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IMPACT OF ETHYL EXTRACTS OF THE LEAVES OF HEXALOBUS MONOPETALUS (ANNONACEAE) ON DIURESIS AND SALIDIURESIS IN WISTAR STRAIN RATS

Rodrigue Akotegnon, Justin Behanzin, Ismael Hoteyi, Alphonse Sezan *
Laboratory of Biomembranes and Signalling Cell, University of Abomey-Calavi P.O. Box 1147, Cotonou, Benin

ABSTRACT

Our study on the treatment of pathologies related to waste disposal through water by urinary excretion focused on the ethyl extract of the dry leaves of Hexalobus monopetalus. The evaluation of the diuretic and salidiuretic properties of this extract is an analytical, prospective and transverse study. Its objective is to analyze the chemical composition of the leaves and to assess the diuretic and salidiuretic activities of the ethyl extract. The method used to analyze theleaves used is phytochemical screening. The aqueous extract is administered by gavage to wistar strain rats. The volume of urine excreted and the quantity of ions are determined on the measuring cylinder by ion spectrophotometry. The results show that the urine of the rats has a pH of between 9.3 and 9.15. The baseline diuresis in six (6) hours is 5mL. Significant diuretic activity (177%, 262.5% and 203%) was observed in rats treated with 67.5 doses, respectively; 135 and 202.5 mg / kg / bw of the Hexalobus monopetalus ethyl extract compared to 200% in furosemide-treated rats at 20 mg / kg. The saluretic activity of this extract is ethyl of 10.34 at a dose of 67.50 mg / kg of the extract, the same value for the furosemide at 20 mg / kg and 4.05 for the distilled water. The 135mg / kg dose of the extract is the best diuretic activity (262.5%) and the dose 67.5 mg / kg / bw of the extract is the best in terms of salidiuretic activity (10, 34 ± 1.5). The ethyl extract of the leaves of the Hexalobus monopetalus helps eliminate blood creatinine and blood does not influence the balance of ions such as calcium, potassium, sodium and chlorine e. However the extract prevents a good elimination of urinary urea through the kidney which would result from an organic deficiency

Key words: *Hexalobus monopetalus, diuretic and salidiuretic activities, ethyl extract, wistar rats.*

INTRODUCTION

In many of the major cities of Third World countries, self-medication is a recurring factor. In order to avoid expenditure related to consultation and the high cost of medicines, access to care is oriented towards self-medication in the absence of the severity of the disease. Self-medication accounts for 54% of therapeutic requests, while 23% of patients use private clinics and 16% of health centers (1). The population, in search of new therapies less expensive and easy to access, is devoted to herbal medicine.

Although herbal medicine remains in Benin, traditional, popular and ancestral, scientific research in recent years has considered this area as a priority. Many analyzes of the active principles of plants, proving their effectiveness and sometimes trying to understand their mode of action, are carried out. It is therefore vital for our country, Benin that researchers and scientists agree to take an even greater interest in studies and research on medicinal plants. This will contribute to sustainable endogenous development desired and achieved by some African populations (2). Similarly, high blood pressure is currently recognized as a global public health problem due to its frequency and cardiovascular complications. More than a quarter (26.4%) of the world's adult population is hypertensive and an estimated 7-8 million people die each year (3). In the Benin flora, several plant species are indicated in the treatment of malaria and high blood pressure. Numerous studies in traditional pharmacopoeia have revealed the diuretic and salidiuretic properties of several plant species. For example, Sanogo et al (2009) evaluated these properties in the Nitrokoudang recipe used in Mali in the treatment of high blood pressure. The same goes for Daouda (2010) who worked on herbal tea adomassin consisting of a mixture of medicinal plants used by the Beninese population in the treatment of several diseases including malaria. These properties are of great importance in the treatment of hypertension. Indeed, they lead to the urinary excretion of water and sodium contained in the blood. This contributes to a decrease in blood volume and thus a decrease in blood pressure (4).It is in this vision that we propose to evaluate the diuretic and salidiuretic effects of the ethyl extract of the leaves of Hexalobus monopetalus in this work.

MATERIALS AND METHODS

Materials

The aqueous hot extracts obtained from the leaves of Hexalobus monopetalus were used as plant material for the study.The animal material consists of blood (serum) and urine from male Wistar rats weighing between 180g and 220g, bred in the laboratory of the Biomembranes Laboratory and Cell Signaling. These animals have free access to food and water.Rat cages are used to collect urine and have a faecal separation glass system.

Photo : Diuresis cage

Methods of Study

Our work was carried out in two phases: a preparatory phase and an experimental phase of in vivo research on diuretic and salidiuretic properties.

Preparatory phase

The leaves of Hexalobus monopetalus were dried in the open air at room temperature. These dried leaves were finely ground. The powder obtained is then stored in glass jars in order to avoid external contamination. The powder was used to obtain the aqueous extract.To obtain the ethyl extract of the leaves of Hexalobus monopetalus, 100 g of the powder obtained from the previously dried Hexalobus monopetalus leaves weighed using an analytical balance of Sartorius® type were macerated in 1000 ml of Ethanol at 96 ° C. and homogenize on a magnetic stirrer for 72 hours at laboratory temperature. The macerate was filtered three times and the filtrate obtained was evaporated at 40 ° C. in a Rotavapor® evaporator. The recovered paste extract is placed in an oven at 45 ° C. for drying. After drying the extract is scraped and crushed in a porcelain mortar and weighed in order to calculate the yield. The yield is determined by the ratio of the weight of the dry extract after evaporation to the weight of the dry plant material used for the extraction multiplied by 100%. It is these extracts that will be used to prepare the concentration ranges tested. Phytochemical screening is a chemical analysis based on reactions of coloration or precipitation more or less specific to each class of active ingredients. It is carried out according to the method of Houghton and Raman (1998).

Phase of experimentation

Animals and accommodation

All animals are of EOPS health status (free from specific pathogenic organisms). Upon receipt, the rats are randomly placed in groups of two (02) in standard cages for an acclimatization period (2 weeks) before being used in the different experiments. During this period the animals have free access to food and water and are kept in a pet shop at a constant temperature (22 ± 2) ° C subjected to a 12 / 12h light / dark cycle.

Evaluation of the diuretic activity of Hexalobus monopetalus

In order to evaluate the diuretic activity with the ethyl extract of the leaves of Hexalobus monopetalus, we have disposed wistar rats weighing between 180 and 220 g, and of male sex. These rats were fasted for 18 hours before each test with free access to tap water only before and after receiving salt water surcharge (except the basic diuresis batch).

Our study was based on 05 lots of rats distributed as follows: lot 1 (base diuresis): 50ml / kg body weight distilled water, lot2 (Furosemide, MR): 20mg / kg body weight of furosemide. Lots 3; 4 and 5 received, in place

of furosemide, the ethyl extract of the leaves of Hexalobus monopetalus at the respective doses of 67.5; 135 and 202.5 mg / kg of bw

Determination of basic diuresis

For baseline diuresis, batch 1, 50 mL / kg / bw of distilled water was administered orally (esophagic gavage). Urinary excretion was measured 6H and 24H after administration.Détermination de l'activité diurétique de l'extrait éthylique des feuilles de Hexalobus monopetalus. The principle of determination of diuretic activity consists in measuring the volume of urinary excretion in wistar rats after administering 50 ml / kg of body weight of 1.8% NaCl solution and then the ethyl extract of the leaves of Hexalobus Monopetalus at the respective doses mentioned above: D67, 5; D100; D135 and D202, 5 mg / kg body weight in batches 3 to 5 respectively by oesophageal gavage. Lot 2 also received 1.8% NaCl solution as well as furosemide (MR), and lot 1 distilled water. After the treatment, each batch being composed of two rats, they are placed (each rat) in metabolic cages (01 rats per cage).Gasting of wistar rats: Administration of the ethyl extract of the leaves of Hexalobus monopetalus The gavage is done orally with a syringe equipped with an oesophageal probe. This ensures that the rat has swallowed the expected dose.

Measuring the volume of urinary excretion

For each group, urinary excretion of 6 h and 24 h was measured after treatment on each wistar rat. After placing them each in the metabolic cages, the following parameters were measured: latency (the appearance of the first urine drops after the gavage), the urinary volume after 6H and 24H and the urinary pH 24H after administration. The volume of urinary excretion (VEU) is obtained by the formula: VEU = VE / VA X 100; With VE = Volume of excretion; VA = Volume administered. VEU <80% = antidiuretic activity; VEU between 80-110 = no activity; VEU between 110-130% = low activity; VEU between 130-150% = moderate activity and VEU ≥ 150% = high activity. Note that a blood sample is taken 24 hours after the treatment on each rat. These blood samples will be used for the study of the salidiuretic activity of the ethyl extract of the leaves of Hexalobus monopetalus.

Evaluation of the salidiuretic activity of Hexalobus monopetalus

The principle is the influence of a diuretic on natriuremia and kaliuria in animals put in aqueous overload. The procedure is the same. The samples of urine collected were used. Urinary concentrations of sodium and potassium ions were determined using an Erbalyte Plus spectrophotometer and the salidiuretic activity was expressed as the [Na +] / [K +] ratio.

DETERMINATION OF URINARY POTASSIUM

Procedure

Measurement in test tubes	Blank	Standard	Assay
Reagent	1ml	1ml	1ml
Demineralized Water	10 µl		
Standard		10 µl	
Serum and urine			10 µl
Mix well. Incubate for 5 min at room temperature. Record the absorbances at 500 nm (450-500) against the reagent blank. The reaction is stable 30 min in the absence of light.			

Table 1: Determination of Potassium

Dosage of urinary sodium

Measurement in test tubes	Blank	Standard	Assay
Reagent	1ml	1ml	1ml
Demineralized Water	10 µl		
Standard		10 µl	
Serum and urine			10 µl
Mix well. Incubate for 5 min at room temperature. Record absorbances at 500 nm (450-500) Against the White reagent. The reaction is stable 30 min in the absence of light.			

Table 2: Determination of Sodium

Determination of other urinary and blood biochemical parameters

Blood sample

Blood sampling is carried out according to the experimental protocol employed by Weiss et al. (2000), and modified by Descat (2002). Puncture of the retro-orbital sinus was performed. The animal is held with one hand in lateral decubitus, and held by the skin of the neck. The pressure of the thumb on the neck, behind the angle of the jaw, allows compression of the jugular vein, and therefore venous stasis towards the head, favoring the filling of the retro-orbital sinus. By making a slight traction on the upper eyelid with the index finger, we create an exophthalmos facilitating the taking of blood by means of tube with hematocrit not heparinized. The end of the tube is slowly introduced into the lateral angle of the eye. Progression through the tissues is facilitated by printing a small pipette rotation. As soon as the venous plexus is reached, the blood springs into the perorbital space and ascends by capillary action in the tube. The volume of blood collected is 0.5 to 2 ml. Before the tube is removed, the compression is released and the bleeding ceases spontaneously when the ocular pressure normalizes. The recovered blood is used for the determination of the various biochemical parameters.

Determination of calcium

Principle

The CPC method described (derived from Moorehead and Briggs) is used to determine the total calcium concentration in serum, plasma or urine. In an alkaline medium, 0-cresolphthalein complexon or CPC reacts with calcium ions to form a dark red colored complex whose absorbance, measured at 570 nm, is proportional to the calcium concentration in the specimen.

Procedure

Measurement in test tubes	Blank	Standard	Assay
Reagent	1 ml	1ml	1ml
Demineralized Water	25 µl		
Standard		25 µl	
Serum and urine			25 µl
Mix well. Incubate 5min at room temperature. Read the absorbance at 570 nm (550-590) against the reagent blank. The color is stable for one hour in the absence of light			

Table 3: Determination of Calcium

Determination of magnesium

Principle

Method based on that described by Gindler, Heth, and Khayam-Bashi. The camalgite, a metallochromic indicator, forms a colored complex, in an alkaline medium, with magnesium. The absorbance of the complex is measured at 510-550 nm and is proportional to the magnesium concentration in the specimen. EGTA limits the interference of calcium, while potassium cyanide reduces that of heavy metals. Polyvinylpyrrolidone and a surfactant reduce the interference of proteins and lipid levels.

Measurement in test tubes	Blank	Standard	Assay
Reagent R1	1ml	1ml	1ml
Demineralized Water	10 µl		
Standard R2(20mg/l)		10 µl	
Serum			10 µl
Mix. Let stand 5 minutes at constant temperature. Read the absorbance of the standard and tests at 530 nm (510-550 nm) against the reagent blank			

Table 4: Determination of Magnesium

Determination of chlorine

Principle

$Hg(SCN)_2 + 2Cl^- \rightarrow HgCl_2 + 2SCN^-$

$3(SCN)^- + Fe^{3+} \rightarrow Fe(SCN)_3$

Procedure

Measurement in test tubes	Blank	Standard	Assay
Reagent	1ml	1ml	1ml

Demineralized Water	10 µl		
Standard		10 µl	
Serum and urine			10 µl
Mix well. Incubate for 5 min at room temperature. Record the absorbances at 500 nm (450-500) against the reagent blank. The reaction is stable 30 min in the absence of light.			

Table 5: Determination of Chlorine

Determination of urea

Principle

The method used is the enzymatic method based on the reaction described by Talke and Schubert (1895). The reaction scheme is as follows:

NH3+oxoglutarate+ NADH + H+ → Glutamate + NAD+ +H20

Urée +H20 → 2NH3 +CO

The decrease in absorbance due to the conversion of NADH to NAD +, measured for a given time at 340 nm, is proportional to the urea concentration in the specimen.

Procedure

Measure in a thermostatically controlled tank (30 ° C or 37 ° C)	Standard	Assay
Reagent	1ml	1ml
Standard	5µl	
urine		5µl
Mix. Read the concentrations at 340nm. 1st A1 reading in 30 seconds. 2nd reading A2 at 90 seconds.		

Table 6: Determination of urea

Dosage of Creatinine

Principle

The method used is the reaction (Jaffé reaction, without pre-treatment step of the specimen) of creatinine with picric acid in an alkaline medium whose developmental kinetics are measured at 490 nm (490-510). This method has been optimized (specificity, speed and adaptability) by the development of a two-point kinetic method.

Procedure

Measure in a 1cm tank of optical path.	Blank (facultatif)	Standard	Assay
Reagent (R1+R2)	1ml	1ml	1ml
Demineralized Water	100µl		
Standard		100µl	
urine			100µl
Mix well after 30 seconds, record absorbance A1 at 490nm (480-510) against reagent blank or distilled water. Read the absorbance.			

Table 7: Determination of creatinine

Statistical analysis

Statistical processing was carried out on independent serial samples using STATISTICA software version 5.5. The results are considered statistically significant when $P < 0.05$.

RESULTS

Extraction yield

The yield is determined by the ratio of the weight of the dry extract after evaporation to the weight of the dry vegetable material used for the extraction multiplied by 100 (Medane, 2012).

Let R be the yield of the extract,

Mass of the extract

$$R = \frac{67.56}{200} \times 100$$

X 100

Mass of the leaves powder

$$R = 33.78\%$$

This study shows that after ethyl extraction of the powder of the leaves of Hexalobus monopetalus, we obtained a yield of 33.78%. The extract has a dark red color.

Phytochemical Screening

Table 8 shows the result of the phytochemical screening of the EEHm

Groupes Chimiques	**Sous-groupes**	**Observations**
The polyphenolic compounds	tannins	+++
	Gallic tannins	++
	Catechic or condensed tannins	++
	anthocyanin	++
	Leuco anthocyanin	+++
flavonoids	flavone	+++
Mucilage		+++
Reducing compounds		+++
saponosides		-
alkaloids		+++
Cyanogenic derivatives		-
Anthracene Derivatives	Free Anthracenics	+++
	O. Heterosides	+
	(A. combined)	-
triterpenoids	C. Heterosides	-
steroids	(A. combined)	+
coumarins	tannins	-
Quinonic derivatives	Gallic tannins	+++

- Absent + little abundant ++ Abundant +++ Very abundant

Table 8: Phytochemical Screening Results

The results of the phytochemical screening showed the presence of several chemical compounds with various properties and in various proportions (see Table 8). These are polyphenolic compounds such as: gallic tannins, catechic or condensed tannins, anthocyanins, and leuco-anthocyanins; Flavonoids; Mucilages; Reducing compounds; Alkaloids; Of certain anthracene derivatives such as: free anthracenics, O-heterosides; Steroids and quinone derivatives. As for alkaloids, they have also been detected in the essential oils of fruits of Hexalobus monopetalus by Hamisi et al. (2014). This supposes that this compound is not found only at the leaf level.

Diuretic activity

Variation of the urinary volume of 6h and 24h

FIG. 9 shows the urinary volumes of 6 h and 24h

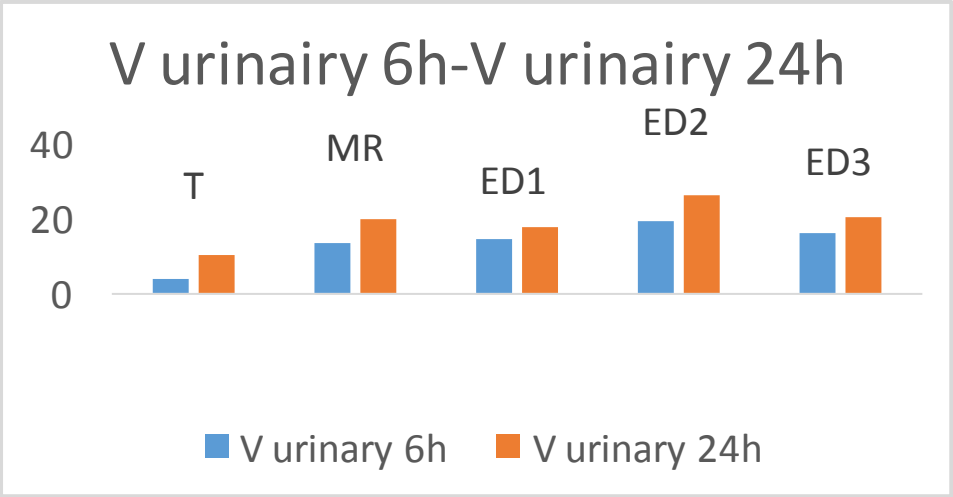

Figure 9: Urinary volumes of 6 hours and 24 hours

The analysis of this figure shows the variation of the urinary volumes of 6 hours and 24 hours.
- Furosemide (MR) at a dose of 20mg / kg bw resulted in an increase in urinary volumes compared to the control from the first hour to the 6th hour. The overload elimination time is within the first three hours after administration.
- EEHm caused a significant increase in urine volumes compared to the control at doses 67.5 and 135 mg / kg bw, as compared to furosemide at 135 mg / kg bw from the second hour after administration and continued until 'At the 24th hour. The action is important at the dose of 135mg / kg bw (27.5 ± 2.5) mL in 24h.

The results of the diuretic activity of the EEHm

The estimated diuretic activity of EEHm and furosemide, based on the value of volumetric urinary excretion, is shown in Table 9.

treatments	Dose/Kg/bW	VA NaCl (ml of bw)	VE/6h (mL)	EUV (%)	Activity
	50 mL		10 2,61	100	Diuretic
Distilled water (control)	20 mg	10	20 2,07	200	No activity
furosemide	67,5 mg	10	17,7 3,56	177	Significant activity
Dose 1	135 mg	10	26,25 3,72	262,5	Significant activity
Dose 2	202,5 mg	10	20,3 2,32	203	Significant activity

Table 9: Results of the diuretic activity of the ethyl extract of the leaves of Hexalobus monopetalus and furosemide in Wistar rats (n = 5)

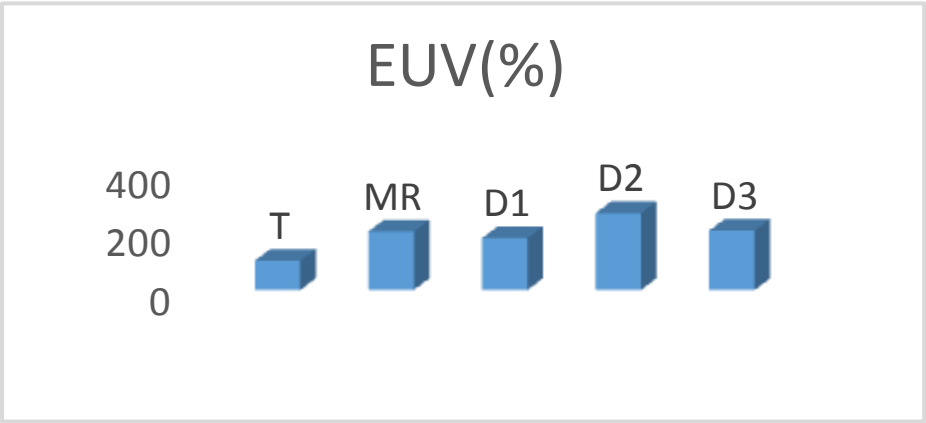

Figure 10: Histogram of volumetric urinary excretion according to the treatments.

Figure 10 shows the results of the evolution of the EUVs. Indeed, it shows an important diuretic activity (VEU ≥ 150%) in the ethyl extract of the leaves of Hexalobus monopetalus at all doses. However, the highest diuretic activity is obtained with the dose of 135 mg / kg bw (262.5). This activity is greater than that of furosemide (200). The first urination was faster: 15 min after the administration of 135 mg / kg of the ethyl extract of the leaves of Hexalobus monopetalus.
From the analysis in Figure 11, we can say that the MHEA has a significant diuretic activity at all doses after 24h. This activity is very important at a dose of 135mg / kg / bw which is even higher than that of the reference molecule (furosemide).

Variation of urinary PH

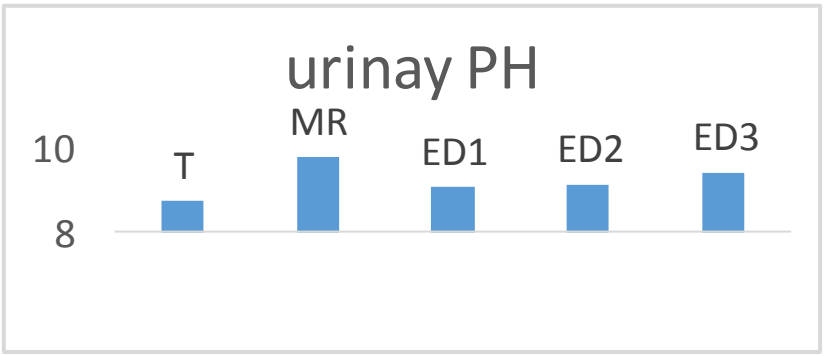

Figure 11: Histogram of urinary pH variation

The graph in Figure 11 shows the pH of the urine samples taken 24 hours later.
The histogram of urinary pH variation shows that EEHm at doses 67.5; 135 and 202.5mg / kg bw resulted in a slight increase in urine pH compared with the control at 9.10; 9.15 and 9.30 with respect to the control (8.75). However, these values are low compared with that obtained with furosemid (9.75). Therefore, EEHm does not influence urinary pH. The urinary medium remains always basic (pH above 7).

Salidiuretic activity
Urinary concentrations of sodium, potassium and chloride ions and the ratio of Na + / K + concentrations are given in Table 10.

Traitements	Concentration des ions (mmol/L)			Rapport
	Na+	K+	Cl-	[Na+]/ [K+]
Eau distillée (témoin)	52,7 ± 1,5	13 ± 1	59,95 ± 1,5	4,05 ± 0,5
67,5 mg/kg (D1)	239,9 ± 0,50	23,21 ± 1.5	154 ± 1	10,34 ± 1,5
135 mg/kg (D2)	121,25 ± 1	15,77 ± 1,5	130,95± 0,50	7,69 ± 0,5
202,5 mg/kg (D3)	184,7 ± 1,5	22,88 ± 0,50	207,9 ± 1	8,07 ± 0,50
Furosemide (50 mg/kg)	152,3 ± 1	14,73 ± 1	169,**45** ± 1	10,34 ± 1

D1 = dose 1; D2 = dose 2; D 3 = dose 3.

Table 10: Urinary ionogram results of wistar rats after treatment.

The observation of Table 10 reveals that the ethyl extract of the leaves of Hexalobus monopetalus allowed the rats to excrete more sodium than potassium. Table 10 shows the mean of the different types of ions quantified in each batch with the ratio [Na +] / [K +]. The analysis of this same table shows that the [Na +] / [K +] ratio of the 67.5 mg / kg bw of the ethyl extract of the leaves of Hexalobus monopetalus was 10.34; The same result for furosemide at 20 mg / kg bw and 4.05 for rats treated with water (control). This ratio is slightly lower at doses of 135 and 202.5 mg / kg bw, which are 7.69 and 8.07, respectively.

Results of the determination of other blood and urinary parameters after administration of the ethyl extract of the leaves of Hexalobus monopetalus.

Changes in serum calcium under the effect of EEHm

Figure 12: Calculation

The analysis of the figure shows that the blood calcium level varies between 95 and 99 mmol / L with the EEHm at the different doses and that of the furosemide is 101 mmol / L. Compared to the control (87.5 mmol / L), there was a slight increase in the blood calcium level due to EEHm, but this increase was lower than that induced by furosemide.

Variation of magnesemia due to EEHm

Figure 13: Magnesemia

The analysis of FIG. 14 shows that the EEHm at doses 135 and 202.5 mg / kg bw does not lead to any change in the level of blood magnesium (22 mmol) compared with the control (22.5 mmol) as well as Furosemide. However, EEHm at a dose of 67.5 mg / kg bw results in a slight decrease in the level of magnesium in the blood.Variation de la Kaliémie-Kaliurie sous l'effet de l'EEHm

Figure 14: Kaliemia-Kaliuria

The serum potassium does not vary at all at the different doses of the extract.On the other hand, kaliuria increases significantly at the different doses of the extract compared to the control (13 ± 1) and furosemide (14.73 ± 1). It peaks at a dose of 67.5 mg / kg bw (23.21 ± 1.5).

• Variation of Natremie-Natriurie under the effect of the EEHm

Figure 15: Natremia-Natriuresis

Natremia does not vary at all by the different doses of the extract. Values are proportional to control and furosemide values. Natriuremia, on the other hand, increases very strongly at the different doses of the extract and has a peak at the dose 67.5 mg / kg bw (240 ± 0.55).

• **Variation of Chloraemia-chloruria under the effect of EEHm**

Figure 16: Chloraemia-Chloruria

Chloraemia does not vary at all at the different doses of the extract.Chloride, on the other hand, increases significantly compared to the control (59.95 ± 1.5) and reaches its peak (207.9 ± 1) at a dose of 202.5 mg / kg bw

• Variation of urinary urea at the different doses of the EEHm

Figure 17: Urinary Urea in g / L

The elimination of urea at different doses of EEHm was small compared to control (13.7 ± 1) g / L and compared with furosemide (10.5 ± 1.5) g / L, Is increased at a dose of 202.5 mg / kg bw.

• Variation of creatinine at the different doses of the EEHm

Figure 18: Creatinine

Figure 19 shows a slight increase in urinary creatinine at doses 67, 5 and 202, 5 mg / kg bw of EEHm (206 and 201 mmol) versus control (160 mmol) and very high Slightly compared to furosemide. At the dose of 135 mg / kg bw of HESE, creatinineuria was almost unchanged

DISCUSSION

Our study is based on the impact of ethyl extracts of the leaves of Hexalobus monopetalus on diuresis and salidiuresis in rats. The yield of the extraction is 33.78% with an extract with a dark red appearance; Shiny and insoluble in water. This result is consistent with that of Elisabeth HOUEZE et al (2015) for an ethyl extraction as well. This could be explained by the fact that the same techniques were used during the extraction and that the leaves of the plant were harvested in the same geographical area during the same period. The leaves of Hexalobus monopetalus therefore contain active ingredients which are extractable by ethanol.

The results of the screening show that the ethyl extract of Hexalobus monopetalus is rich in polyphenolic compounds such as: gallic tannins, catechic or condensed tannins, anthocyanins, and leuco anthocyanins; Flavonoids; Mucilages; Reducing compounds; Alkaloids; Of certain anthracene derivatives such as: free anthracenics, O-heterosides; Steroids and quinone derivatives. Alkaloids were also detected in the

essential oils of fruits of Hexalobus monopetalus by Hamisi et al. (2014). This supposes that this compound is not found only at the leaf level. According to Bruneton (2010), flavonoids are powerful diuretics and especially those of the flavonone type. Réméssy et al. (1996), by analyzing the mechanism by which flavonoids act, find that the protective and dilating effects of blood vessels and their ability to make blood flowing would be due to flavonoids. They argue that the elimination of toxins is an essential condition for promoting weight loss (Réméssy et al., 1996). However, the phytochemical analysis of the leaves of Hexalobus monopetalus reveals an abundant presence of flavonoids. This would explain the strong diuretic activity of the EEHm. Treatment with furosemide also gave a significant diuretic activity with a EUV of 200%. This result is similar to that of Toutain (2007), which showed that furosemide exerts the diuretic effect on Henle's loop, which makes him a diuretic. Doses 67.5; 135 and 202.5 mg / kg bw all lead to significant diuretic activity with respectively 197%, 262.5% and 203% volumetric urinary excretion in twenty-four (24) hours. Indeed, the urinary volume of furosemide (20 ± 2.07 mL) is low in EEHm at doses of 135 mg / kg bw (26.25 ± 3.72 mL); And; 202.5 (20.3 ± 2.32mL) and strong at that dose of 67.5mg / kg bw (17.7±3.56 mL) in 24h.The elimination of sodium was significant whereas that of potassium was less in animals treated with EEHm. The salidiuretic activity of this ethyl extract was 10.34 at the dose of 67.50 mg / kg / bw, 7.69 at the dose of 135 mg / kg / bw and 8.05 at the dose 202.5 mg / Kg / bw compared to 10.34 for furosemide 20 mg / kg and 4.05 for rats in the control group treated with distilled water. HEME therefore has a salidiuretic effect. These diuretic and salidiuretic effects of Hexalobus monopetalus may be beneficial in the management of certain cases of hypertension insofar as they act by urinary elimination of part of the water and sodium contained in the blood. This will result, according to Sanogo et al (2009), a decrease in blood volume and thus a decrease in blood pressure.In addition, potassium ion is the major cation of the intracellular medium and its gradient on both sides of the cell membrane is mainly determinant of the transmembrane electrical potential that influences the excitability of tissues such as nerves and muscles (Hannedouche, 2007). However, the results show that at 67.50 mg / kg, there is a high excretion of sodium and potassium retention. Then the aqueous extract of the leaves of Hexalobus monopetalus can be used to, on the one hand maintain the electrochemical composition of the intracellular medium and on the other hand to treat arterial hypertension at this dose. Urinary pH remained basic after administration of EEHm doses. This result is not in line with the results of ADEROMOU A. et al (2010) who evaluated the diuretic and salidiuretic properties of aqueous extracts of Elaeis guineensis and obtained urinary pH values between 6.1 and 8.6. Indeed, the latter observes a difference between the values of the pH which would be a consequence of the physiological difference of the organisms of the animals tested; or diet. However, with our extract, the different pH values obtained show that the EEHm does not influence the acid-base balance of the organism at different doses 67.5; 135 and 202, despite the physiological difference in the organisms of the test animals; or diet. The 135mg / kg dose of the extract is the best diuretic activity (262.5%) and the dose 67.5 mg / kg / bw of the extract is the best in terms of salidiuretic activity (10 , 34 ± 1.5).In addition the EEHm by stimulating renal urinary excretion facilitates the elimination of creatinine, a toxic waste of the body. However, EEHm would not facilitate removal of urea. The elimination of urea at different doses of EEHm was small compared to control (13.7 ± 1) g / L and compared with furosemide (10.5 ± 1.5) g / L, Is accentuated at a dose of 135 mg / kg bw. This would be due either to a negative influence of the EEHm on the metabolism of the amino acids, the urea being a waste resulting from this metabolism, or to an attack on the functioning of the kidneys. The ratio of sodium to potassium is normally greater than 1. A ratio of less than 1 indicates a functional renal insufficiency generally linked to hypovolemia by soda leak from Burtis C. A. et al. A ratio greater than 1 indicates hypernatriuresis and is found in the organic renal insufficiency according to Frey J. et al.The idea that MHEA alone would have a negative influence on amino acid metabolism is implausible. The low elimination of urea is therefore due to an organic kidney failure as the sodium / potassium ratio is greater than 1 with all the doses used.In addition blood levels of calcium; Magnesium; Sodium; Potassium and chlorine did not vary significantly under the effect of EEHm. The functioning of the ion channels is therefore not influenced by the EEHm.In other words, the EEHm is a diuretic and a salidiuretic which does not influence the functioning of the ion channels. The effective dose for this diuretic and salidiuretic activity is 135 mg / kg body weight. However, the removal of the urea is disturbed by the effect of the extract.

CONCLUSION

The leaves of Hexalobus monopetalus consist of four (04) subclasses of polyphenolic compounds (gallic tannins, catechic or condensed tannins, anthocyanins, and leuco-anthocyanins), flavonoids, mucilages, reducing compounds, Alkaloids, two (02) types of anthracene derivatives (free anthracenics, O-heterosides), steroids, and quinone derivatives.The ethyl extract of the dried leaves of Hexalobus monopetalus has a high diuretic activity from 67.5 mg / kg / bw. This extract therefore contains active ingredients which give it diuretic action.The ethyl

extract of the dried leaves of Hexalobus monopetalus also favors the elimination of certain ions such as Na +, cl- and K + retention by urinary excretion. It then has a salidiuretic activity.The effective dose of the ethyl extract of the dried leaves of Hexalobus monopetalus with respect to the diuretic and salidiuretic results is 135 mg / kg.With their diuretic and salidiuretic activities, the ethyl extract of dried leaves of Hexalobus monopetalus has no significant influence on blood levels of calcium, potassium, sodium, and chlorine in wistar rats.It also promotes the elimination of blood creatinine through the urine. However, the extract does not promote a good elimination of urea, consequence of an organic kidney failure that the latter would entail.

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