

Original Research Article

Phytochemical study and evaluation of the biological activity of anorectic plants used in the Seno province (Burkina Faso).

ABSTRACT

Background: In Africa plants have always been a good source of medicine for health care. Obesity is a pathology that is growing dramatically in developing countries. Anorectic plants are likely to cause a reduction of exaggerated weight gain. The aim of the study is to determine the phenolic compound content of five anorectic potential plants of Burkina Faso (*Ceratotheca sesamoides*, *Gardenia erubescens*, *Brachystelma bingeri*, *Raphionacme daronii* and *Vernonia kotschyana*), to determine also their antioxidant potential and their acetylcholinesterase inhibitory capacity.

Place and Duration of Study: Laboratory of Biochemistry and Applied Chemistry (LABIOCA), Research Institute for Health Sciences (IRSS).

Methodology: For the determination of the acute toxicity of the extracts a group of six (6) mice NMRI race were constituted for each plant extract. A dose of 3000 mg / kg of weight was administered to the animals. The methods of screening were used to detect secondary metabolites like tannins, steroids and terpen, flavonoids, coumarins. For the phenol content, the concentration of total phenolics, flavonoids and tannins were determined. The antioxidant property of the extracts was evaluated in vitro using 2,2-diphenyl-1-picrylhydrazyl acid (DPPH), 2,2-azino-bis (3-ethylbenzthiazoline-6-sufonic) (ABTS) and Ferric Reducing Antioxidant Power (FRAP). The acetylcholinesterase activity of the extracts 0.1 mg / ml was determined by a spectrometric assay method.

Results: Acute toxicity evaluated in NMRI mice showed that the methanolic extracts of five extracts show no toxicity. The coumarins and tannins were detected in all five species of plants. The polyphenol contents of *Ceratotheca sesamoides* gave the highest total phenolic compound content with 221.97 ± 1.206 mg EAG / g and also the best flavonoids content with 39.58 ± 0.068 mg EQ / g. Antioxidant tests show that *Vernonia kotschyana* Sch-Bip and *Ceratotheca sesamoides* Endl presented the best inhibitions of the DPPH radical with $82.63 \pm 3.29\%$ and $83.62 \pm 2.12\%$ at 100µg/ml. This activity is also better than that of quercetin which is a reference substance. For the reducing power of radical cation ABTS \cdot^+ the most active macerates of our extracts were obtained with *Vernonia kotschyana* ($51,388 \pm 0,133$ mmol ET / g extract) and *Ceratotheca sesamoides* ($50,748 \pm 0,395$ mmol ET / g extract). *Ceratotheca sesamoides* showed a best activity on reducing power of the ferric ion (7.03 ± 0.44 mmol EAA / g extract), this activity on ferric ion is superior to that of quercetin, which is a reference substance. *Raphionacme daronii* exhibited the greatest inhibition of acetylcholinesterase with a percentage inhibition of 53.542 ± 4.053 at 100µg / ml.

Conclusion : The study demonstrated that anorexigenic plant extracts have a good antioxidant potential that is necessary for any weight-reducing activity. They also have an ability to inhibit acetylcholinesterase.

Keywords: anorectic plants, antioxidant, acetylcholinesterase activity

14 1. INTRODUCTION

15

16 Since the earliest times, plants have been used by humans first to feed themselves, then to
17 heal themselves. The multiple knowledge accumulated during these past centuries have
18 allowed humans to first distinguish between edible plants and toxic plants and medicinal
19 plants called medicinal plants. In Africa, as in most low-income countries, because of the low
20 accessibility of conventional medicine to populations, more than 80% use traditional
21 medicine for their health care [1]. Several plants are used for the management of metabolic
22 diseases such as obesity. Obesity is a chronic condition characterized by excess body fat
23 that results in increased body weight [2].

24 Today, it is the world's fifth-highest mortality risk factor, with nearly three million people dying
25 each year. This pathology is most often associated with diseases such as hypertension,
26 heart failure, stroke, type II diabetes, insulin resistance, dyslipidemias, certain cancer [3] On
27 the market, drugs Pharmaceuticals are mostly of synthetic origin are used but they have
28 many side effects. Medicinal plants are still an important arsenal for the fight against this
29 disease. Indeed some plants are already known and exploited in this sense. It is recognized
30 that specific chemical constituents such as glycosilated pregnanes [4]. Caffeine [5].),
31 mucilages, phenylalanine [6] [7] [8], hydroxycitric acid [9] found in these plants are
32 responsible for the suppressive effects used to treat the disease.

33 Burkina Faso, like Sahelian countries, has often been confronted in times of famine [10].
34 During these periods of food shortage, people usually resort to plants that have appetite
35 suppressant or thirst-quenching effects. These provide them with satiety, usually without
36 significant energy, which can lead to weight loss. So taking a supplement of these appetite
37 suppressants may help you lose weight by reducing appetite and cravings. This anorectic
38 property could be used in the fight against obesity. *Ceratotheca sesamoides*, *Gardenia*
39 *erubescens*, *Raphionacme daronii*, *Brachystelma bingeri* and *Vernonia kotschyana* are
40 anorectic plants consumed during periods of famine in Burkina Faso [10].

41 So the purpose of our work is to do a phytochemical screening of the five species and to
42 evaluate their biological activities in vitro.

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44 2. MATERIAL AND METHODS

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46 2.1. MATERIEL

47 2.1.1. Plant material

48 The fruits of *Gardenia erubescens*, the leaves of *Ceratotheca sesamoides*, the roots of
49 *Vernonia kotschyana*, the tuber of *Raphionacme daronii* and *Brachystelma bingeri* (Photo 1)
50 were harvested in Dori (locality located 271 km from Ouagadougou in northern Burkina
51 Faso). The species were authenticated by Professor MILLOGO R. Jeanne, botanist at the
52 UFR / SVT of the University of Ouagadougou. Herbarium were deposited at the UFR / SVT
53 under the identification codes of 01ID.16691, 02ID.16693, 03ID16691, 04ID.16692 and
54 05ID.16693 respectively for *Ceratotheca sesamoides*, *Brachystelma bingeri*, *Vernonia*
55 *kotschyana*, *Gardenia erubescens* and *Raphionacme daronii*. The leaves of *Ceratotheca*
56 *sesamoides* and *Vernonia kotschyana* roots were dried under laboratory conditions and then
57 reduced to powder and stored in freezer bags for extractions. The tubers of *Raphionacme*
58 *daronii*, *Brachystelma bingeri*, as well as the fruits of *Gardenia erubescens* were kept in the
59 freezer before extractions.

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(a)

(b)



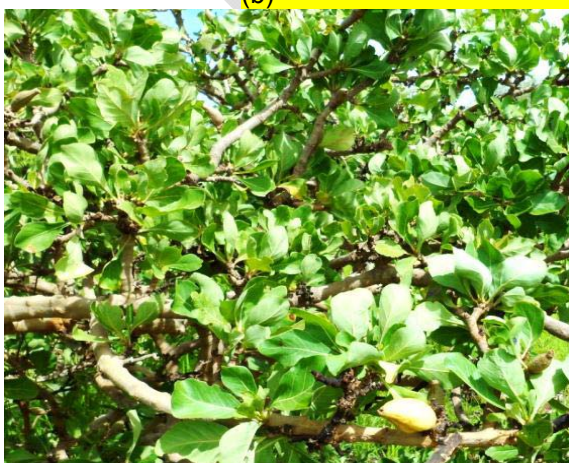
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(b)

(d)



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(e)

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Photo 1: *Vernonia kotschyana* (a), *Ceratotheca sesamoïdes* (b), *Raphionacme daronii* (c), *Brachystelma bingeri* (d), *Gardenia erubescens* (e).

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71 2.1.2. Animal material.

72 White NMRI mice of both sexes between 7 and 8 weeks of age and body weight between 17
73 and 39 grams were used for the study. They come from the UFR / SVT animal shop of
74 University of Ouagadougou. They were raised under the following conditions:

- 75 - Food granules feed at 29% protein; running water from town;
- 76 - Stabulation at a temperature of 25 ° C; humidity level 30%

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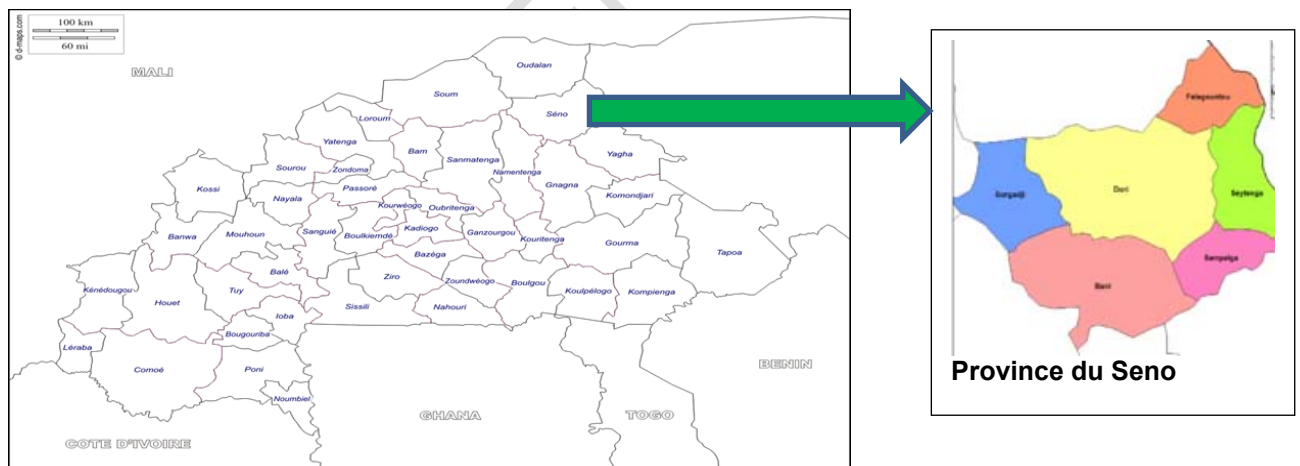
78 2.2. METHODS

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80 2.2.1. Study area: Dori

81 Seno Province, whose capital is Dori (Fig 1), is located in the north eastern area of Burkina
82 Faso. It has 215 villages and an area of 6979 km² with a population of 264,815 people [8].
83 This locality has a Sahelian climate, characterized by a long dry season (May to October)
84 and a short rainy season (average rainfall of 400 mm), with varying temperatures (10–43°C),
85 low humidity, wind and a large amounts of sunshine, typical of the Sahel. The vegetation is
86 characterized by wooded and shrubby steppe that is heavily damaged. However, there are a
87 few gallery forests which are generally located along the rivers (like the swamp of Dori or the
88 Yakouta River). The dominant types of vegetation are thorn trees [11]. Famine is recurrent in
89 this province. The predominant population is the Fulani group, who are nomadic herders.
90 They have survived drought in this region through their knowledge of appetite suppressing
91 plants. *Ceratotheca sesamoides*, *Gardenia erubescens*, *Brachystelma bingeri* *Raphionacme*
92 *daronii* and *Vernonia kotschyana* are five plants use as anorectic plant

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Burkina Faso
Fig.1. Study area

100 2.2.2. Extraction

101 The samples were extracted by methanolic maceration and 25 g of powder of *Ceratotheca*
102 *sesamoides* leaves and roots of *Vernonia kotschyana* were extracted in 250 ml of methanol.
103 For *Gardenia erubescens*, *Brachystelma bingeri*, *Raphionacme daronii*, the fruit and tuber
104 pulps previously stored in the freezer were milled, then 25 g of the ground material of each
105 sample is put into 250 ml of methanol. These different mixtures obtained were stirred
106 magnetically for 24 hours. The extracts obtained are concentrated using a rotary evaporator
equipped with a vacuum pump. The dry extract obtained was used for the different tests.

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108 **2.2.3. Acute toxicity of extracts**

109 The toxicity was determined according to the method described by OCDE [12].

110 2.2.3.1. Distribution of mice

111 The animals were divided into five (5) lots of six (6) mice. Each animal is identified by a
112 different mark. The animals are pre-fasted for 16 hours, then the weight of each mouse is
113 taken, and they receive a dose of plant extract given by batch. After a follow-up for 72 hours,
114 the mortality in each batch was determined.

115 2.2.3.2. Administration of the extract

116 Extracts were administered by gavage (oral) using an esophageal tube. For the evaluation of
117 the acute toxicity of the extracts, the 5 lots of 6 mice received a single dose limit of 3000 mg
118 / kg of plant extract once at the beginning of the experiment. The extracts were administered
119 to the animals for a volume not exceeding 0.5 ml.

120 2.2.3.3. Animal Tracking

121 After the administration of the extract, the animals were observed for 2 hours for the
122 evaluation of signs of intoxication (toxidrome). After having restored a normal diet (water,
123 granules), the animals were then observed at 24, 48 and 72 hours after which the cumulative
124 number of deaths in each batch was noted.

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126 **2.2.3. Phytochemical studies**

127 2.2.3.1. Screening test for secondary metabolites

128 The purpose of the tests is to detect the main phytochemicals present in plant extracts.
129 These tests were performed on the extracts of the plant studied. The procedures described
130 by Ciulei [13] have been used for the demonstration of the different chemical groups. So:

- 131 • The reaction with iron trichloride (FeCl_3) is used for the detection of tannins and
132 polyphenols,
133 • The Shibata test for flavonoids,
134 • The Feiggl-Frehden test for coumarins,
135 • The Liebermann / Buchard test for triterpenes / steroids,
136 • The foam test for saponosides.

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138 2.2.3.1. Determination of polyphenols

139 Total phenolics were estimated by the Singleton method [14]. It evaluates all the phenolic
140 compounds that reduce the phosphomolybdotungstic reagent (Folin-Ciocalteu reagent).
141 Thus the content of the total phenolics is determined by extrapolation on a standard curve
142 obtained with gallic acid (200 mg / l). In each test tube were added, according to the
143 solutions obtained after dilution, 0.125 ml of the sample to be assayed (gallic acid or sample)
144 and 0.625 ml of Folin Ciocalteu FCR reagent (0.2 N in distilled water). After waiting for 5
145 minutes, 0.5 ml of sodium carbonate (75 g / l) was added. After stirring, the various solutions
146 were allowed to stand in the dark for 2 hours. The reading was made using a
147 spectrophotometer at 760 nm against a blank consisting of a mixture of 0.5 ml of FCR and
148 0.5 ml of sodium carbonate. Three readings are made per sample. The total phenolic
149 content is expressed in mg Equivalent of Gallic Acid (EGA) per 100 mg of solids.

150 2.2.3.2. Determination of flavonoids

151 The contents of the flavonoids were determined by the method by Arvouet Grand [15]. The
152 method evaluates all compounds reacting with aluminum chloride (AlCl_3). A volume of 0.75
153 ml of 2% AlCl_3 (in analytical methanol) is mixed with an equal volume of extract according to
154 the dilution obtained (1/10 or 1/100) in methanol. The optical densities were read after 10
155 minutes of incubation at 415 nm using a spectrophotometer against a calibration curve
156 previously drawn. The calibration curve is plotted using quercetin as a reference from a
157 dilution. Three readings were performed per sample and the results are expressed in mg
158 Equivalent Quercetin (EQ) per 1g of extract (mg EQ/ 1g).

159 2.2.3.3. Tannin dosage

160 The tannin contents of the samples were determined using the method of the European
161 Commission [16]. A mixture of 1 ml of water, 0.2 ml of extract according to the dilution
162 obtained with 0.2 ml of ferric ammonium citrate (CAF) with a concentration of 3.5 mg / ml in
163 water and 0.2 ml of NH_4OH 8mg / ml concentration in water is performed. The
164 concentrations are read after 15 minutes of incubation at 525 nm using a spectrophotometer
165 against a standard curve previously drawn using the tannic acid used as a reference
166 substance. Three readings are carried out for each sample and the results are expressed in
167 mg Tannic acid equivalent (E.A.T) per 1g of dry extract (mg EAT / 1g).

168 2.2.4. Biological activities

170 2.2.4.1. Antioxidant activity

171 a. DPPH (2,2diphenyl-1-picrylhydrazyl) method

172 The anti-radical activity of the extract (1 mg/ml) was evaluated by the DPPH (2,2diphenyl-1-
173 picrylhydrazyl) method [17]. This method is based on the reduction in absorbance at 517 nm
174 of the stable free radical DPPH, in the presence of a hydrogen radical donor (Koleva et al.,
175 2002) three (03) tests were carried out by mixing 100 μl of the sample and 200 μl of DPPH
176 (20 mg / l in methanol). After 15 minutes of incubation, the absorbance is read at 517 nm
177 against a blank (100 μL of methanol and 200 μL of DPPH) using a spectrophotometer.
178 Quercetin was used as reference substances. The antiradical activity was expressed in
179 percent inhibition.

180 b. ABTS [2,2'-azinobis- (3-ethylbenzothiazoline-6-sulfonic acid)] method

181 It is based on the discoloration of the stable radical cation ABTS^+ [2,2'-azinobis- (3-
182 ethylbenzothiazoline-6-sulfonic acid)], in ABTS, in the presence of antiradical compounds.
183 The monitoring is done by measuring the absorbance at 734 nm because the chromophoric
184 radical cation ABTS^+ blue-green color produced by reaction of ABTS with potassium
185 persulfate at λ_{max} at 734 nm. The method of Re et al. (1999) [18]. was used.
186 Preparation of the ABTS solution: A mass of 10 mg of ABTS was dissolved in 2.6 ml of
187 distilled water. 1.7212 mg of potassium persulfate is added and the mixture is kept in the
188 dark at room temperature for 12 hours. The mixture is then diluted in ethanol so as to obtain
189 an absorbance of 0.70 ± 0.02 at 734 nm.

190 Test on the samples: In 3 eppendorf tubes containing 10 μl of sample solution (1 mg / ml)
191 were added to 990 μl of ABTS solution. + freshly prepared. The same operation was carried
192 out for the Trolox used as reference. The whole is protected from light for 15 minutes and
193 the absorbances are read at 734 nm spectrophotometer against a standard Trolox curve.

194 The concentration of compounds having a reducing effect on the radical cation ABTS^{•+}
195 (antiradical compounds) is expressed in mmol Trolox equivalent (mmET) / g of dry extract

196 c. Reducing power FRAP (Ferric reducing antioxidant power)

197 The ferric ion (Fe³⁺) to ferrous ion (Fe²⁺) by reducing compounds follows an electron mono
198 electron transfer [19]. In test tube containing 0.5 ml of extract (1 mg / ml), 1.25 ml of
199 phosphate buffer (0.2 M, pH 6.6) and 1.25 ml of potassium hexacyanoferrate (1% aqueous)
200 were added. The mixture was heated at 50 ° C in a bain-marie for 30 minutes. After cooling,
201 trichloroacetic acid (1.25 mL, 10%) was added, and the mixture was then centrifuged (2000
202 rpm for 10 minutes). Three aliquots (125 µl) of the supernatant were transferred to 96-well
203 microplate to which 125 µl of distilled water and then 25 µl of FeCl₃ (0.1% aqueous) were
204 added. The reductive power was evaluated at 700 nm against a standard curve of ascorbic
205 acid using a spectrophotometer (Epoch 251465, Biotek Instruments, USA). The experiment
206 is carried out in triplicate (independent tests), and the reduced activity of the extract is
207 expressed in mmol Equivalent Ascorbic acid per gram of extract (mmol EAA / g extract).
208 Quercetin was used as reference substances.

209 2.2.4.2. Inhibition of acetylcholine esterase

210 The inhibitory activity of the extracts was evaluated using the procedure described by Lopez
211 [20]. 100 µl of sample (0.1 mg / ml in 50 mM Tris-HCl buffer, pH 8, 10% methanol) were
212 mixed with 100 µl of AChE (0.22 U / ml in 50 mM Tris-HCl buffer). HCl, pH 8, 0.1% BSA) and
213 200 µl of buffer (50 mM Tris-HCl, pH 8, 0.1% BSA). The mixture was incubated for 5 minutes
214 at 30 ° C in a 1 ml vat. 500 µl of DTNB (3 mM in TrisHCl buffer, pH 8, 0.1 M NaCl, 0.02 M
215 MgCl₂) and 100 µl of ATCI (15 mM in water) were added thereafter. A blank was also
216 prepared under the same conditions by replacing AChE with 100 µl of buffer (50 mM Tris-
217 HCl, pH 8, 0.1% BSA). The reaction was monitored for 5 minutes at 405 nm using a
218 spectrophotometer Buffer (0.1% in 50 mM Tris-HCl, pH 8, 10% methanol) been used as a
219 negative control. Anti-acetylcholinesterase activity (I%) is expressed as percentage inhibition

220 2.2.5. statistical analyzes

221 For statistical analyzes, Microsoft Excel was used to obtain standard curves and graphs,
222 percentages of inhibition, averages, and standard deviation of results. One-way ANOVA
223 followed by the Turkey test was used to measure the degree of statistical significance of the
224 results using the XL stat module. A significant difference is considered for p <0.05 .
225

226 3. RESULTS AND DISCUSSION

227 3.1. RESULTS

228 3.1.1. Acute toxicity of extracts

231 The results showed that up to 3000 mg / kg of body weight extracts of *Gardenia erubescens*,
232 *Ceratotheca sesamoïdes*, *Vernonia kotschyana*, *Raphionacme daronii* and *Brachystelma*
233 *bingeri* showed no mortality (Table 1). The LD₅₀ values are therefore greater than 3000 mg /
234 kg of body weight.
235

236 Table 1. Acute toxicity of extracts

237

Extraits	Doses (mg/kg)	Number of mice	Average weight (g)	Number of dead	% of mortality	toxidromes
<i>Vernonia kotschyana</i>	3000	6	29,5±5,8	00	00	reduced displacement
<i>Gardenia erubescens</i>	3000	6	26,9±5,5	00	00	Agitation
<i>Ceratotheca sesamoïdes</i>	3000	6	22 ± 3,65	00	00	Agitation
<i>Brachystelma bingeri</i>	3000	6	23,33 ±1,36	00	00	Agitation
<i>Raphionacme. daronii</i>	3000	6	25,33 ±1,36	00	00	Agitation

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240 3.1.2. Results of Screening test

241 We can note that two main groups of compounds were found in all these plant extracts
 242 namely the group of sterols and triterpenes and coumarins. flavonoids were detected only in
 243 *Gardenia erubescens* and *Ceratotheca sesamoïdes* extract (Table 2)

244 **Table 2. Screening test**

species	<i>Ceratotheca sesamoïdes</i>	<i>Gardenia erubescens</i>	<i>Raphionacme daronii</i>	<i>Brachystelma bingeri</i>	<i>Vernonia kotschyana</i>
Test					
Saponosids	+	-	+	+	+
Tannin and polyphénols	+	-	-	-	-
Flavonoïds	+	+	-	-	-
Steroids and triterpènes	+	+	+	+	+
Coumarins	+	+	+	+	+

245 += Presence - = Absence

246 3.1.3. Phenolic content

247 The overall results of total phenolics, flavonoids, flavonols and tannins are recorded in Table
 248 3. We find that the extract of *Ceratotheca sesamoïdes* which has a content of 221.97 ±
 249 1.206 (mg EAG / 1g), is the richest in phenolic compounds than the other four extracts. The
 250 lowest content of phenolic compounds was obtained with *Brachystelma bingeri* extract
 251 (01.70 ± 0.090mg EAG / 1g), a content is not statistically different from that of *Gardenia*
 252 *erubescens* and *Raphionacme daronii*. With regard to the total flavonoid assay, the extract of

253 *Ceratotheca sesamoides* is the only one with a content of 39.58 ± 0.068 (mg EQ / 1g of
254 extract) and for the other plants no content was detected

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256
257

Table 3. Phenolic contents

Contents species	Total Phenolic (mg EAG/1g)	Total flavonoids (mg EQ/1g)	Total tannins (mg EAT/1g)
<i>Brachystelma bingeri</i> . A	$01,70 \pm 0,090^c$	Traces	Traces
<i>Ceratotheca sesamoides</i> . Endl	$221,97 \pm 1,206^a$	$39,58 \pm 0,068$	Traces
<i>Gardenia erubescens</i> .	$14,55 \pm 0,106^c$	Traces	Traces
<i>Raphionacme daronii</i> .	$05,26 \pm 0,256^c$	Traces	Traces
<i>Vernonia kotschyana</i>	$43,84 \pm 0,178^b$	Traces	Traces

258 Results indicated by different letters are statistically distinct ($p < 0.05$)
259 Mean \pm S.E.M = Mean values \pm Standard error of means of three experiments.
260

261 3.1.4. Biological activities

262 3.1.4.1. Antioxidant activity

263 a) Inhibition of the radical DPPH

264 *V. kotschyana* and *C. sesamoides* presented the best inhibitions of the DPPH radical with
265 $82.63 \pm 3.29\%$ and $83.62 \pm 2.12\%$ at $100\mu\text{g/ml}$. The activity of these species is also better
266 than that of quercetin which is a reference substance. *Gardenia erubescens*, *Brachystelma*
267 *bingeri* and *Raphionacme daronii* had the lowest anti-radical activity (Table 4)

268 **Table 4. Results of DPPH activity**

Espèce	<i>V. kotschyana</i>	<i>C. sesamoides</i>	<i>G. erubescens</i>	<i>B. bingeri</i>	<i>R. daronii</i>	Quercetin
Inhibition (%)	$82,63 \pm 3,29^a$	$83,62 \pm 2,12^a$	32.95 ± 1.45^b	06.39 ± 0.03^c	8.57 ± 0.029^c	82.17 ± 0.30^a

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270 Different letters in the same column indicate significance difference ($p < 0.05$)
271 Mean \pm S.E.M = Mean values \pm Standard error of means of three experiments.
272

273 b) Activity on the ABTS

274 The reducing power of the radical cation ABTS⁺ obtained is 51.388 ± 0.133 mmol ET / g of
275 extract; 50.748 ± 0.395 mmol ET / g; 33.544 ± 0.213 mmol ET / g extract; 32.954 ± 0.707
276 mmol ET / g extract and 31.881 ± 0.585 mmol ET / g extract respectively for extracts of
277 *Vernonia kotschyana*, *Ceratotheca sesamoides*, *Gardenia erubescens*, *Brachystelma bingeri*
278 and *Raphionacme daronii*. Thus we note that by this method, the most active macerates of
279 our extracts were obtained with *Vernonia kotschyana* ($51,388 \pm 0,133$ mmol ET / g extract)
280 and *Ceratotheca sesamoides* ($50,748 \pm 0,395$ mmol ET / g extract) but these activities are
281 less than quercetin used as a reference who gave 69.00 ± 1.41 mmol ET / g extract (fig 2).

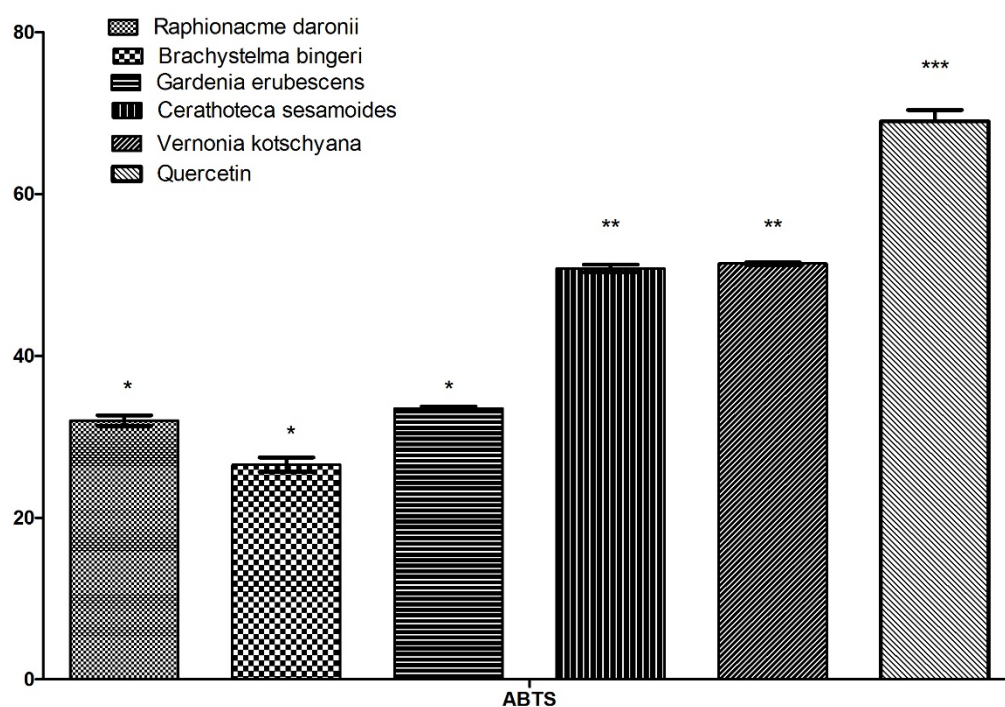


Fig. 2. Effect of extract on ABTS

***P-value is significant at $p < 0.05$

C) Reducing activity (FRAP)

The FRAP method evaluates all compounds capable of reducing ferric ion by the transfer of an electron. The values expressed in (mmol EAA / g) are as follows: *C. sesamoides* (7.03 ± 0.44); *V. kotschyana* (1.44 ± 0.08); *R. daronii* (0.015 ± 0.001); *B. bingeri* (0.013 ± 0.004); *G. erubescens* (0.012 ± 0.003). We note through these values that the highest reducing power was obtained with the extract of *Ceratotecha sesamoides* (7.03 ± 0.44 mmol EAA / g extract), this activity is superior to that of quercetin, which is a reference substance (table 5).

Table 5. Results of reducing activity (FRAP)

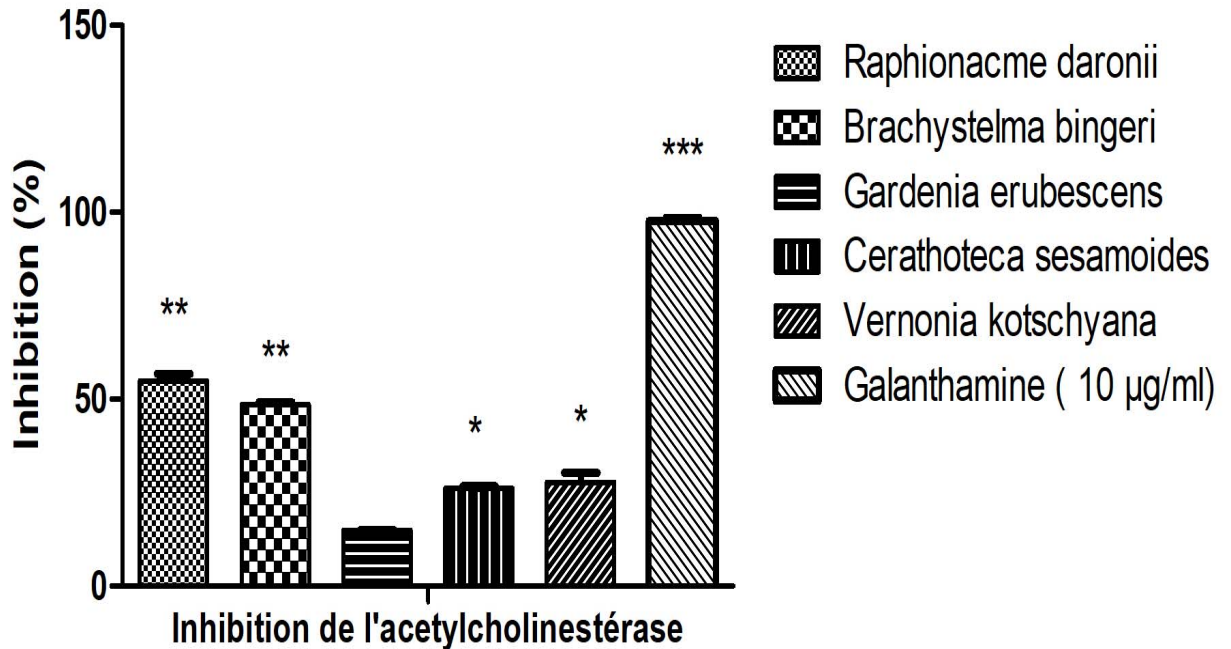
Species	<i>V. kotschyana</i>	<i>C. sesamoides</i>	<i>G. erubescens</i>	<i>B. bingeri</i>	<i>R. daronii</i>	Quercetin
Antioxidant capacity (mmol EAA/g d'extract)	$1,44 \pm 0,08^c$	$7,03 \pm 0,44^a$	$0,012 \pm 0,003^d$	$0,013 \pm 0,004^d$	$0,015 \pm 0,001^c$	4.69 ± 0.05^b

Different letters (a, b, c, d) in the column indicate significance difference ($p < 0.05$)

Mean \pm S.E.M = Mean values \pm Standard error of means of three experiments.

301 3.1.4.2. Inhibition of acetylcholinesterase

302 The extract of *R. daronii* and *B. bingeri* showed the highest acetylcholinesterase inhibitions
303 with percentages of $53.542 \pm 4.053\%$ and 48.188 ± 6.106 at the concentration of $100 \mu\text{g} / \text{ml}$
304 and the lowest inhibition was obtained with the extract of *G. erubescens* ($14.88 \pm 2.616\%$ at
305 a concentration of $100 \mu\text{g} / \text{ml}$) (fig 3).
306



307
308 **Fig. 3. Acetylcholinesterase inhibition**

309
310 Mean \pm S.E.M = Mean values \pm Standard error of means of three experiments.

311 3.2. DISCUSSION

312 The extracts of *Gardenia erubescens*, *Ceratoteca sesamoides*, *Vernonia kotschyana*, *R.*
313 *daronii* and *Brachystelma bingeri* showed no mortality at $3000 \text{ mg} / \text{kg}$ body weight.
314 Considering the toxicity scale of Hodge and Sterner [21]., macerates of our species are not
315 toxic orally in NMRI mice. This low toxicity could justify the fact that these species are
316 consumed by the populations.

317 The five species showed high levels of total polyphenols and showed the presence of
318 flavonoids. Indeed these compounds endowed with anorectic activity. Black tea polyphenols
319 are able to reduce weight gain through their appetite suppressant effects [22] [23]. The
320 presence of these compounds in plant extracts may explain the traditional use of these plant
321 species as anorectic plants. Also the presence of inulin in the tuberous roots of *V. kotschyana*
322 [24], of saponosides, steroids, triterpenes in *Brachystelma bingeri*, *Gardenia erubescens* and
323 *Ceratoteca sesamoides* [25] [26].) could justify their uses because these molecules have
324 an anorectic potential [27].

325 The extracts also gave a good antioxidant potential. They showed an ability to reduce the
326 DPPH radical, neutralize the ABTS radical cation and reduce the ferric ion. *V. kotschyana*
327 and *C. sesamoides* presented the best inhibitions of the DPPH radical with $82.63 \pm 3.29\%$
328 and $83.62 \pm 2.12\%$ at $100\mu\text{g}/\text{ml}$. These extracts are therefore a good way to fight against
329 oxidative stress. Indeed, obesity is associated with an increase in reactive oxygen species

(responsible for oxidative stress) due to the presence of excess adipose tissue. Adipocytes and preadipocytes have been identified as a source of pro-inflammatory cytokines, including TNF- α , IL-1 and IL-6. These cytokines are potent stimulators for reactive oxygen species (ORS) production by macrophages and monocytes; therefore, an increase in cytokine concentration may be responsible for an increase in reactive oxygen species (ORS). Oxidative stress can be a cause and consequence of obesity. Polyphenols have good antioxidant capabilities, they have a wide range of biological actions, such as free radical scavenging, metal chelation, and enzyme modulation capabilities [28]. The presence of polyphenols in the extracts could explain the good antioxidant activity observed. Flavonoids are also endowed with antioxidant activity. They are mainly recommended for their antioxidant action. Some flavonoids have the ability to chelate metal ions such as Fe²⁺ and Cu²⁺ which play a vital role in oxygen metabolism and free radicals. They are also able to chelate free radicals immediately by giving a hydrogen atom or a single electron transfer. Thus the complete mode of action of flavonoids includes: the extinction of the element free radical, chelating the metal, suppressing the enzymes associated with the generation of free radicals. Quercetin, kaempferol, naringenin and hesperidin are examples of antioxidant activities [29] [30]. The presence of polyphenol and flavonoids could explain the good antioxidant activity. The antioxidant activity of these anorectic species could be used in the fight against oxidative stress diseases most often associated with obesity. All our extracts showed an interesting acetylcholinesterase inhibitory activity but low compared to galanthamine which is a reference inhibitor compound of acetylcholinesterase with an inhibition of $98.28 \pm 1.52\%$ at $10 \mu\text{g} / \text{ml}$ [31]. However, *R. daronii* and *B. bingeri* with inhibition percentages of $53.542 \pm 4.053\%$ and 48.188 ± 6.106 are potential sources of inhibitor of acetylcholinesterase activity and could be used for the search for treatments for related diseases. oxidative stress [32]. Inhibition of acetylcholinesterase is also a strategy for the treatment of Alzheimer's disease, senile dementia, ataxia, myasthenia, and Parkinson's disease [33]. These plant extracts in addition to their potential anorectic could be used in the fight against Parkinson's disease.

4. CONCLUSION

The results of this study show that *Ceratotheca sesamoïdes*, *Gardenia erubescens*, *Brachystelma bingeri* *Raphionacme daronii* and *Vernonia kotschyana*, anorectic species consumed in Burkina Faso, are not acutely toxic. The methanolic extract of these plants also has a good antioxidant potential. Antioxidant capacity is necessary in the anti-obesity activity of an extract. So these species traditionally used as anorectic plants may have a good ability to reduce body weight..

COMPETING INTERESTS

Authors have declared that no competing interests exist.

ETHICAL APPROVAL (WHERE EVER APPLICABLE)

All experimental animal protocols had complied with the instructions of the Institutional Animal Ethics Committee (directive 2010/63/EU on protection of animals used for scientific purposes). Ethical approval code: 2010/63/EU, Date of approval: 20 October 2010. The institutional animal ethical guidelines were strictly observed. All authors hereby declare that "Principles of laboratory animal care were followed, as well as specific national laws where applicable

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