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Phytochemical analysis and acute toxicity of two medicinal plants (*Anogeissus leiocarpus* and *Daniellia oliveri*) used in traditional veterinary medicine in Burkina Faso

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ABSTRACT

The traditional veterinary pharmacopoeia is a common practice of smallholders and pastoralists to treat small ruminant gastro-intestinal parasitism in the Central Plateau of Burkina Faso. The remedies made from plants such as Anogeissus leiocarpus and Daniellia oliveri are commonly used to treat this disease. In this study, surveys were conducted to understand the traditional practice use of both plants, to identify their phytochemical composition and to evaluate their acute toxicity in mice. The results showed that leaves and stem bark are plant parts used for A. leiocarpus and D. oliveri respectively. They are traditionally used as a decoction of which the lyophilized product is administered orally to small ruminants at a rate of 160 mg/kg for A. leiocarpus leaves and 242.5 mg/kg for D. oliveri stem bark. In both plants, flavonoids, tannins and polyphenols are present. On the other hand, the lethal doses (LD_{50}) obtained were 2403.6 mg/kg for A. leiocarpus leaves extract and more than 3500 mg /kg for D. oliveri stem bark extract. Thus, these plant extracts can possibly use as alternative products in the treatment of animal diseases and the result of this study can be used as the basis of clinical studies on animals.

INTRODUCTION

In Burkina Faso, the small ruminant livestock play an important role for rural farmers in the daily struggle for food security and income growth needed to ensure the education of children and family health [1]. The mode of animal breeding in those farms is mostly traditional and the

main objectives of rural farmers are herd numerical productivity and meat production, which pass through the control of diseases as gastrointestinal parasites. Indeed, these parasites cause enormous losses to farmers through rural high mortalities in young and decreases animal performance in adults. In practice, the use of chemical anthelmintics is the appropriate method to fight gastrointestinal parasites in the animal infested. Unfortunately, these costs are high and modern products out of reach of rural farmers. Faced with this situation, these farmers have adopted alternative solutions based on the use of medicinal plants to treat animal diseases [2] [3]. Among these plants, *Anogeissus leiocarpus* (DC.) Guill. and Perr. and *Daniellia oliveri* (Rolfe) Hucht. and Dalza. are commonly used to treat small ruminant gastrointestinal parasitism in the central region of Burkina Faso [4]. These plants are widely distributed in Burkina Faso and have diverse traditional medicinal use to treat human and animal diseases [3] as many medicinal plants that are natural sources of compounds used against many diseases thought the world [5]. This study was carried out (i) to describe the first case of the use of those two plants in traditional veterinary in Burkina Faso, (ii) to realize their phytochemical screening and (iii) to determine their acute toxicity level (LD_{50}) in mice.

MATERIALS AND METHODS

1 - Field surveys

The results of ethno-veterinary exploratory surveys carried out in the central plateau of Burkina Faso by 101 farmers and 21 traditional healers have been used for this study. To this end, we are interested in results for the uses of *A. leiocarpus* and *D. oliveri* used by traditional healers in the treatment of gastrointestinal parasitism in small ruminants [4].

2 - Tests of toxicity and phytochemical analysis

2.1-Plant material and preparation of plant extracts

A. leiocarpus leaves and *D. oliveri* stem bark used by traditional healers in Burkina Faso are collected from experimental station in Kamboinsé, Institute for Environmental and Agricultural Research. The plant specimens were identified at the herbarium of National Centre for Scientific Research and Technology, Ouagadougou.

Both plant material (leaves and stem bark) were then washed with water and dried to obtain powder. 100 g of each powder are used to make a decoction in one liter of distilled water during one hour. The obtained extract of each plant is filtrated, lyophilized and kept in a drier until being reconstituted for phytochemical screening and toxicity test.

2.2- Phytochemical screening of aqueous extracts

The lyophilised decoction of *A. leiocarpus* leaves and *D. oliveri* stem bark were subjected to various tests to identify the major groups of secondary metabolites. To this end, the test tube procedures were used [6].

2.3- Acute toxicity of extracts

The method adapted from Litchfield and Wilcoxon [7] were used for acute toxicity assessment on both sex mice NMRI, weighing 35 ± 5.6 g. During the test, mice were put in the laboratory conditions: standardised boxes, food made of granules containing 20% of proteins, water ad libitum, natural light and ambient temperature at 25-30°C.

For each extract, seven groups were used including six groups receiving six different concentrations (500, 1000, 2000, 2500, 3000 and 3500 mg / kg body weight) and one untreated control group that received distilled water. For this, animals of each group were receiving a

specific dose of the extract to be tested and distilled water by oral route using intragastric syringe. Then, all mice were observed systematically during 72 hours for intoxication syndromes and number of death. The number of mice died was recorded and used in the calculation of the acute toxicity value (LD_{50}).

3 - Statistical analysis

The results of the chemical composition characterization of two extract plants were notified negative (-) or + (positive).

The lethality of mice was estimated as a percentage of deaths observed during the test for each dose of extract. SPSS (version 10.0.5) for Windows (95, 98 and NT) was used to determine by probit analysis the values of DL_1, DL_5, DL_{50}, DL_{95} and DL_{99} and the relative ratios DL_5/DL_{50}, DL_{50}/DL_{95} and DL_5/DL_{95}. The scale of Hodge and Sterner [8] were used to characterize the safety level of each plant extract.

RESULTS

1 - Traditional method use

The extraction yields of *A. leiocarpus* leaves and *D. olivieri* stem bark aqueous decoctions prepared according the traditional healers procedure were respectively 8% and 9.7%.

The Table 1 show the traditional method use of two plant extracts in the treatment of small ruminant gastrointestinal parasitism in central region of Burkina Faso. From our estimations according traditional healer recommendations, doses administered by traditional healers to small ruminant are follows by extrapolation:

- *A. leiocarpus*: a handful (estimated at 52 g) of powder of dried leaves per animal (25 kg average live weight) and orally, once daily for 3 days (52 g x 3) or 156 g per animal for the duration of treatment. These doses are equivalent to about 2 g of dry matter per kg of body weight per day or 160 mg of lyophilized dry matter per kg of body weight per day of treatment;

- *D. oliveri*: a handful (estimated at 63 g) powdered dried bark of stems per animal (25 kg average live weight) and orally, once daily for 3 days (63 g x 3) or 189 g per animal for the duration of treatment. These doses are equivalent to about 2.5 g of dry matter per kg of body weight per day or 242.5 mg of lyophilized dry matter per kg of body weight per day of treatment.

Table 1: Traditional method uses of *A. leiocarpus* leaves and D. oliveri stem bark in the treatment of small ruminant gastrointestinal parasites of in Burkina Faso

Plant extracts	Route and method of administration of plant extracts
Anogeissus leiocarpus leaves	orally with a bottle of 66 cl for 3 consecutive days
Daniella oliveri stem bark	

2 – Chemical composition of extract plants

The Table 2 presents the result of chemical characterization of both two extracts. *D. oliveri* stem bark contains more chemicals than *A leiocarpus* leaves. Indeed, *A leiocarpus* leaves contains tannins and flavonoids and *D oliveri* stem bark shows the presence of flavonoids, tannins, saponin and steroids/triterpenes.

Table 2: Phytochemical screening of aqueous extracts of *A. leiocarpus* leaves and *D. oliveri* stem bark

Chemical Groups	Aqueous extracts	
	A. leiocarpus leaves	*D. oliveri* stem bark
Flavonoids	(+)	(+)
Tannins and polyphenols	(+)	(+)
Saponins	(-)	(+)
Coumarins	(-)	(-)
Steroids / triterpenes	(-)	(+)
Alkaloids	(-)	(-)

(+) : présence *(-) : absence*

1 - Acute Toxicity of plant extracts in mice

Table 3 and 4 present the results of acute toxicity of *A. leiocarpus* leaves and *D. oliveri* stem bark extract in mice. The different groups treated with *D. oliveri* stem bark extract showed no specific abnormalities signs and mortalities like the control group. The all animals move and fed normally. The LD_{50} value of *D. oliveri* stem bark extract was estimated at greater than 3500 mg / kg. On the other hand, the behavior of mice has changed at the dose of 2000 mg/kg with *A. leiocarpus* leaves extract. Mice showed incorrect signs (fatigue and loss of appetite) and a mortality. All animals were died at the dose of 3500 mg / kg. LD_{50} value calculated by-probit analysis was 2303.6 mg / kg body weight within 95% confidence limits (1899.0 – 2651.4 mg / kg) (Table 4).

Table 3: Acute toxicity of the extract of *A. leiocarpus* leaves and *D. oliveri* stem bark in mice by oral route after 72 hours of observation

Number of mice	Dosis (mg/kg)	*A. leiocarpus* leaves		*D. oliveri* stem bark	
		Number of mice died	Percentage of mice died	Number of mice died	Percentage of mice died
6	500	0	0	0	0
6	1000	0	0	0	0
6	2000	1	16	0	0
6	1500	4	66	0	0
6	3000	5	83	0	0
6	3500	6	100	0	0

Table 4: Lethal doses values (mg/kg) of *A. leiocarpus* leaves aqueous extract after 72 hours and intoxication syndromes in mice

lethal doses (mg/kg)	Quotient of lethal doses	Intoxication syndromes
LD_1 = 1655.525 LD_5 = 1823.760 LD_{50} = 2303.649 LD_{95} = 2909.811 LD_{99} = 3205.08	LD_5/LD_{50} = 0.79 LD_{50}/LD_{95} = 0.79 LD_5/LD_{95} = 0.62	asthenia and lack of appetite

DISCUSSION

Our study was conducted to identify the major chemical groups contained in *A. leiocarpus* leaves and D. *oliveri* stem bark extracts used by traditional healers of Burkina Faso against gastrointestinal parasites of small ruminants and to establish their toxicological limits use in mice.

The unit of measurement used (handle) by traditional healers to administer medicines prescribed illustrates the existence of therapeutic and toxic doses concepts in the system of traditional

animal health in our study. This observation was also made in Fouta Djallon (Guinea) at traditional healers by Barry et *al.* [9]. Although imprecise, this traditional unit concept of measurement has the advantage to guide the user in the treatment of animal diseases in farming areas.

During the toxicological study of *D. oliveri* aqueous extract, the mice behaved normally. Indeed, neither their mobility nor their breathing and feeding have been modified. The LD_{50} of this extract is greater than 3500 mg / kg by oral route. Consequently, the decoction of *D. oliveri* stem bark extract in our study could be classified as slightly toxic on the scale of Hodge and Sterner [8]. These results are comparable to those of Ahmadu et *al.* [10] who obtained an LD_{50} of 4 000 mg / kg with ethanol extract of *D. oliveri* leaves in mice by intraperitoneal route in Nigeria. However, our results are different from those of Jegede et *al.* [11]. Indeed, these authors noted behavioral problems at a dose of 500 mg / kg and a LD_{50} of 447.2 mg / kg with a macerated aqueous extract of stem bark in intraperitoneal route.

As the aqueous extract of *A. leiocarpus* leaves, the LD50 was estimated at 2303.6 mg / kg body weight of mice treated orally. The ratios of LD5/LD50 and JD50/LD95 are quite equal for the plant studied, confirming the validity of our LD50 test-value [12]. Thus, this extract of *A. leiocarpus* can be considered as low toxicity because the LD_{50} is between 500 and 5000 mg [8]. In Nigeria, Agai et *al.* [13] observed no mortality in albino rats treated with increasing doses (800 to 3200 mg / kg) of aqueous extract of *A.. leiocarpus* leaves unlike our results in mice. These authors obtained an LD_{50} of 1400 mg / kg in rats treated with the same extract of *A. leiocarpus* by intra-peritoneal route. These results confirm the low toxicity obtained in our study of the aqueous decoction of *A. leiocarpus* leaves.

In the context of our study, the safe use of tested extracts and active substances that they contain, explain probably their common uses by traditional healers in the treatment of numerous human and animal diseases [3 [14] [15]. Indeed, flavonoids and polyphenolic compounds (polyphenols and tannins) were observed in extracts of both plants. In more, saponins and steroids/triterpenes were found in *D. oliveri* extract. This difference of chemical composition between the two extract plants suggests that there are probably more polar compounds in *D. oliveri* stem bark than in *Anogeissus leiocarpus* leaves. The therapeutic properties of these large chemical groups have been reported by various authors [16] [17]. According Arbonnier [18], all parts of *D. oliveri* are exploited in traditional medicines. The macerated extract of the plant stem bark presents an anti inflammatory effect in rats [11]. The methanol extract of the plant stem bark causes smooth muscle relaxant effect [19] and the n-butanol extract of plant leaves possess an anti-diarrheal effect in mice [10]. Also, *A. leiocarpus* leaves possess antiplasmodium [20] and antitussive [21] activities. Besides, the terpenoidal fraction isolated from *A. leiocarpus* have antibacterial activity against *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* [22].

In conclusion, the toxicity study which is essential for an adaptation of the traditional medicine was conducted to identify the tolerance limits of both plant extracts such as veterinarians prepared by traditional healers. According the obtained LD_{50} values, the two extracts are very low toxicity. Comparatively, the *D. oliveri* stem bark extract is less toxic than *A. leiocarpus* leaves extract. The various chemical groups contained in both two plants justified their use by traditional healers in animal diseases.

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REFERENCES

[1] M. Sangaré. Synthèse des résultats acquis sur l'élevage des petits ruminants dans les systèmes de productions animales d'Afrique de l'Ouest. Document UE-Procordel/Cirdes. Bobo-Dioulasso, Burkina Faso, **2005,** 163 pages.
[2] H. H. Tamboura, H. Kaboré, S. M. Yaméogo. *Biotechnol. Agron. Soc. Environ.* , **1998**, 2 (3): 181-191.
[3] Centre d'Analyse des Politiques Economiques et Sociales (CAPES). Etat des lieux des savoirs locaux au Burkina Faso. Inventaire des bonnes pratiques et propositions pour leur contribution au développement. Ouagadougou, **2006**; 448 pages.
[4] A. Kaboré, H. H. Tamboura, A. M. G. Belem, A. Traoré. *Int. J. Biol. Ch. Sci.,* **2007**, 1(3): 297-304.
[5] J. B. Githiori, Hôglund J., P. J. Walker. *Animal Health Research Reviews*, **2007**, 6(1):91-103.
[6] I. Ciulei. Methodology for analysis of vegetable drugs. Bucharest: Ministry of Chemical Industry, Bucharest, **1982**; 67 pages.
[7] C. J. T. Liechtfield, F. Wilcoxon. *J. Pharmacol. Exp. Ther.*, **1949**, 57:261-113.
[8] H. C. Hodge, J. H. Sterner. *American industrial Hygien Association*, **1943**, 10:93.
[9] M. S. Barry, K. Oularé, A. Diallo, Y. Camara, M. Camara, M. A. Baldé. *Revue Méd. Pharm. Afr.* , **2001**, 15: 95-102.
[10] A. A. Ahmadu, A. U. Zezi, A. H. Yaro. *Afr J Tradit Complement Altern Med.* **2007**, 4(4): 524–528.
[11] I. A. Jegede, F. C. Nwinyi, I. Muazzam, D. D. Akumka, A. A. Njan, M. Shok. *African Journal of Biotechnology*, **2006**, 5 (10): 930-935.
[12] N. Somé, L. Sawadogo, M. Lompo, J. L. Pousset, I. P. Guissou. *Rev. Sci. & Tech. Série: Sciences de la Santé*, **1995-1996**, 22(1): 38-46.
[13] B. M. Agaie, P. A. Onyeyili, B. Y. Muhammad, M. J. Ladan. *African Journal of Biotechnology*, **2007**, 6 (7): 886-889.
[14] O. G. Nacoulma/Ouédraogo. Thèse de Doctorat d'Etat ès Sciences Naturelles. Université de Ouagadougou,(Ouagadougou, **1996**).
[15] Désert Margins Program (DMP). Les savoirs traditionnels de préservation des terres et de la diversité biologique dans les sites de recherches du programme d'action sur les zones en marge du désert (DMP). Rapport 07/CNRST, Ouagadougou, **2004** ; 61 pages.
[16] P. Delaveau. *Aliments*, **1988**, 137-139.
[17] M. M. Cowan. *Clinical Microbiology Reviews*, **1999**, 12(4): 564-582.
[18] M. Arbonnier. Arbres, arbustes et lianes des zones sèches d'Afrique de l'Ouest. CIRAD/MNHN/UICN. **2000** ; 542 pages.
[19] D. N Onwukaema et F.Udoh. *Nig. Journ. Of Nat. Prod. and Med.* , **199**9, 3: 39-41
[20] C. Vonthron-Sénécheau, B. Weniger, M. Ouattara, F. T. Bi, A. Kamenan, A. Lobstein, R. Brun, R. Anton. *Journal of Ethnopharmacology*, **2003**, 87 : 221–225.
[21] M. Baoua, J. Fayn , J. Bassiere. *Plantes Médicinales et Phytothérapie*, **1976**, 10: 251–266.
[22] A. Mann, J. O. Amupitan, A. O. Oyewale, J. I. Okogun and K. Ibrahim. *African Journal of Pharmacy and Pharmacology*, **2009**, Vol. 3(1). pp. 022-025