Androgenic Effect and Phytochimie of the Bark of Trunk of Buchholzia coriacea Engl. (Capparidaceae)

Ondele R1*, Peneme. B.M.L1, Miguel L1, Akassa H1, Etou Oscibi A. W.2, Elion Itou R.D.G1. Nkounkou Loumpangou C.2, Abena A.A.1

1 - Laboratory of Biochemistry and Pharmacology of the Faculty of the. Sciences of Health. University Marien Ngouabi Republic of Congo
2 - Chemistry Unit and of the Plant Faculty of them. Sciences and Technique. University Marien Ngouabi Republic of Congo

ABSTRACT

The testosterone is the main androgen produced by the testes among the males. Her absence is responsible for erectile dysfunction and the loss of the sexual performance. The aim of the present study is to evaluate the androgenic effect and to determine the profile phytochemistry of the barks of trunk of B. coriacea. Fifteen days after the castration, the animals left in group and, received the aqueous extract of B. coriacea (100, 250 and 500 mg/kg, p.o). The sexual parameters (mounts sexual, number of erections, number of ejaculations and the time of latency) have been valued, and compared, to the witnesses groups: water distilled (0,5 ml/100 mg, p.o) and Enanthate of testosterone (1 mg/mL, i.m). The dosage of the testosterone has been achieved by the technic of ELISA analysis. The determination of the chemical families has been achieved by the technique of coloration in tubes. The aqueous extract of B. coriacea (100,250 and 500 mg/kg, p.o) has an effect androgenic because managed daily during 15 days, it provokes an increase of the rate plasmatic of the testosterone and the bodily weight of the rats, maintains the number of ejaculations, increase the latency and the sexual performance. The aim of the present study is to evaluate androgenic effect and to determine the profile phytochemistry of the barks of trunk of B. coriacea. The barks of trunk of B. coriacea would contain the stérols, the flavonoids, the saponosids that could be responsible for the sexual effects and androgénic observed. The aqueous extract of the plant would possess the properties androgenic would be to advise the patients presenting problems of masculine barrenness.

Keywords: Androgen - Phytochimie - Buchholzia - coriacea

INTRODUCTION

The Fertility is a quality important of the man’s life. But many factors as the deficit in sexual hormone caused to age contributed to its reduction. The physiology of the human reproduction shows that age influences the reduction of the blood free testosterone caused to the absence of link between the testosterone and the SBHG. What explains the reduction of the performance and the sexual impotence thus at the aged person. Culpabilised in this physiological state, the man uses today the medicinal plants to make again sexually. B. coriacea is called in languages vernacular by ombanda; songo lama, respectively in gangoulo and lari. The plant was the subject of numerous pharmacological studies that is the case of the aphrodisiac potential of the aqueous extract of the bark of the trunk1; effect of the aqueous extract of the barks of the trunk on the arterial pressure2; effects on the sexual and contraceptive cycle of the leaves 3,4; Anti-inflammatory and healing effect of the aqueous extract and ethanolic of the barks of the trunk5 and The assessment of the pain-killing activities and anti oxidizing6. The present work aims the assessment of the effect androgénic and phytochemistry of the barks of trunk of B. coriacea among the rat.

MATERIAL AND METHOD

Plant material: The barks of trunk of Buchholzia coriacea harvested in Brazzaville in April 2016, have been used. A specimen has been deposited to the National herbarium of the Center of study on the Plant Resources (Congo) under the n° 2456 (IEC) of the 17-2-1968. The harvested barks have been dried to the ambient temperature (28 ± 1 °C), safe from the solar rays during 20 days and pulverized with the help of a mortar made of wood.

Animal material: The male adult rats aged of 20 to 24 weeks and weights understood between 200 and 250 g have been
used. They have been provided by the animalerie of the Faculty of the Sciences and Techniques where they are maintained in conditions standard of éclairement (12 hours of lighting 12 hours of obscurity) to the ambient temperature of 25 ± 1 °C. These rats had the free access to a standard food and to the water of faucet.

**Chemicals**

The reagents of the different chemical families have been used, to have itself/themselves:

- HCl (20%); HCl (1 N) HCL extract, alcohol éthanolique.; Reactive of Meyer, alcohol isoamylique, Cuttings of magnesium, Chloroform, acetic anhydride, acid sulphuric extract (H2SO4), the reagent of Stiany and the chloride of fer (FeCl3 1%)

**Preparation of the aqueous extract**

The decocte to be prepared from 50 g of powder of barks of trunk of *B. coriacea* in 500 ml of water distilled during 15 minutes. After cooling then filtration on the absorbent cotton and the Wattman paper, the gotten décocé to been concentrated to the bath gets married thermostat to 55 °C. what permitted to get 15 g of a strong residual of brown color, either an output of 30%.

**Study of the effect of the aqueous extract on the sexual parameters among the castrated rat**

This study has been achieved in the goal to determine the effect substutif of the testes by the aqueous extract of the plant. It has been made according to the method of Roubinian7, modified slightly.

**Preparation of the animals**

The rat is anesthetized to the ethylic ether then placed in dorsal decubitus on a tray of dissection. With the help of scissors the animal deplated around the testicular scrotum. One makes an incision of the different muscular layers, on the median line of the scrotum separating the two testes, then. By a light pressure, appears the whitish color testicle. Two sons of attachment are placed then on both sides of the épидidyme and the vaginal. With the help of scissors, the testes are dereed, then one conducts the suture of the wound thus formed.

**Treatment of the Animals**

15 days after the castration, thirty (30) rats, distribute in six (6) shares of five (5) rats have been called the following manner daily:

- the group 1 (neutral witness) received the distilled water (0,5 ml/100g, per bone) during 7 days;
- the group 2 received the élanthane of testosterone (1 ml/100g, i.m) during the 3 days that precede the tests of copulation;
- the groups 3, 4 and 5 received the aqueous extract to the respective doses of 100, 250 and 500 mg/Kg per bone during 7 days of treatment. The observation of the different sexual parameters (numbers of mount , of erections, of ejaculations and the time of latency) has been made during one hour.

**Study of the effects androgenics of the aqueous extract of the Barkes of trunk of *B. coriacea* among the rat**

Twenty (20) rats left in four (4) shares of five (5) animals each has been called the following manner daily:

- the group 1 (witnes) received by oral way of water distilled to the dose of g 0,5ml/100 of bodily weight;
- the group 2, 3 and 4 received by oral way the aqueous extract of *B. coriacea* to the respective doses of 100, 250 and 500 mg/Kg.

The treatment of the animals is achieved during fifteen (15) days as well as the measure of the weight of every animal. Three (3) hours after the last administration, the animals have been lullè by imitation to the ethylic ether. With the help of the tubes to hematocrite, the blood of every animal has been appropriated by orbital way then recovered in tubes héparinés for the dosage of the testosterone.

The dosage of the testosterone has been achieved by the technique of Immunoanalyse while using the Diamed kit * as follows: 25µl of samples and control have been distributed in wells. In these, one adds 200µl of conjugué(Anticorps marked to the peroxydase).

The whole preparation is hatched during 60minutes between 20 and 25°C to ambient temperature. With the help of a washer of plates of RAYTO mark, the plates are washed 3fois with 400µl tampon of washing. Thereafter 200µl of substratum are distributed in the wells. After incubation during 15 minutes to 20 - 25°C (in obscurity), one adds 100µl of the stop solution (sulphuric acid). The absorbances is read to 40nm while using a reader of RAYTO mark. The concentration plasmatique of testosterone has been calculated according to the formula: The dosage of the testosterone has been achieved by the technique of Immunoanalyse while using the Diamed kit * as follows: 25µl of samples and control have been distributed in wells. In these, one adds 200µl of conjugué(Anticorps marked to the peroxydase).

The dosage of testosterone has been calculated according to the formula: 

\[
Cs = Ce \times \frac{DOS}{DOe}
\]

\(Cs = \text{concentration plasmatique of the substance in ng / ml}
\)

This = concentration of the sample in mg/ml

DOS = optic density of the substance

Doe = optic density of the sample

**Determination of the big chemical families**

This survey has for goal to search for the constitutes chemical of the peels of trunk of *B. coriacea* by classic methods of chemical screening® . They consist in identifying the chemical families (that could be responsible for the observed effects) by characteristic tests. These chemical families are the anthraquinones, the free quinones, the tannins, the stérols and terpénoïdes, the anthoyanthes, the Saponosideses, the alkaloids, the flavonoïdes and the hétéròsidès cardiotonics.

**Statistical analysis of the results**

The results expressed affected on average of the standard mistake are submitted to an analysis of variance to a factor followed of a t test of Student - Fischer. The observed difference is meaningful when the calculated t value is in absolute value, superior to the read t value in the t table of Student for d.d.l = n1+n2 - 2 and the risk of first species of 5%, n1 and n2 are the number of values for every measure.
RESULTS

Effect of the aqueous extract of Buchholzia coriacea on the sexual parameters among the castrated male rat

The table 1 shows that the aqueous extract of the barks of *B. coriacea* (100, 250 and 500 mg/kg) increases the number slightly of mount sexual in relation to the rats witnesses (NaCl 0,9%). The respective percentages of increase being of 38,29; 21,27 and 38,29%. With the énanthate of testosterone 1 mg/ml used like molecule of reference, the number of go up sexual passed from 9,4 ± 1,69 in 28,2 ± 1,06 mount sexual (p <0,001), either an increase of 66,67%. Concerning the number of erections, the aqueous extract of the barks of *B. coriacea* provokes an increase of 100% of this one to the studied doses.

The rats having received the physiological solution (NaCl 0,9%) didn't present any erection. The figure 1 shows otherwise that the énanthate of testosterone 1 mg/ml and the extract of this plant to the studied doses are without effect on the number of ejaculations as the physiological solution (NaCl 0,9%). Concerning the time of latency, the aqueous extract of the barks of *B. coriacea* to the doses of 100, 250 and 500 mg/kg, induced of the respective non meaningful increases of the time of latency of 15,05; 1,05 and 13,89%. The énanthate of testosterone 1 mg/ml leads a reduction of the time of latency of 51,81%.

Effects of the extract aqueous barks of trunk of *Buchholzia coriacea* on the ponderal evolution among the rat

The figure 1 watch the ponderal evolution of the rats submitted to the treatment with the aqueous extract of the barks of trunk of *B. coriacea* during 15 days. The aqueous extract of the barks of trunk of *B. coriacea* (100,250 and 500 mg/kg) managed to the rats watch an increase of the weight in relation to the first day of the treatment. The weight of the animals pass of 99,58 ± 1,26 to 109,95 ± 0,9g (p <0,05); 104,35 ± 1,84 in 114,83 ± 2,36 g (* * p <0,01) and 103,29 ± 1,5 to 109,53± 2,36 g (p <0,05) are increases of 9,43; 9,12 and 5,43% respectively to 100, 250 et 500 mg/kg.

Effects of the aqueous extract of the bark of trunk of *Buchholzia coriacea* on the rate plasmatic of testosterone among the rat

The present figure 2 the variation of the testosterone concentration plasmatic among the rats treated to the increasing doses of *B. coriacea* (100, 250 and 500 mg/kg). To these doses, this extract increases the concentration plasmatic of testosterone. This increase is not meaningful in relation to the rate plasmatic among the animals of the share witness. The rate plasmatic passes to 1,072 ± 0,13; 1,42 ± 0,56 and 0,75 ±0,14 ng/ml is increases of 49,25; 61,97 and 28% respectively to 100, 250 and 500 mg/kg, against 0,54 ± 0,24 ng/ml for the witness group. The rate plasmatic of the animals having received the aqueous extract of the barks of *B. coriacea* to 250 mg/kg is two times superior to the one of the animals treated to 500mg/kg.

Determination of the big chemical families of the bark of trunk of *Buchholzia coriacea*

The II picture presents the results of the characterization in tubes achieved on the extracts of barks of *B. coriacea*. The chemical screening achieved on this species permitted to put in evidence six big chemical families: the flavonoïds, the alkaloids and anthraquinons in these extracts.

Table 1: Effect of the aqueous extract of the bark of *B. coriacea* on the sexual parameters among the castrated rat

<table>
<thead>
<tr>
<th>Traitements</th>
<th>Parameters sexuals</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Number of mountes</td>
</tr>
<tr>
<td>NaCl (0,9%)</td>
<td>9,4 ± 1,69</td>
</tr>
<tr>
<td>ET 1mg/ml</td>
<td>28,2±1,06***</td>
</tr>
<tr>
<td>B. c 100 mg/kg</td>
<td>13 ± 1,78</td>
</tr>
<tr>
<td>B.c 250 mg/kg</td>
<td>11,4 ± 3,02</td>
</tr>
<tr>
<td>B.c 500 mg/kg</td>
<td>13 ± 0,89</td>
</tr>
</tbody>
</table>

* ET : Ethanate de testostérone ; B.c : *Buchholzia coriacea*. The values are middle ± ESMS, with n = 5, * * p <0,01, * * * p <0,001, meaningful difference in relation to witness NaCl 0,9%.
DISCUSSION

The present study had for objective, to value the effect androgenic and the survey of the phytochemistry of barks of trunk of B. coriacea.

The results of the treatment of the rats castrated with the aqueous extract of the barks of trunk of B. coriacea show the maintenance of the sexual activity observed among the non castrated rats (number of mount sexual and erections). The maintenance of this sexual activity could be assigned to the aqueous extract of the barks of trunk of B. coriacea. Indeed, it is possible that this extract contains some substances analogous to the androgens (phytohormones), responsible of the sexual activity, whose testes are the seat of the production. Other authors also showed the persistence of the sexual activity among the male rats castrated treated with the extracts of medicinal plants. It is the case of the aqueous extracts and ethanolic of the peels of trunk of Bridelia ferruginea and of the extract of Dracaena arborea. Otherwise, it is noted in the present study that the androgen of reference (enanthate of testosterone 250 mg to 1 mg/ml, i.m) seems to act in a more efficient way in relation to the aqueous extract of the barks of trunk of B. coriacea (100, 250 and 500 mg/kg, p.o) on the numbers of mount sexual and of erection and on the time of latency among the castrated rats. The administration by oral way (p.o) this extract could explain its weak efficiency. Indeed, the administration by oral way of the products can come with effects of first passages intestinal and hepatic at the origin of a reduction of the biodisponibility of the active principle and therefore his action. it is established that the action pharmacodynamic of a product is bound closely to the number of receptors activated and therefore to her concentration to the level of the organs targets.
The effect of the aqueous extract of the barks of trunk of B. coriacea (100, 250 and 500 mg/kg, p.o) on the rate plasmatic of the testosterone among the rats was sought-after. The results show that this extract provokes a not meaningful increase of the rate of the testosterone in relation to the witness having received the distilled water (0.5 g ml/100, p.o). This increase suggests that the aqueous extract of the barks of trunk of B. coriacea could have an effect androgénique (stimulative effect of the production of the testosterone).

The effects androgenic has also been demonstrated with the extract of medicinal Zingiber, of Pentadipandra brazzeana, of Aframomum melegueta and to Pipe guineense14 15. Comitantly, the weight of the animals submitted to the treatment of the aqueous extract of the bark of trunk of B. coriacea (100, 250 and 500 mg/kg, p.o) was measured.

He is to note that this extract provokes a meaningful increase of the weight of the rats. This ponderal gain could explain itself by the increase of the rate plasmatic of the testosterone among these rats. This result corroborates the one of our study on the sharp toxicity that showed a ponderal gain among the mice dealt with the aqueous extract of the bark of this plant to the doses of 2000 and 5000 mg/kg. It is also in agreement with the works of Hamat16 that showed that the administration of extract of Securinega virosa (2ml/kg) drag the increase of the rate plasmatic of the testosterone and a ponderal gain of the male rats. On the other hand, another survey shows that the administration of Hollarena floribunda (150 and 200 mg/kg) among the male rats provokes an increase of the rate plasmatic of the testosterone without increasing the bodily weight of the rats meaningfully17. The action anabolisite of the androgens consists mainly in stimulating the synthesis of the proteins. The accumulation of these proteins to the level of the skeletal muscles and cloths renal and bony explains the ponderal gain among the animals18, to confirm the presence of the substances analogues to the testosterone evolved high here. The results gotten by the reactions of coloration in tubes return the presence of the flavonoids, of the Saponidosides, the cardiac heterosids, the tannins, the steroids and terpenes and the anthocyanes. These results are in agreement with the works of Ep19, on the barks of trunk of the same plant. And this let think that the extract t by the presence of these chemical groups (stérols, flavonoïdes,...) would stimulate the stéroidogenése at the origin of the increase of the rate of testosterone stérique among the rat. This stury shows that our survey on the aphrodisiac activity of the aqueous extract of B. coriacea, comes to confirm the therapeutic use of the plant again in the treatment of the masculine barrenness.

CONCLUSION

The objective of this work was to value the effect androgenique and the determination of the chemical profile of the bark of trunk of B. coriacea. To the term of our study it agrees to say that the barks of trunk of B. coriacea contain some phytodrugs. This plant would be therefore capable to correct the deficit in sexual hormone and to reinforce the fertility the refoire among the male of mammals.

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