Infusion of *Chenopodium ambrosioides* consumed by rabbits: effects on carcass, meat and burger quality

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**Abstract**

Plants with high levels of antioxidant compounds have been used to feed animals and increase stability of their meat and meat products. The objective of this study was to evaluate the effects of *Chenopodium ambrosioides* infusion on carcass, meat and burger quality. Rabbits were assigned to each treatment (0, 5 or 10 g.L⁻¹ of *Chenopodium ambrosioides*). Animals were slaughtered after 28 days of fattening, after which carcass and meat quality was measured, and the meat obtained was processed into burgers. Results indicate that all variables measured were not significant, except for dissectible fat, pH, adhesiveness, L* value, and initial total plate counts in burgers. It is concluded that *Chenopodium ambrosioides* infusion could be used to feed rabbits, acquire meat, and use the meat for processing burgers, which have low bacterial growth and low oxidation development.

**Keywords:** antioxidant properties; *Chenopodium ambrosioides*; meat and carcass quality; microbiological quality.

**Practical Application:** If plant infusion is used to feed rabbits, additives to improve meat quality are not required.

1 Introduction

Rabbit meat has been considered as a functional food due to its nutritional properties (Dalle-Zotte & Szendrő, 2011). However, lipid oxidation has a negative impact on flavor, color, texture and the nutritional value of meat and meat products (Trebušak et al., 2014; Shah et al., 2014). Consequently, meat processing uses ingredients to provide suitable sensorial and functional characteristics, such as antioxidants (Sebranek, 2009). However, some of these additives have been under scrutiny due to their toxicity (Movileanu et al., 2013). Nevertheless, the use of antioxidants as additives in animal feed has improved the shelf life of meat products, mainly decreasing lipid oxidation (Qwele et al., 2013), and natural sources of antioxidants should be used to offer healthier meat products (Kumar et al., 2015). Bioactive compounds produced by plants have antibacterial or antioxidant properties (Hashemi & Davoodi, 2011), and it is for this reason that several plants, either in their complete form or as extracts, have been used to feed rabbits (Dalle-Zotte et al., 2016). The use of natural resources is a good alternative for feeding animals and producing meat products (Kumar et al., 2015). Aqueous, ethanolic or methanolic extracts obtained from grapefruit, ginger, peppermint and other plants have been used on meat products to inhibit lipid oxidation (Shah et al., 2014). Aromatic plants have bioactive compounds with medicinal properties which can lead to an improvement in performance and increased meat quality (Kovitvadhí et al., 2016).

With regard to feeding rabbits, the main aim of using plants as additives is to improve productive performance, meat and carcass quality, such as using olives and mushrooms (Trebušak et al., 2014), spirulina and thyme (Dal Bosco et al., 2014), rosemary (Cardinalli et al., 2015) and oregano (Soutlos et al., 2009). However, Meineri et al. (2010) showed a prooxidant effect in rabbit meat with animals fed on chia seeds during the fattening period, which suggests that it is not only botanical sources that have beneficial properties.

*Chenopodium ambrosioides* is an aromatic plant used in traditional Mexican gastronomy, and has some bioactive compounds such as flavonoids, phenolic acids and terpenes (Villalobos-Delgado et al., 2017). Villalobos-Delgado et al. (2016) demonstrated that an infusion of *Chenopodium ambrosioides* decreased lipid oxidation in ground beef. The objective of this study was to evaluate *Chenopodium ambrosioides* infusion as a beverage for fattening rabbits and its effects on the microbiological and oxidative stability of the carcass, meat quality and burgers.

2 Materials and methods

2.1 Animals and diets

In this trial, growing rabbits were housed in an experimental rabbit station located in Tulancingo, Hidalgo, Mexico. All animals were subjected to the care and management approved by the institutional committee for this purpose (Protocol no. CICUA/002/18). Eighteen weaned rabbits (35 days of age, eight female and 10 male) were randomly assigned to three treatments (n=6 by treatment and n=3 by repetition). Rabbits were housed in cages (90 x 60 x 40 cm) adapted with manual feeders and automatic drinkers. The breeds of rabbits were hybrids of New Zealand, California and English Pot with an average weight of 1165.52 ± 124.06 g. Feed was pelletized using a SKJ120 model pellet machine (Shandong, China). Isoprotein (16% of crude protein) and isoenergetic (2.3 Mcal/kg of digestible energy)
experimental diets were used to feed animals and were designed according De Blas and Mateos (2010). This trial was achieved by using three treatments: control (T1), 5 g L⁻¹ (T2), 10 g L⁻¹ (T3) of Chenopodium ambrosioides infusion as indicated in Table 1. The infusion was prepared by adding 5 or 10 g of dry ground Chenopodium ambrosioides to one liter of water and filtered using a coffee maker (Hamilton Beach, Glen Allen, Virginia, USA) at 80 °C for 15 min.

2.2 Carcass quality

Rabbits were slaughtered at 63 days of age in the facilities of a meat laboratory. The rabbits were not subjected to fasting before slaughtering and were mechanically stunned and processed according to national legislation (NOM-033-SAG/ZOO-2014, México, 2014). Once the rabbits were slaughtered, they were dissected to obtain weight measurements of hot carcasses, liver, kidney, full and empty digestive system, full and empty bladder, and skin. Carcasses were then stored under refrigeration at 4 °C for 24 h. The dorsal length of the live animal and carcass length were determined by measuring from the atlas to the last ischia vertebra. Hip circumference of the live animal and carcass were measured using a measuring tape.

2.3 Carcass jointing

The carcases of the animals were sectioned after 24 h of refrigeration storage as indicated by Blasco et al. (1993). The head was cut at the level of the atlas, the forequarter was obtained by cutting the last rib, and the loin was obtained in the 6 and 7th lumbar vertebra cutting the abdominal wall transversally to the vertebral column to obtain the foreleg. Kidney and scapular fat were also dissected. All of these parts were weighed separately, while all fat, bone and meat were removed from the legs and also weighed.

2.4 Meat quality

Meat color was measured on the surface of the loin between the last rib and 6th lumbar vertebra at cooling temperature (8 °C). A portable colorimeter i-Lab SS60 (Microptix, Wilton, Maine, USA) was used to determine color values by means of CIEL*a*b* color space with a standard illuminant D65 and observer of 2° as indicated in American Meat Science Association (2012). L* indicates brightness, a* redness and the b* axis yellowness. The pH was determined using a pH meter for meat, Hanna model HI99163 (Hanna instruments, Cluj-Napoca, Romania). Water holding capacity (WHC) was carried out according to Honikel (1987) and was expressed as a percentage of water loss. Cooking losses were measured in loins, with samples put in a plastic bag and cooked at 80 °C until reaching 68 °C internal temperature using a check temp digital thermometer (Hanna instruments, Portugal). Cooked samples were cooled at room temperature, weighed and cooking losses were calculated by differences in weight before and after cooking and expressed as a percentage. Once samples were cooled a texture profile analysis (TPA) was carried out following the method used by Bourne (1978) by cutting cubes (1 cm each side) and then compressing to 50% perpendicular to the muscle fiber direction using 1 mm/s of crosshead speed. Hardness, adhesiveness, cohesiveness, springiness and resilience were calculated using Texture Pro CT software and a CT3 Texture Analyzer (Brookfield, Middleboro, MA, USA).

2.5 Processing burgers

Meat obtained from each treatment was ground in a meat grinder (Torrey, Monterrey, Mexico). For the preparation of the burgers, only salt was added (NaCl, 12 g.kg⁻¹). After mixing, 80 g of meat was weighted to form the burger. It was packed using plastic film and stored at 4 °C for 14 d. Microbiological, lipid oxidation, color and pH were determined every seven days during storage.

2.6 Microbiological analysis of burgers

Microbiological analysis was carried out on day 0, 7 and 14 after the burgers were prepared. One g of meat burger was weighed and mixed with 9 mL peptone water (Bioxon, Becton Dickinson de México SA de CV, Mexico) at pH 7.2, and decimal dilutions were then made. Afterwards, one mL was inoculated in soybean trypticase agar to determine the total count of microflora, Staph 10 agar was used to count staphylococcus species and McConkey agar (Bioxon, Bionxon, Becton Dickinson de México SA de CV, Mexico) was used to grow enterobacteria.

2.7 Lipid oxidation determination in burgers

The 2-thiobarbituric acid reactive substances (TBARS) were determined according to the method as described by Nam & Ahn (2003). Ten g of meat sample were homogenized in 90 mL of distilled water using an ultra-Turrax T25 (IKA-Labortechnik,
Staufen, Germany) at 13500 rpm for 20 s. A 1 mL of homogenate was transferred to a test tube to which 0.05 mL of ethanolic solution of butylated hydroxytoluene (7.2%, v/v) was added along with 2 mL of thiobarbituric acid 0.02 M in trichloroacetic acid (20 mM TBA/15% TCA, w/v). This mixture was vortexed and incubated at 80°C for 30 min to develop color. The mixtures were then cooled for 10 min in running water. The tubes were then centrifuged at 3500 rpm for 20 min at 4°C to obtain a supernatant. The absorbance was measured at 532 nm. The amounts of TBARS were expressed as mg of malondialdehyde per kg of meat. For the control, 1,1,3,3-tetra-etoxi-propano (TEP) was used.

### 2.8 Color and pH of burgers

Color was measured in meat as described above using a MicroOptix i-Lab VRV-300 handheld Visible Analyzing spectrophotometer to obtain L*, a* and b* values. For pH, 10 g of meat were homogenized with 90 mL of distilled water and measured using an electrode attached to a pHmeter (Hanna Instruments) until pH was stable.

### 2.9 Statistical analysis

All data were analyzed under a one-way experimental design. An analysis of variance was carried out with the data following the General Linear Model procedure with feeding treatment as the fixed factor using SAS software (SAS Institute Inc., 2004). The statistical model used was: Yi = μ + βi + εij, in which Yij = dependent variable; μ = mean for the variable; βi = the fixed effect of i-th rabbit of the group; and εij = experimental error associated with the observation Yij. When statistical differences were found (*P<0.05) a Tukey comparison test was used.

### 3 Results

#### 3.1 Morphometric characteristics of rabbits consuming Chenopodium ambrosioides infusion

The morphometric characteristics measured were not significant (P>0.05) among treatments (Table 2). However, live weight in control group was slightly higher (1941.6 g) than the groups supplemented with Chenopodium ambrosioides (1879.1 and 1904.1 g, respectively). Therefore, the hot carcass weight was 1047.6, 999.3 and 1030.1 g for control group, and Chenopodium ambrosioides infusion, respectively. Live weight and hot carcass weight has a higher relationship. Animal length and hip circumference in live animals are related to carcass length and hip circumference of the carcass, these variables were also slightly higher in control group, except for the hip circumference in carcass (22.48, 22.50 and 23.50 for C, CA5 and CA10, respectively).

#### 3.2 Carcass quality

The quantity of Chenopodium ambrosioides during the fattening period of the rabbits was not significant (P>0.05) on carcass quality (Table 3). Dressing percentage and empty body weight were not significant (P>0.05) among groups. Likewise, the average weights of lungs, heart, spleen, liver, bladder and kidneys were not significant (P>0.05) indicating that these organs function normally and are not affected by the treatments. This is also an indication that Chenopodium ambrosioides in drinking water used in this work is not toxic for the rabbits. The weight of primal cuts among the different groups was similar (P>0.05), with hind part and legs weighing the most. Dissectible fat obtained from legs was different (P<0.05) among groups. Rabbits which ingested Chenopodium ambrosioides infusion produced a lower fat content compared to control group (22.71 vs 7.46% control and CA5 group, respectively), but there no was difference (P>0.05) among CA5 and CA10 groups (7.46 vs 18.24%, for CA5 and CA10, respectively). Moreover, meat from legs did not show differences (P>0.05) among treatments, but the CA5 group produced a higher content of lean meat (P<0.05).

#### 3.3 Meat quality

The pH, adhesiveness and L* value in meat quality were significant (*P<0.05) with a higher pH value for CA5 treatment, while the control treatment was more adhesive than rabbit meat from rabbits that had consumed Chenopodium ambrosioides infusion (Table 4). The L* value was lower in CA10 treatment, while cooking losses and WHC were not significant (P>0.05) among treatments. Likewise, TPA parameters were not significant (P>0.05) except for adhesiveness as previously mentioned. Chenopodium ambrosioides infusion used as drinking water for rabbits during fattening periods did not have an effect on meat quality, except for L* value which decreased (*P<0.05).

#### 3.4 Burger quality

The total plate count in burgers processed with rabbit meat from animals which consumed Chenopodium ambrosioides have lower counts (P>0.05) than other treatments at day 0 (Table 5). After this period, counts increased and all treatments had no significant differences (P>0.05). During the first week, enterobacteria and staphylococcus groups showed a lower growth in the burgers though there no was differences (P>0.05) among treatments.

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Table 2. Morphometric measurement of rabbits consuming Chenopodium ambrosioides infusion during rabbit fattening.

|                  | C    | CA5  | CA10 | SEM  |
|------------------|------|------|------|------|
| Live weight (g)  | 1941.66 | 1879.16 | 1904.16 | 105.10 |
| Animal length (cm)| 33.31  | 30.66  | 31.66  | 1.03  |
| Hip circumference in live animal (cm) | 26.33 | 25.16 | 23.66 | 1.28 |
| Carcass length (cm) | 30.78  | 30.78  | 30.16  | 0.84  |
| Hip circumference in carcass (cm) | 22.48  | 22.50  | 23.50  | 0.90  |
| Hot carcass (g)  | 1047.66 | 999.33 | 1030.13 | 68.94 |

*C, control; CA5, 5 g.L⁻¹; CA10, 10 g.L⁻¹ of Chenopodium ambrosioides; SEM, standard error of the mean.*
Chenopodium ambrosioides on carcass, meat and burger quality

Finally, after 14 d of storage under refrigeration, temperature enterobacteria and staphylococcus counts in burgers did not show any differences (P>0.05), but CA10 treatment had lower counts than others treatments.

The pH of burgers during storage period did not show any differences (P>0.05) among treatments. The pH for control group fluctuated between 6.05 and 6.44, for the CA5 and CA10 groups were 5.87 and 6.79. The color of burgers processed with rabbit meat from animals who consumed Chenopodium ambrosioides infusion recorded differences between treatments (P<0.05) on day 0 and 7 (Table 6). The L and b* values were lower in CA10 burger group on day 0. After 14 d, the changes in color were significant as both L and b*values were higher on day 0, L value was higher in C group (55.4), b* value decreased after 14 d of storage and CA10 group was the highest among the treatments (1.81). TBARS on day 0 was different among groups (P<0.05) as indicated in Table 6 (0.55, 0.27 and 0.18 for C, CA5 and CA10, respectively). After 7 and 14 days of storage there no were

Table 3. Carcass quality of rabbit drinking Chenopodium ambrosioides infusion during fattening period.

| Treatment       | C           | CA5         | CA10         | SEM  |
|-----------------|-------------|-------------|--------------|------|
| Dressing percentage, % EBW | 53.91       | 53.03       | 54.05        | 0.86 |
| Empty body weight, g | 1764.33     | 1700.16     | 1745.00      | 70.18|
| Skin weight, g.kg⁻¹ EBW | 169.89      | 178.48      | 181.64       | 3.78 |
| Feet, g.kg⁻¹ EBW | 17.96       | 18.97       | 18.72        | 0.09 |
| Lungs, g.kg⁻¹ EBW | 10.19       | 10.29       | 8.16         | 0.66 |
| Heart, g.kg⁻¹ EBW | 4.32        | 3.73        | 5.56         | 0.69 |
| Gastrointestinal tract, g.kg⁻¹ EBW | 89.52       | 90.22       | 93.95        | 3.01 |
| Spleen, g.kg⁻¹ EBW | 0.75        | 0.92        | 0.96         | 0.19 |
| Liver, g.kg⁻¹ EBW | 50.18       | 50.91       | 54.97        | 2.98 |
| Bladder, g.kg⁻¹ EBW | 1.60        | 2.03        | 2.16         | 0.26 |
| Kidneys, g.kg⁻¹ EBW | 7.76        | 8.35        | 8.56         | 0.56 |
| Fore part, g.kg⁻¹ EBW | 247.86      | 249.06      | 244.87       | 2.48 |
| Intermediate part, g.kg⁻¹ EBW | 82.29       | 71.17       | 84.03        | 6.29 |
| Hind part, g.kg⁻¹ EBW | 214.83      | 224.65      | 221.03       | 8.25 |
| Legs, g.kg⁻¹ EBW | 341.19      | 336.42      | 332.16       | 5.19 |
| Head, g.kg⁻¹ EBW | 105.02      | 100.43      | 100.94       | 4.36 |
| Meat, g.kg⁻¹ Legs | 679.18      | 706.75      | 696.66       | 19.27|

Table 4. Meat quality of rabbits drinking Chenopodium ambrosioides infusion during fattening period.

| Treatment       | C           | CA5         | CA10         | SEM  |
|-----------------|-------------|-------------|--------------|------|
| pH₂₄             | 5.50b       | 5.56a       | 5.51ab       | 0.02 |
| Cooking losses, % | 24.14       | 23.01       | 26.11        | 2.04 |
| Water holding capacity, % | 43.73       | 39.15       | 45.51        | 4.40 |
| Adhesiveness, g.s⁻¹ | 0.05b       | 0.02ab      | 0.01b        | 0.01 |
| Resilience       | 0.21        | 0.21        | 0.20         | 0.01 |
| Cohesiveness     | 0.52        | 0.54        | 0.52         | 0.03 |
| Springiness      | 2.81        | 2.76        | 2.52         | 0.73 |
| Hardness, N      | 12.56       | 10.86       | 9.10         | 1.76 |
| L*               | 56.58b      | 55.90ab     | 54.28b       | 0.90 |
| a*               | 5.14        | 5.52        | 4.23         | 1.16 |
| b*               | -14.47      | -16.39      | -12.90       | 2.17 |

C, control; CA5, 5 g.L⁻¹; CA10, 10 g.L⁻¹ of Chenopodium ambrosioides; SEM, standard error of the mean. ab Indicates significant differences using the Tukey test (P<0.05).

Table 5. Bacterial counts in rabbit meat burgers from rabbits drinking epazote infusion during fattening period.

| Treatment       | CTV          | Enterobacteria | Staphylococcus |
|-----------------|--------------|----------------|----------------|
|                 | Log₁₀ UFC g⁻¹ |                |                |
| Day 0           |              |                |                |
| C               | 0.81 ± 0.06a | SC             | SC             |
| CA5             | 0.65 ± 0.06ab| SC             | SC             |
| CA10            | 0.40 ± 0.06b | SC             | SC             |
| Day 7           |              |                |                |
| C               | 2.65 ± 0.21  | SC             | 0              |
| CA5             | 2.18 ± 0.21  | SC             | 0.24           |
| CA10            | 2.22 ± 0.21  | SC             | 0.30           |
| Day 14          |              |                |                |
| C               | 2.72 ± 0.15  | 2.08 ± 0.07    | 0.40 ± 0.09    |
| CA5             | 2.50 ± 0.15  | 1.90 ± 0.07    | 0.24 ± 0.09    |
| CA10            | 2.40 ± 0.15  | 1.64 ± 0.07    | SC             |

C, control; CA5, 5 g.L⁻¹; CA10, 10 g.L⁻¹ of Chenopodium ambrosioides. ab Indicates significant differences using the Tukey test (P<0.05). SC, without bacterial growth.
differences among groups with regard to TBARS (P>0.05), but CA5 and CA10 groups had lower levels than control group.

4 Discussion

To the best of our knowledge, this is the first report on the use of Chenopodium ambrosioides infusion to feed rabbits, although studies have been carried out on the beneficial effects of Chenopodium ambrosioides extracts and essential oils when used directly on pork and beef (Villalobos-Delgado et al., 2016, 2017). The results of this study found similar morphometric measures among treatments that indicate Chenopodium ambrosioides may not have toxins or molecules inhibiting the growth of rabbits after weaning at the concentrations used in this trial. The morphometric characteristics are normal for the age and the number of days of fattening (Average = 63 d) of the rabbits. Similar results were obtained by Oloruntola et al. (2015) who reported that rabbit fed with different forages have an average slaughter weight of 1343 g, while Ozung et al. (2011) produced fattened rabbits after 140 d obtaining an average slaughter weight of 1974 g using different legumes for their feed. Mancini et al. (2018) reported a final body weight of 2438 g in rabbits during 90 d of fattening. Sun et al. (2017) found a live weight average for control treatment of 2236 g after 80 d of fattening.

One part of this study was focused on evaluating two quantities of Chenopodium ambrosioides for growth efficiency, effects on organs and rabbit carcass. Organ weight changes in animals are important as they can indicate ill health. The results of this trial indicate that the organs were similar among animal groups. Oloruntola et al. (2015) found no change in the weight of kidneys in their study, which implied a safe mixture of leguminous and non-leguminous plants. In the case of fore, intermediate and hind parts, legs and head were not significant (P>0.05). When legs were dissected into meat, bone and fat, only the last value was higher (P<0.05) in control group. Cardinalli et al. (2015) showed that weight and carcass dressing were higher in rabbits fed with oregano and rosemary. Dal Bosco et al. (2012) added different kinds of olive pomaces to feed rabbits, and differences were observed in cold carcass weight, while Rotolo et al. (2013) and Kone et al. (2015) found no differences in carcass quality and dressing when adding oregano and their essential oil, respectively, to feed for growing rabbits.

With regard to previous studies using different plants or extracts to feed rabbits, Dal Bosco et al. (2012) showed no color change in rabbits fed with olive leaves. Furthermore, Dalle-Zotte et al. (2014) did not find differences when rabbits were fed with spirulina and thyme, while Sun et al. (2017) found that neither pH nor drip loss were modified in meat when Moringa oleifera was consumed by growing rabbits. In the same study, L* value measured in two different muscles was similar between treatments. Simonová et al. (2010) found similar results using oregano and sage to feed rabbits, reporting pH from 5.6 to 5.7, L* value of between 48 and 50.9, and WHC between 32 and 33% without significant differences between treatments (P<0.05).

Ranucci et al. (2015) analyzed cooked ham for microbiological analysis and links to pH at 0, 10 and 20 d, and no differences were found between treatments when pigs were fed with oregano extract and sweet chestnut. Soultos et al. (2009) added oregano essential oil to rabbit feed and found that total plate count decreased after 12 d storage. Villalobos-Delgado et al. (2017) reported a low total plate count in ground pork meat added with aqueous and methanolic Chenopodium ambrosioides extracts. Biswas et al. (2012) added curry and peppermint extract to ground pork meat and color was analyzed over 12 d and showed differences in L*, a* and b* values. Martinez et al. (2012) developed a burger with vitamin E and olive oil as additives and found differences in L*, a* and b* values over 8 d of study.

Burgers processed using rabbit meat obtained from animals who had consumed Chenopodium ambrosioides infusion have a reduced lipid oxidation (*P<0.05) at day 0. Similar results were found by Mancini et al. (2016) in hamburgers elaborated using curcuma or ascorbic acid during 7 d of storage. Cardinalli et al. (2015) fed rabbits with oregano extract and vitamin E and recorded lower levels of malonaldehyde.

5 Conclusion

Dietary supplementation of Chenopodium ambrosioides infusion does not affect carcass and meat quality during rabbit fattening. However, the microbiological and oxidative properties in burgers resulted in low bacterial growth and oxidation at the initial time of processing.
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