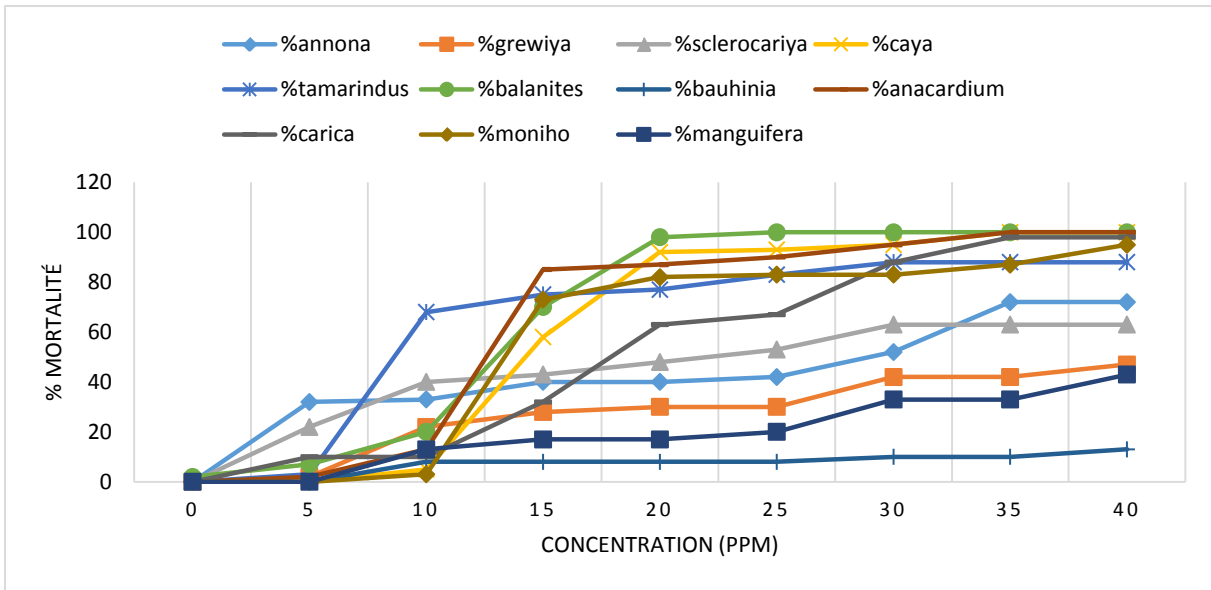


Figure 1 a: percentage of 24h mortality

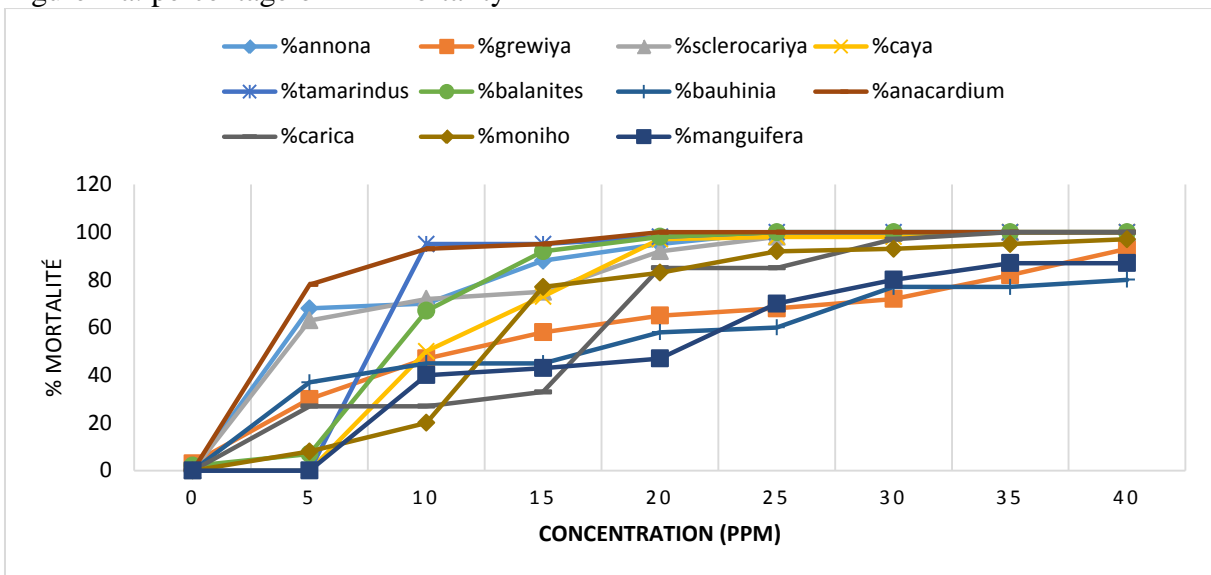


Figure 1 b: percentage of 48h mortality

Statistical analyses performed on larval mortality indicate a significant difference ( $H=0.87256$ ,  $P<0.05$ ) after 24 h and ( $H=0.79926$ ,  $p\text{-value} < 0.05$ ) after 48 h. This explains the differences observed in the mortality means (Table 5). As the mortality observed in the control lot was less than 5%, no correction was made when calculating mortality rates. The results show that there is a strong positive correlation between the recorded mortality rates and the exposure time and/or the concentration of the extracts. Moreover, the exploitation of the regression lines obtained by the probit of the concentrations allowed us to determine the LD50 in 24 and 48 hours (table 6). Thus, after 24 h, *Balanites aegyptiaca*, *Anacardium occidentale* and *Khaya senegalensis* having recorded 100% mortality require respectively 1.02 mg/L; 1.23 mg/L and 1.62 mg/L to induce 50% mortality in *Anopheles* larvae. After 48 h, these same plants require 0.81 mg/L, 0.28 mg/L and 1.41 mg/L respectively. This confirms that the effectiveness of the extracts varies from one plant to another and that *Balanites aegyptiaca* is more active on *Anopheles* larvae than *Anacardium occidentale* and *Caya senegalensis*. These plants are more effective than those used (Merabti et al. 2015) in Algeria. In substance, these authors showed that 50 mg/L of aqueous extract of *Citrullus colocynthis* is needed to obtain 50% mortality of *Culex pipiens* larvae. Our results appear to be more

effective than those obtained in Algeria (Benhissen et al. 2019) which showed that 22.17 g/L of *R. chalepensis* is necessary to obtain 50% mortality in *Culiseta longiareolata* larvae. On the other hand, our results are identical to those of the same authors who also showed that the longer the exposure time to the extracts, the more effective the plant is.

**Table 6:** Mortality rate of Anopheles larvae to aqueous plant extracts in 24h and 48h of exposure

Plants	24h	48h
<i>Anacardium occidentale</i>	12,7+8,6 a	17,03+6,3 a
<i>Annona senegalensis</i>	8,1+4,3 abc	16,03+ 6,3 a
<i>Balanites aegyptiaca</i>	13,3+8,6 a	14,7+7,9 ab
<i>Bauhinia rufescens</i>	1,5+1,7 d	10,6+5,1 cd
<i>Khaya senegalensis</i>	12+9 ab	13,7+8,2 abcd
<i>Carica papaya</i>	10,3+7,7 abc	12,2+7,6 abcd
<i>Grewiya bicolor</i>	5,5+3,5 bcd	11,5+5,5 bcd
<i>Manguifera indica</i>	3,9+4,0 cd	9,9+6,6 d
<i>Moniho esculentus</i>	11,2+8,3 ab	12,5+8,0 abcd
<i>Sclerocariya birea</i>	8,7+4,4 abc	15,5+6,2 abc
<i>Tamarindus indica</i>	12,7+6,8 ab	15,2+8,3 a

**Table 6:** LD50 and equation of lines in 24 and 48 hours

Plantes	24H		48H	
	DL <sub>50</sub> mg/l	Equation	DL <sub>50</sub> mg/l	Equation
<i>Annona senegalensis</i>	2,13	Y=1,13x+1,23	0,52	Y=3,61x-4,83
<i>Grewiya bicolor</i>	3,98	Y=1,91x-1,88	1,09	Y=1,85x-0,63
<i>Sclerocariya birea</i>	2,04	Y=1,22x+0,96	0,6	Y=3,58x-4,96
<i>Khaya senegale</i>	1,62	Y=8,73x-23,06	1,41	Y=8,24x-21,01
<i>Tamarindus indica</i>	1,2	Y=3,01x-4,29	1,17	Y=7,91x-19,33
<i>Bauhinia rufescens</i>	6,3	Y=3,39x-7,91	1,23	Y=1,37x-0,76
<i>Anacardium occidentale</i>	1,23	Y=5,74x-12,79	0,28	Y=2,94x-2,21
<i>Moniho esculentus</i>	1,9	Y=6,90x-17,69	1,2	Y=3,86x-6,92
<i>Manguifera indica</i>	3,63	Y=4,41x-10,71	0,85	Y=5,35x-10,68
<i>Carica papaya</i>	1,51	Y=4,07x-7,95	1,09	Y=4,48x-8,64
<i>Balanites aegyptiaca</i>	1,02	Y=6,02x-13,15	0,81	Y=5,35x-10,68

## CONCLUSION

Our research work is oriented on the valorization of plants of the biodiversity of Niger, used in the framework of the fight against malaria. The aim is to test the larvicidal activity of aqueous extracts of some plants and to determine their phytochemical compositions in order to develop products that could be an alternative to the use of synthetic chemicals. This study showed the larvicidal power of aqueous extracts of *Balanites aegyptiaca*, *Carica papaya*, *Tamarindus indica*, *Anacardium occidentale*, *Sclerocarya birea*, *Annona senegalensis* and *Khaya senegale* which allowed 100% mortality of *Anopheles* larvae after 48h of exposure. Indeed, the doses that allow 50% mortality of L2 *Anopheles* larvae with the aqueous extracts of these plants are relatively low. Moreover, the phytochemical screening showed that secondary metabolites such as polyphenols, flavonoids, polyterpenes sterols, tannins, saponosides, quinones and alkaloids can be effective in the control of *Anopheles* larvae vector of malaria in Niger. Finally, the aqueous extract of *Balanites aegyptiaca*, *Tamarindus indica*,

*Anacardium occidentale* and *Khaya senegale*, can be used in mosquito control campaigns by spraying in potential breeding sites. Indeed, the LD50 of the aqueous extracts of these plants are respectively 1.02; 1.2; 1.23 and 1.62 after 24h and 0.81; 1.17; 0.28 and 1.41 after 48h.

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