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Effects of *Chenopodium ambrosioides* meal as food additive in the diet, on feed intake, *in vivo* digestibility and caecal flora dynamysm of cavies

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ABSTRACT

Aim: The study was aimed to evaluate the effects of *Chenopodium ambrosioides* meal, ethanolic or aqueous extracts on feed intake digestibilty and caecal flora varation in guinea pigs.

Method and materials: *Chenopodium ambrosioides* meal, ethanolic or aqueous extracts were incorporated at a level of 0.5% into the compound feed. Pelleted ration was offered to the animals. Eighty cavies of local breed with an average weight of 431.67±52.63 g were used. The digestbility trial last for 17days (10 days of adaptation and 07 days of data collection), each ration was repeated on 14 guinea pigs (7 males and 7 females), for a total of 56 animal. For the evaluation of the caecal flora, 24 animals randomly selected, i.e. 6 per treatment, were sacrificed at the end of the adaptation period for the identification of the digestive microflora. Then at the end of the data collcetion period, 24 other animals were randomly selected from the 56 animals above and sacrificed for determination of evolution of the digestive microflora.

Results: The inclusion of *Chenopodium ambrosioides* meal, ethanolic or aqueous extract significantly ($p<0.05$) lowered the feed intake (23.83 g; 23.42 g; 21.58 g) of cavies compared to the control ration (26.58 g). Feed intake of males (28.97 g) on the control diet was significantly ($p<0.05$) higher than that of females (24.20 g). However, animals fed on ration containing 0.5%*Chenopodium ambrosioides* meal, ethanolic or aqueous extracts, feed intake was higher in females. Regardless of sex, DM digestibility of ration containing 0.5% *Chenopodium ambrosioides* meal (48.27%) was comparable to that of the control (41.81%) but, significantly higher than that of ethanolic extract (T2) (34.42%) and aqueous (T3) (37.79%) ration. Meanwhile, concerning OM, CP and CC digestibilty showed no significant difference ($P>0.05$) between treatments.

Conclusion: It was concluded that incorporation of the *Chenopodium ambrosioides* meal and the various extracts significantiy lowers the ingestion. On the other hand improves feed conversion and has a positive effect on the intestinal microbiota.

Keywords: Cavies, *Chenopodium ambrosioides*, *Chenopodium ambrosioides* meal, extracts feed, additive.

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Introduction

Food security and protein safely is a real challenge in most African countres, especially in Cameroon (Miegoue *et al*., 2018). Indeed, population growth creates an imbalance between demand and supply of animal proteins, leading to malnutrition especially in low-income families (Noumbissi *et al*., 2014). Development of mini-breeding appears to be a better alternative for the fight against protein malnutrition, poverty and food insecurity in Africa in general and Cameroon in particular (Miegoue *et al*., 2018). Among the different species in this category, the cavies presents itself as one of the opportunities to be seized in order to help poor households escape from the situation of food insecurity and poverty to which they are subjected (Faihum et *al*., 2020). Since it does not require large initial investments, guinea pig farming has the characteristics of an economically profitable mini-farm that can effectively contribute to food security (Niba et *al*., 2012). Guinea pig is a monogastric animal with a strictly herbivorous diet that makes better use of local fodder resources that have no market value (Mounchili et *al*., 2019), and provides good-quality meat, making it a genuine meat animal (Kouakou et *al*., 2017). Thus, previous works have reported that feeding remains a real constraint for its

production due to digestive disorders inherent to the imbalance of the caecal flora frequent in this species (Tobou et *al.,* 2020). Indeed, the digestive use of food in guinea pigs is highly dependent on the caecal microbial flora whose balance is easily altered by a sudden change in diet. The use of antibiotic in this animal is not elficient and, antibiotic as growth promoter is not accepted in animal breeding. To overcome this problem of digestive disorders in guinea pig production, the use of phytobiotics, including herbs, spices and essential oils, has increased in recent years (Brenes and Roura, 2010). Many plants used in traditional medicine contain bioactive substances with beneficial health effects, including immunostimulant effects (Akram et *al*., 2014). In particular, plants from the Chenopodiaceae family have a high content of polyphenols known for their beneficial role in animal and human health (Nowak et *al*., 2016). Among these plants is *Chenopodium ambrosioides* which is an annual or perennial plant 40-100 cm high. It is one of the most widely used plants in folk medicine in Latin America, for its potent anthelmintic activity against roundworm and tapeworm as well as to treat parasitic infections in livestock (Keddad, 2018). According to Daoudi et *al* (2017), phytochemical analysisof the plant revealed the presence of many chemical constituents including alkaloids, quinones, tannins and terpenes as well as the absence of anthocyanins and polyphenolic compounds. Studies conducted by Houngnimassoum *et al* (2020) on traditionally reared chickens showed that *Chenopodium ambrosioides* has a nematodicidal effect on *Ascaridia galli.* In addition, Garcia *et al* (2020) showed the microbiological and oxidative properties of *Chenopodium ambrosioides* in rabbits. Furthermore, the works of Kouam *et al.* (2015) showed that essential oil soaps of *Chenopodium ambrosioides* leaves were to*in vitro* and *in vivo*toxic on*Ripicephalus lunatus*; and that tick mortality rates increased progressively in terms of dose and time. The present work was initiated with the objective of evaluating the effects of *Chenopodium ambrosioides* meal, ethanolic or aqueous extracts on feed intake digestibilty and caecal flora varation in guinea pigs.

Materials and Methods

Experimental site

The study was conducted at the univesity of Dschang at the Application and experimental Farm (AEF). This locality is located in the western high lands of cameroon at the altitude of 1,410M, longitude East 10° 26′ and latitude 5° 26′.

Plant material

The plant material consisted of chenopod (*Chenopodium ambrosioides*) collected from a field in the city of Dschang. It was washed with water, dried at room temperature (25-30°C) during the day and ground with a harmed mill machine. It was then preserved in hermetically sealed boxes. Part of this *Chenopodium ambrosioides* meal, each of these form of the plant product was include a 0.5% in the compound feed as feed additive.

Extraction process

Fresh leaves of *Chenopodium ambrosioides* were harvested, dried and ground. The powder obtained was used for aqueous and ethanolic extraction according to the process in (Fig. 1)

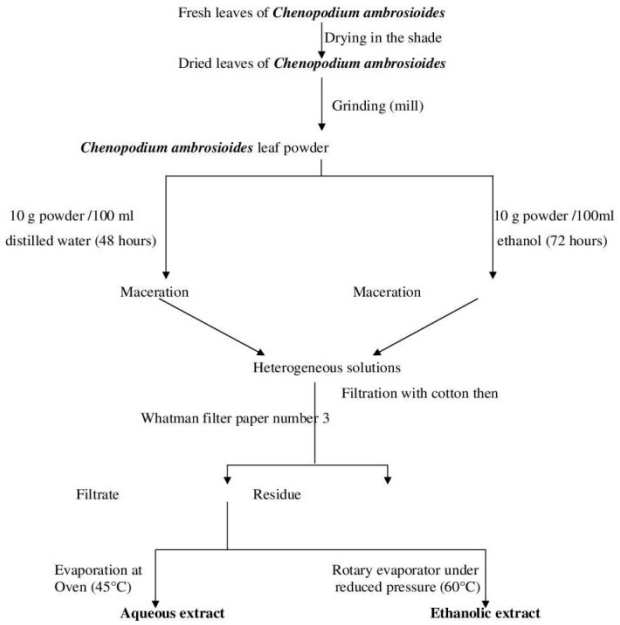

Fig. 1: *Chenopodium ambrosioides* leaf extraction process

Animal materials and housing

For thistrial, 80 cavies of local breed (40 males and 40 females) of the species *Cavia porcellus*, all born at the University of Dschang apply and experimenalFarm were used in this study. They were approximately 5 months old with a mean weight of 431.67±52.63 g. The animals were housed in individual wire mesh cages each measuring 80 cm x 50 cm x 30 cm with a plastic feeder of 100 g capacity and a plastic drinker.

Experimental rations

The proportions of the different ingredients used in the experimental feed or ration and its chemical composition were presented (Table 1).

Table 1: Formula and chemical composition of animal feed

Ingredients	$T_{0\%}$
Maize	22
Trypsacum laxum	25
Soybean meal	04
Cottonseed cake	03
Palm kernel cake	09
Fish meal	09
Bone meal	01
Wheat bran	22
10% concentrate	02
Shell	01
Molasses	02
Total	100

Chemical composition of the ration $T_{0\%}$	
Crude protein (% DM)	17.82
Crude Cellulose (% of DM)	16.60
Fat (% DM)	1.47
Gross Energy (Kcal/Kg DM)	3993.55
Digestible Energy (Kcal/ Kg DM)	2803.05

**VitA: 3000000UI, Vit D: 600000UI, VitE: 4000mg, VitK: 500mg, Vit B1: 200mg, Vit B2: 1000mg, Vit B6 :400mg, Vit B12: 4mg, Iron: 8000mg, Cuvre: 2000mg, Zinc: 10000mg, Selenium: 20mg, Manganese: 14000mg, Methionine: 200000mg, Lysine: 78000mg DM: Dry matter.*

The daily rations served to each animal were made up as follows:

- T0: pelleted compound feed containing 0% *Chenopodium ambrosioides* (Batch 0);
- T1: pelleted compound feed containing 0.5% *Chenopodium ambrosioides* meal (batch 1);
- T2: pelleted compound feed containing 0.5% aqueous extract of *Chenopodium ambrosioides* (batch 2);
- T3: pelleted compound feed containing 0,5 % ethanoic extract of *Chenopodium ambrosioides* (batch 3).

Evaluation of intake, in vivo digestibility of rations and in caecal flora dynamism

Animals were randomly allocated to individual cages following a completly randomised design. For feed intake assessment, feed quantities were recorded, and refusals were collected daily and weighed before any further distribution. Refusals were quantified to determine the amount of feed ingested. Feed intake or food consumption was calculated according to the below formula:

Feed intake **=** Daily amount of feed served **-** Amount not consumed (refusals).

The digestibility test was preceded by a 10-day adaptation period of the animals to the digestibility cage and the pelleted compound feed. During this period, the quantities of the pelleted compound feed served were adjusted to the animal's estimated consumption of 60 g/animal/day. During the data collection period, which lasted 7 days, each morning before the distribution of the feed, faeces were collected, weighed and dried at 60°C in a ventilated oven in the laboratory. Subsequently, the analysis of their dry matter (DM), organic matter (OM), crude protein (CP) and crude cellulose (CC) content was done according to the method described by AOAC (2000) cited by Mouchili (2019). The apparent digestive utilization coefficients of Dry Matter (DM), Organic Matter (OM), Crude Protein (CP), and Crude Cellulose (CC) were calculated according to the formula of Roberge and Toutain (1999):

- aDC DM= 100 x [Ingested DM (g)Excreted DM (g)] / [Ingested DM (g)]
- aDC OM= 100 x [Ingested OM (g) Excreted OM (g)] / [Ingested OM (g)]
- aDC CP= 100 x [Ingested CP (g) Excreted CP (g)] / [Ingested CP (g)]
- aDC CC= 100 x [Ingested CC (g) Excreted CC (g)] / [Ingested CC (g)]

For the caecal flora parameter, 24 randomly selected animals, i.e. 6 per treatment, were sacrificed at the end of the adaptation period for the identification of the digestive microflora. Subsequently, at the end of the digestibility test itself, 24 animals were again selected and sacrificed for the determination of the evolution of the digestive microflora. During each sacrifice, the caecum of the animal was cut open and the contents were collected. The swabs were used to collect the faecal material and stored aseptically in sterile boxes in the refrigerator at -20°C, in the FASA Laboratory of Physiology and Animal Health where the identification and quantification of*lactobalillus*, *Escherichia coli*, coliforms and salmonella were done in the following respective and specific culture media; MRS AGAR, Mac Conkey AGAR, Endo AGAR and SS AGAR following the method described by Benson et *al*., (2002).

Statistical analysis

In *vivo* feedintake and *iv vivo* digestibility data were submitted to a 2-factor analysis of variance (ration and sex) and caecal flora data were subjected to a 1-factor analysis of variance (ration), using SPSS 20.0. To compare treatments means at a 5% threshold level, Dunca's multiple range test was used (Steel and torrie 1984) ; probality values less than 0.05 were considered as significant.

Results and Discussion

The extrat yeild and secondary metabolite was presented (Table 2). Some compounds were present in the *Chenopodium ambrosioides* meal and absent in the extract (alkaloids, tauins, saponins onther extrat cqanins and anthraquinones) On the other hand, flavonoids were present in the extract and absent in the *Chenopodium ambrosioides*meal aqueous extract was higher than ethanolic extract.

Table 2: Extraction yeild and some secondary metabolits of *Chenopodium ambrosioides* meal or extract

	Chenopodium ambrosioides meal	Ethanoic extract	Aqueous extract
Yield (%)		7.5	15.90
Alkaloids	+	-	-
Phenols	+	+	+
Flavonoids	+	-	-
Sterols	-	+	+
Triterpenoids	+	+	+
Tannins	+	-	-
Saponins	+	-	-
Anthocyanins	+	-	-
Anthraquinones	+	-	-

+ = present; - = absent; Rd= Extraction efficiency (%)

The effects of *Chenopodium ambrosioides* meal, ethanolic or aqueous extracts on feed intake of compound feed, dry matter (DM), organic matter (OM), crude protein (CP) and crude fiber (CF) show that, in males, the control ration resulted in significantly ($p < 0.05$) higher feed intake than the other rations. Meanwhile this time, the different feed intake of the animals submitted to treatment T2 were significantly ($p < 0.05$) higher than those of the animals fed on treatment T1. The lowest values ($p < 0.05$) were recorded in the animals fed on treatment T3. On the other hand, in females, these different feed intakes were significantly ($p < 0.05$) higher in animals submitted to treatment T1. The feedintakes of the animals fed on treatment T0 were significantly higher ($p < 0.05$) than those of the animals subjected to treatment T2. The lowest values were recorded in the animals fed on treatment T3. Irrespective of sex, the intake of animals on the T0 ration was significantly ($p < 0.05$) higher. However, the T1 and T2 rations gave comparable intakes ($p > 0.05$) but significantly ($p < 0.05$) higher than that obtained with the T3 ration.

Concerning the comparison between males and females, the highest intake values of DM (27.09 gMS/d), OM (23.79 gMS/d), CP(3.92 gMS/d) and CF (3.09 gMS/d) were observed with males the Control batch. On the other hand, the highest intake values of DM (23.75 gMS/d), OM (20.85 gMS/d), CP (3.44 gMS/d) and CF (2.71 gMS/d) in females were observed with the T1 ration. In addition, the intake of males in the control batch was significantly higher ($p < 0.05$) than that of females; however, the opposite trend was observed with animals in the other treatments. The effects of the inclusion of *Chenopodium ambrosioides*meal, ethanoic or aqueous extracts of *Cheopodium ambrosioides* on ADUC.DM showed that in males, no significant difference ($p > 0.05$) was observed. However, in females, the ration containing the inclusion of *Chenopodium ambrosioides*meal resulted in a significantly ($p < 0.05$) higher ADUC.DM than the other rations. On the other hand, the control ration and the ration containing the inclusion of aqueous extract gave statistically comparable ADUC.DM ($p > 0.05$) but remained higher than the ration containing the inclusion of ethanoic extract. Regardless of sex, the ADUC.DM of treatment T1 was significantly ($p < 0.05$) higher than that observed with treatments T2 and T3. On the other hand, the ADUC.DM obtained with treatment T0 remained comparable ($p > 0.05$) to those of rations T2 and T3 on the one hand and to that of ration T1 on the other hand.

In females, the ration containing the inclusion of *Chenopodium ambrosioides* powder resulted in a significantly higher ADUC. OM than that of the control ration and the ration containing the inclusion of the ethanoic extract of *Chenopodium ambrosioides*, but comparable to that obtained from the ration containing the inclusion of the aqueous extract of *Chenopodium ambrosioides*. On the other hand, the latter was higher than that of the T2 ration and remained statistically comparable to that of the T0 ration. The lowest value was recorded in animals fed ration T2 but remained comparable to ration T0. On the other hand, in males and independently of sex, the inclusion of the *Chenopodium ambrosioides*meal and the different extracts had no significant effect.

In males, the significantly ($p < 0.05$) highest ADUC.CF value was obtained in animals fed the ration containing added ethanoic extract, and the lowest in those fed the ration containing added powder. However, no significant difference ($p > 0.05$) was observed between the ADUC.CF value of animals fed on the T1 ration and that of animals fed on the T3 ration. The results obtained with the animals on ration T0 were also statistically comparable ($p > 0.05$) to those fed ration T2 on the

one hand and those fed ration T1 and T3 on the other. In females, the statistically highest ADUC.CF value was observed in animals fed ration T1 and the lowest in those fed ration T2. However, the value of the animals fed on T0 remained comparable to that of the animals fed on T1; while that of the animals fed on T2 remained comparable to that of the animals fed on T3. Regardless of sex, no significant difference ($p > 0.05$) was observed between the different treatments. Regarding ADUC.CP no significant difference ($p > 0.05$) was observed between the different treatments.

In males the best values of ADUC.DM (51.79%) and ADUC.CP (62.19%) were obtained in the control animals; while the best values of ADUC.OM (54.15%) and ADUC.CF (58.18%) were obtained in the T1 and T2 animals respectively. However, in females, the best values of ADUC. DM (48.81%), ADUC. OM (50.10%) and ADUC.CF (56.48%) were observed in animals of batch T1 while the highest values of ADUC.CP (56.48%) were observed in animals of batch T2.

Concerning the comparison between males and females, the (ADUC) of the different nutrients in males was significantly ($p < 0.05$) higher than that of females in animals subjected to treatments T0, T2 and T3 with the exception of ADUC.CF for the T0 ration and ADUC.CPfor T3 where the values were comparable ($p > 0.05$) between males and females. However, no significant difference ($p > 0.05$) was observed between males and females in animals subjected to T1 treatment.

Table 3. Feed intake of cavies fed on ration containing powder, ethanolic or aqueous extracts of *Chenopodium ambrosioides*.

Feed intake (g DM/d/animal)		**Treatments**				***SME***	**p**
		T0	T1	T2	T3		
Compound feed (DM)	♂(7)	28.97^{aA}	22.28^{cB}	23.33^{bA}	20.56^{dB}	0.72	0.00
	♀(7)	24.20^{bB}	25.39^{aA}	23.51^{cA}	22.59^{dA}	0.24	0.00
	♂♀(14)	26.58^{a}	23.83^{b}	23.42^{b}	21.58^{c}	0.37	0.00
SEM		0.81	0.52	0.08	0.34		
P		0.00	0.00	0.36	0.00		
Nutrients							
Dry matter (DM)	♂(7)	27.09^{aA}	20.84^{cB}	21.82^{bA}	19.23^{dB}	0,67	0.00
	♀(7)	22.63^{bB}	23.75^{aA}	21.98^{cA}	21.13^{dA}	0,23	0.00
	♂♀(14)	24.86^{a}	22.29^{b}	21.90^{b}	20.18^{c}	0,35	0.00
SEM		0.75	0.49	0.08	0.32		
P		0.00	0.00	0.36	0.00		
Organic matter(OM)	♂(7)	23.79^{aA}	18.30^{cB}	19.16^{bA}	16.89^{dB}	0,59	0.00
	♀(7)	19.87^{bB}	20.85^{aA}	19.31^{cA}	18.55^{dA}	0,20	0.00
	♂♀(14)	21.83^{a}	19.58^{b}	19.23^{b}	17.72^{c}	0,31	0.00
SEM		0.66	0.43	0.07	0.28		
P		0.00	0.00	0.36	0.00		
Crude protein(CP)	♂(7)	3.92^{aA}	3.01^{cB}	3.16^{bA}	2.78^{dB}	0,09	0,00
	♀(7)	3.27^{bB}	$3,44^{aA}$	3.18^{cA}	3.06^{dA}	0,03	0,00
	♂♀(14)	3.60^{a}	3.23^{b}	3.17^{b}	2.92^{c}	0.05	0.00
SEM		0.11	0.07	0.01	0.04		
P		0.00	0.00	0.36	0.00		
Crude fiber(CF)	♂(7)	3.09^{aA}	2.37^{cB}	2.49^{bA}	2.19^{dB}	0.07	0.00
	♀(7)	2.58^{bB}	2.71^{aA}	2.51^{cA}	2.41^{dA}	0.02	0.00
	♂♀(14)	2.83^{a}	2.54^{b}	2.50^{b}	2.30^{c}	0.04	0.00
SEM		0.08	0.05	0.00	0.03		
P		0.00	0.00	0.36	0.00		

a, b and c: means with the same lowercase superscript letters in the same row are statistically identical; A, B: means with the same uppercase superscript letters in the same column are statistically identical; SEM: Standard Error on the Mean; P : Probability; ( ): number; ♂:male; ♀:female; ♂♀: male and female combined, T0: Control; T1: Control + 0.5% Chenopodium meal; T2: Control + 0.5% Chenopodium ethanoic extract; T3: Control + 0.5% Chenopodium aqueous extract.

Table 4: Apparent digestive utilization coefficients (ADUC) of nutrients in cavies fed on ration containing *Chenopodium ambrosioides* meal, ethanolic or aqueous extracts.

ADUC (%)		Treatments				*SME*	P
		T0	T1	T2	T3		
ADUC.DM	♂(7)	51.79^{aA}	47.72^{aA}	44.44^{aA}	41.13^{aA}	1.55	0.07
	♀(7)	31.83^{bB}	48.81^{aA}	24.40^{cB}	34.46^{bB}	2.14	0.00
	♂♀(14)	41.81^{ab}	48.27^{a}	34.42^{b}	37.79^{b}	1.59	0.01
SEM		3.75	0.98	3.81	1.63		
P		0.00	0.62	0.00	0.04		
ADUC.OM	♂(7)	53.86^{aA}	54.15^{aA}	49.92^{aA}	52.21^{aA}	1.66	0.82
	♀(7)	40.41^{cbB}	50.10^{aA}	35.60^{cB}	46.80^{abB}	1.88	0.01
	♂♀(14)	47.14^{a}	52.12^{a}	42.76^{a}	49.50^{a}	1.44	0.12
SEM		3.28	2.45	3.53	1.32		
P		0.03	0.44	0.04	0.04		
ADUC.CP	♂(7)	62.19^{aA}	54.00^{aA}	53.10^{aA}	56.34^{aA}	1.68	0.22
	♀(7)	49.89^{aA}	54.48^{aA}	56.81^{aA}	51,59^{aA}	1.99	0.65
	♂♀(14)	56.04^{a}	54.24^{a}	54.95^{a}	53.97^{a}	1.31	0.95
SEM		3.31	2.75	2.99	1.44		
P		0.06	0.93	0.56	0.10		
ADUC. CP	♂(7)	53.09^{abA}	46.74^{bA}	58.18^{aA}	50.31^{bA}	1.46	0.02
	♀(7)	52.26^{abA}	56.48^{aA}	35.46^{cB}	42.26^{bcB}	2.49	0.00
	♂♀(14)	52.67^{a}	51.61^{a}	46.82^{a}	46.28^{a}	1.49	0.31
ESM		2.54	2.67	4.22	1.88		
P		0.88	0.06	0.00	0.02		

a, b and c: means with the same lowercase superscript letters in the same row are statistically identical; A, B: means with the same uppercase superscript letters in the same column are statistically identical; SEM: Standard Error on the Mean; P : Probability; ( ): number; ♂:male; ♀:female; ♂♀: male and female combined, T0: Control; T1: Control + 0.5% *Chenopodium* meal; T2: Control + 0.5% *Chenopodium* ethanoic extract; T3: Control + 0.5% *Chenopodium* water extract.

Table 5: Effect of *Chenopodium ambrosioides* powder, ethanoic and aqueous extracts on the variation of caecal flora in guinea pigs.

Collection days	Caecal flora Log_{10} (CFU/µl)	Treatments				SME	P
		T0	T1	T2	T3		
J_{10}	Coliforms	2.77^{c}	3.05^{bc}	3.61^{a}	3.33^{ab}	0.11	0.02
	E. coli	2.00^{b}	2.90^{a}	2.85^{a}	2.93^{a}	0.13	0.00
	Salmonella	1 .72^{a}	3.17^{a}	2.74^{a}	3.40^{a}	1.05	0.22
	Lactobacilli	3.71^{c}	4.65^{b}	5.21^{a}	4.73^{b}	0.16	0.00
J_{17}	Coliforms	3.59^{a}	3.71^{a}	3.62^{a}	3.39^{a}	0.05	0.91
	E. coli	1.79^{a}	1.23^{a}	2.15^{a}	2.77^{a}	0.43	0.71
	Salmonella	2.80^{a}	0.86^{a}	1.66^{a}	1.46^{a}	0.38	0.40
	Lactobacilli	4.54^{a}	3.64^{ab}	3.46^{b}	4.07^{ab}	0.17	0.07

a, b and c: Means with the same letters on the same line are not significantly different at the 5% level; SM: Standard Error on the Mean; P: Probability; ( ): number; ♂ :male; ♀ :female; ♂♀ : male and female combined, T0 : Control; T1 : Control + 0.5% *Chenopodium* powder; T2 : Control + 0.5% *Chenopodium* ethanoic extract; T3 : Control + 0.5% *Chenopodium* water extract.

From this table it can be seen that at D10 the highest value of coliforms in the cecum was significantly ($p < 0.05$) observed in animals subjected to treatment T2 and the lowest in animals subjected to control treatment. However, the value of animals subjected to treatment T3 remained statistically comparable to that of animals subjected to treatment T1 and T2 while that of animals subjected to treatment T1 remained statistically comparable to that of animals subjected to treatment T0 and T3. *E. coli* levels in the cecum of animals fed T1, T2 and T3 were comparable ($p > 0.05$) and significantly ($p<0.05$) higher than in animals fed T0. No significant difference ($p > 0.05$) was observed between the different treatments regarding Salmonella levels in the cecum. The levels of Lactobacilli in the cecum of animals fed T1 and T3 were comparable ($p> 0.05$) but significantly ($p<0.05$) lower than in animals fed T2 and higher

than in animals fed T0.

Furthermore, at D_{17}, no significant difference ($p > 0.05$) was observed between the different treatments, regardless of the bacterial group.

Comparison ofdiferent bacteria group according to differents treatment in cavies.

It showed that lactobacilli levels were significantly ($p<0.05$) higher than coliform, *E.coli* and salmonella levels in animals subjected to treatments T1, T2 and T3. In contrast, no significant difference ($p>0.05$) was observed between the different bacterial groups in animals fed on treatment T0.

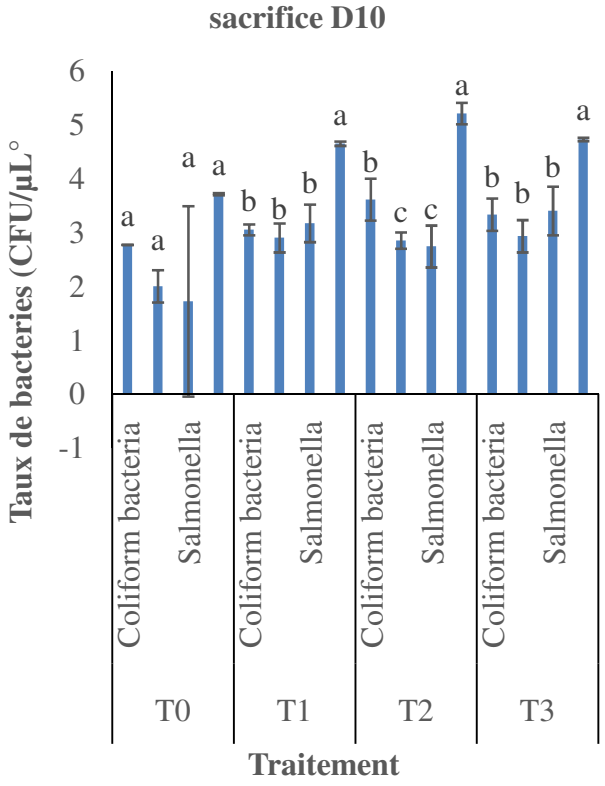

Fig. 2: Comparison of the different bacteria per treatment in cavies

a, b and c: means with the same letters for the same ration are not significantly different at the 5% threshold. T0: Control; T1: Control + 0.5% Chenopodium powder; T2: Control + 0.5% Chenopodium ethanoic extract; T3: Control + 0.5% Chenopodium aqueous extract.

The figure shows that lactobacilli levels were significantly ($p<0.05$) higher than coliform, *E.coli* and salmonella levels in animals subjected to treatments T1, T2 and T3. In contrast, no significant difference ($p>0.05$) was observed between the different bacterial groups in animals fed on treatment T0.

Comparison of different bacteria group in cavies according to different treatment at the end of in vivo digesbility trial (Day 17)

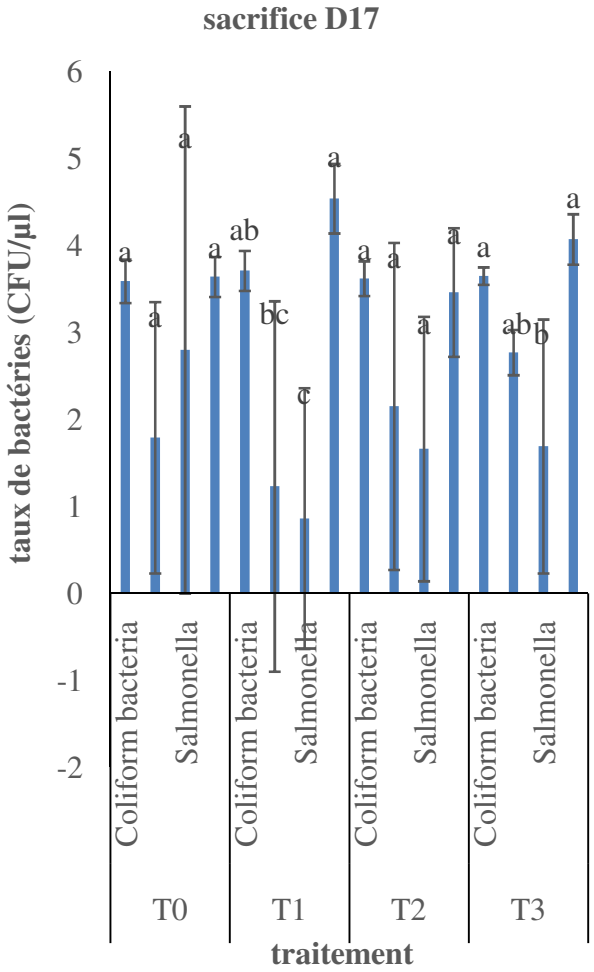

Fig. 3. Comparison of different bacteria per treatment in cavies at the end of second trial (17 days).

a, b, c: means with the same letters on the same line are not significantly different at the 5% threshold. T0: Control; T1: Control + 0.5% *Chenopodium* powder; T2: Control + 0.5% *Chenopodium* ethanoic extract; T3: Control + 0.5% *Chenopodium* aqueous extract.

From this figure it can be seen that the lactobacillus levels of animals fed on treatment T1 were significantly ($p<0.05$) higher than the levels of coliforms, *E.coli* and salmonella. On the other hand, lactobacillus levels in animals fed on T3 treatment remained comparable to coliform levels but were higher than Salmonella and *E. coli* levels. For the animals fed on treatments T0 and T2, no significant difference ($p>0.05$) was observed between the different bacterial groups.

Comparison effect of treatment on bacteria groups according to the number of sacrifice in cavies

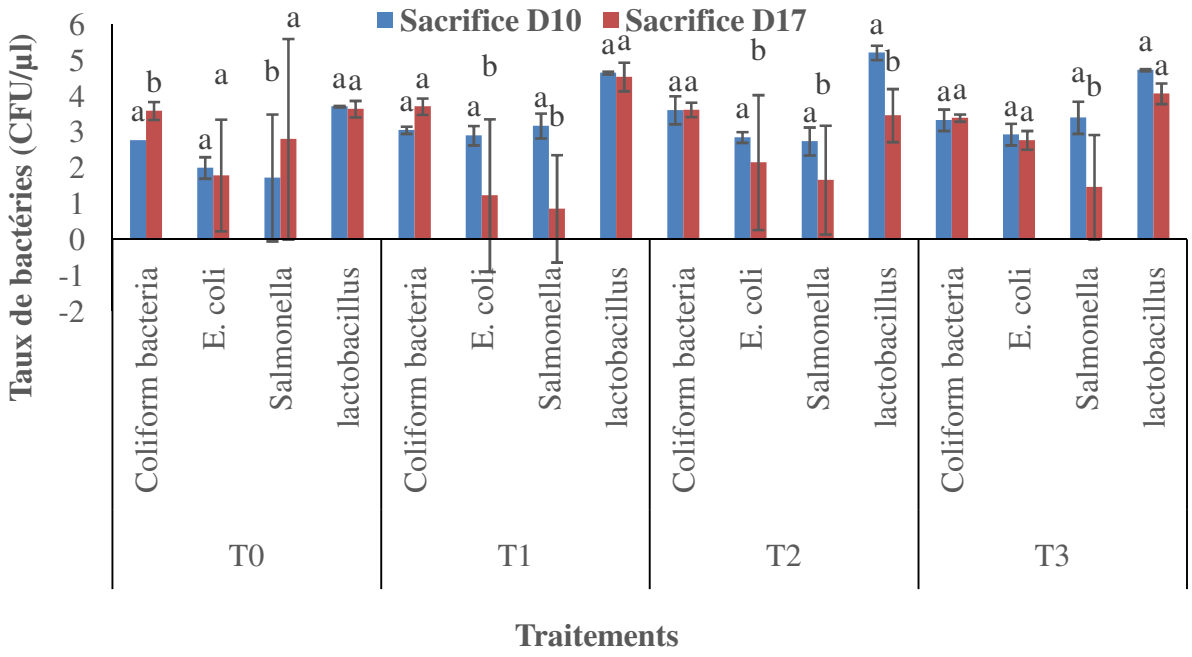

Fig. 4. Comparative effect of treatments on bacteria group according to the number of sacrifices
a: Means bearing the same letters for the same parameter are not significantly different at the 5% threshold for the same treatment; T0: Control; T1: Control + 0.5% Chenopodium powder; T2: Control + 0.5% Chenopodium ethanoic extract; T3: Control + 0.5% Chenopodium aqueous extract.

Salmonella levels were significantly ($p<0.05$) lower between day 10 and day 17 sacrifice regardless of the treatment considered; the same observation was donefor *E. coli levels with* the exception of T3 animals which remained comparable. The coliform leve remained comparable between the sacrifice on day 10 and day 17 in the batches receiving treatments T1, T2 and T3, but in the animals submitted to treatment T0, this rate was significantly higher ($p<0.05$) at 2sacrifice. Regarding the lactobacillus level, no significant difference was observed between the different days of sacrifice in animals submitted to treatments T0, T1 and T3, but in those subjected to treatment T2, this level was higher on day 10 sacrifice compared to day 17 sacrifice. At al the leve of lactobacillus remain higher during treatmeant.

In general, the inclusion of the powder and the different types of extracts in the feed decreased the feed intake of the animals compared to the control batch. This decrease could be explained by the fact that *Chenopodium ambrosioides* being a feed additive has bioactive molecules such as ascaridole and caryophyllene oxide which give it a bitter taste and a cytotoxic activity contributing to the inhibition of the respiratory chain in mammalian cells and mitochondria (Gille et *al*., 2010), hence the reduction of feed intake. These results corroborate the work done by other authors on phytobiotics of the same family as *Chenopodium;* notably those conducted by Barazesh et *al*. (2013) and Tariq (2017), who noted that the increasing addition of ginger powder to the ration, decreased feed intake in chickens. However, these observations are in contrast to those of Nouboussi et *al*. (2021) who noted that supplementation of bean fan biochar as a feed additive at 0.8% to guinea pigs significantly improved feed intake.

The incorporation of the *Chenopodium ambrosioides* meal in the diet increased numerically the digestibility of all nutrients in general. This suggests that varying the inclusion leve may produce more noticeable effects. In addition, for organic matter, inclusion of aqueous extracts resulted in significantly higher digestive utilisation in female guinea pigs. This would be related to the fact that *Chenopodium* due to its pharmacological, laxative and vermifuge properties seems to

facilitate digestion. These observations are in agreement with the work of Florian et *al*, (2013); Tatsinkou et *al*, (2020); Nouboussi et *al*, (2021) who respectively noted an improvement in dry matter digestibility without degradation of nitrogen assimilation following the ingestion of aqueous extract of *C. ambrosioides* leaves, aqueous and hydroethanol extracts of avocado kernel or biochar based on bean fans in the ration.

Different bacteria group level (coliforms, *E. coli*, *salmonella and lactobacilli*) were significantly affected by the meal, aqueous and ethanolic extracts of *C. ambrosioides* between the sacrifice on day 10 and day 15. Indeed, the highest level of lactobacilli was obtained with the ration containing *Chenopodium ambrosioides* meal. However, the level of pathogenic bacteria in the gut of the guinea pigs (*E. coli* and *salmonella*) decreased significantly between day 10 and day 15 in favour of beneficial bacteria *(lactobacilli*). This can be explained by the fact that fermentation leads to the production of large quantities of lactic and acetic acids in the ileum and colon, which have an indirect effect on the concentration of propionic and butyric acids. Their presence reduces intestinal pH, thereby inhibiting pathogenic bacterial proliferation (Broderick and Duong, 2016; Wang et *al.*,2016). In particular, Chenopodiaceae plants have a high content of polyphenols known for their beneficial role in animal and human health (Nowak et *al*., 2016). This result is in agreement with the studies done by Fayez althobaiti (2020) in the use of RAMP-PCR assay showed that methanol extract of the leaves allowed the multiplication of Gram positive bacteria (*Bacillus subtilis* and *Staphylococcus aureus*) compared to Gram negative bacteria (*Escherichia coli* and *Proteus vulgaris*) and fungi (*Crytococcus neoformas* and *Candida albicans*). In the same sense, Maldonado-Garcia et *al*, (2019) recorded identical results but in fish feed. These results could also be explained by the fact that, *Chenopodium ambrosioides* is rich in natural antioxidants that can inhibit the proliferation of gram negative bacteria. In addition, Nouboussi et *al* (2021) showed that supplementation of bean tops with biochar as a feed additive at 0.8% in the diet of guinea pigs has a positive effect on the gut microbiota by increasing the rate of lactobacilli and clostridia and decreasing the rate of enterobacteria. In contrast, the rate of inclusion of *Arachis glabrata* in the guinea pig diet had no significant difference between the lactobacilli and enterobacteria levels of the caecal flora (Miégoué et *al*., 2019).

Conclusion

The incorporation of 0.5% of the *Chenopodium ambrosioides* meal, ethanolic and aqueous extracts of *Chenopodium ambrosioides* in the rations on the ingestion, digestibility and variation of the caecal flora in guinea pigs (*Cavia porcellus*), it appears that :

- The different forms of incorporation led to adecrease in feed intake;
- The incorporation of the powder induced an improvement in the digestibility of nutrients in these animals;
- The different forms of incorporation stimulated the multiplication of beneficial bacteria (*lactobacilli*) by inhibiting the growth of pathogens (*salmonella*, *E. coli* and coliforms) present in the intestinal microbiota;
- The best results were obtained with 0.5% inclusion of *Chenopodium ambrosioides* meal.

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