



Dietary supplementation effects with *Ruta graveolens* on performance, carcass traits and meat quality on rabbits



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Abstract:

Ruta graveolens is a weed that can be used to feed rabbits. The aim of this study was to determine growth performance, carcass and meat quality of rabbits after their dietary supplementation with *Ruta graveolens*. Sixty (60) weaned rabbits were randomly assigned to five treatments; control diet (C) or diets supplemented either with leaves (25RL or 50RL) or complete plant of *Ruta graveolens* (25CP or 50CP). The use of *Ruta graveolens* has a similar ($P>0.05$) growth performance to the control group and feed conversion rate. Carcass quality was different ($P<0.05$) among treatments in empty body weight, empty gastrointestinal tract and fat. The pH decreased when *Ruta graveolens* was used to feed growing rabbits, but meat produced better texture parameters than control group. The results obtained in the present study suggest that *Ruta graveolens* can be considered as an alternative feed source in the diets of rabbits.

Key words: Aromatic plant, Meat quality, Growth efficiency, Rabbit.

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In recent years, there has been a growing interest in the use of plants in animal production due to their bioactive compounds that improve productive performance, carcass traits and meat quality⁽¹⁾. Several medicinal plants are used in diets of fattening rabbits as they are a source of phytochemicals that possess properties such as antioxidants or antimicrobials^(1,2). Bilberry pomace dietary supplementation was used as a feeding strategy to produce favourable nutritional performance and changes in fatty acids in rabbit meat content⁽³⁾. Moreover, the supplementation with oregano and rosemary had positive effects on growth performance and carcass traits of fattening rabbits⁽⁴⁾. But, dietary supplementation with onion, cranberry, strawberry and their extracts did not produced differences on productive performance and meat quality and oxidative stability in weaned rabbits⁽⁵⁾.

Ruta graveolens is a plant used in traditional medicine around the world and is known by different names, such as rue, herb of grace, and others in several languages⁽⁶⁾. This plant is recognised due its antimicrobial^(7,8,9) and antioxidant^(1,2) properties. *Ruta graveolens* is a plant with a high content of secondary metabolites, such as coumarins, alkaloids, volatile oils, flavonoids and phenolic acids, which are responsible for several biological effects⁽⁶⁾.

It seems that, this is the first time that *Ruta graveolens* is been used in diets of growing rabbits. However, the extracts or leaves of this herb were previously used to investigate *in vitro* antibacterial activity⁽⁷⁾. *Ruta graveolens* and its flavonoids can be used as antimicrobial⁽⁸⁾ or antioxidant agent^(8,10). Rabbit production faces a problem during rabbit growth. Weaning is a crucial period for rabbits, since increased digestive disturbances are observed at this age, possibly due to increased susceptibility to several pathogens caused by enhanced stress rates. One disease that is characterised by diarrhoea, abdominal bloating and distention of intestinal cavity is the epizootic rabbit enteropathy; this disease has high morbidity and mortality rates⁽²⁾. As *Ruta graveolens* has antimicrobial and antioxidant properties could be used as a feed additive to increase productive parameters and obtain better carcass and meat quality.

Based on the above considerations, this study was conducted to determine effects of feeding *Ruta graveolens* using leaves or complete plant at two different dietary levels on productive performance, carcass traits and meat quality of growing rabbits.

Trial was conducted in the experimental rabbitry of the Instituto de Ciencias Agropecuarias (Tulancingo, Hidalgo, México) and was approved by the Animal Care Committee of the Universidad Autónoma del Estado de Hidalgo. Sixty (60) weaned rabbits (35 d of age, 25 males and 35 females) were assigned randomly to five treatments (n= 12 by treatment), each with three replications; rabbits were housed in cages (90 cm x 60 cm) equipped with manual feeders and

automatic drinkers. Rabbits were reared in controlled conditions with average temperature of 20 °C. Rabbits were hybrids of New Zealand, California and English Spot breeds with an average weight of 756.79 ± 97.69 g. The feed used was pelletized using a pellet machine (SKJ120 model, Shandong, China). Animals were fed *ad libitum* using isoproteic (17 % of crude protein) and isoenergetic (2.4 Mcal/kg of digestible energy) experimental diets, according to De Blas and Mateos⁽¹¹⁾. Animals were divided in the following treatments: control, supplemented with 25 and 50 g of rue leaves/kg of food, or supplemented with 25 and 50 g of complete plant/kg of food (C, 25RL, 50RL, 25CP, 50CP diets, respectively) as indicated in Table 1.

Table 1: Experimental diets ingredients

Ingredient	Treatments				
	C	25RL	50RL	25CP	50CP
	Kg				
Corn	1.79	1.81	1.82	1.81	1.82
Oat straw	1.61	1.37	1.14	1.37	1.14
Bran wheat	1.07	0.99	0.99	0.99	0.99
Soybean husk	0.78	0.78	0.78	0.78	0.78
Soybean meal	1.54	1.60	1.60	1.60	1.60
Canola meal	0.77	0.77	0.77	0.77	0.77
Sorghum	1.90	1.90	1.90	1.90	1.90
Molasses	0.25	0.25	0.25	0.25	0.25
Vit. and minerals premix	0.30	0.30	0.30	0.30	0.30
<i>Ruta graveolens</i> (rue)	0	0.23	0.46	0.23	0.46
Calculated composition					
Crude protein, %	16.6	16.7	16.6	16.7	16.7
NDF, %	16	16	16	16	16
ADF, %	8.7	8.7	8.7	8.7	8.7
ED, Mcal/kg	2.5	2.5	2.5	2.5	2.5
Ca, %	0.8	0.8	0.8	0.8	0.8
P, %	0.5	0.5	0.5	0.5	0.5

C=Control, 25RL= 25 g.kg⁻¹ of rue leaves; 50RL= 50 g.kg⁻¹ of rue leaves; 25CP= 25 g.kg⁻¹ of complete plant; 50CP= 50 g.kg⁻¹ of complete plant.

The plant was obtained in Tulancingo, Hidalgo State in the centre of México. After transporting to the lab, plants were separated into leaves (RL) and complete plant (CP) and dried at room temperature for 5 d in shadow. All parts were grounded into a miller (Mexicana de Suministros Agropecuarios SA de CV, Tulancingo, Hidalgo, México) using a sieve with a 5 mm diameter. Once

grounded, the plant was stored in a dark plastic container until use. During the experiment, rabbits were weighed individually each week, and feed intake was daily measured during the fattening period. From this data, daily average weight gain (DAWG), total weight gain (TWG), and feed conversion ratio (FCR) were calculated.

Rabbits were slaughtered at 63 d old in the meat lab belonging to the Instituto de Ciencias Agropecuarias in Tulancingo, Hidalgo, Mexico. Before rabbits were slaughtered there was no fasting. Animal length was determined in live animals by measuring distance from the atlas to last ischion vertebra while animal was in a dorsal position. Pelvis and lumbar circumference on live animal and carcass were measured using a measuring tape. Animals were weighed, mechanical stunned and processed according to the national legislation⁽¹²⁾. Hot carcasses, liver, kidneys, gastrointestinal tract full and empty, bladder full and empty, scapular fat, perirenal fat, and skin were weighed, and then carcasses were stored under refrigeration at 4 °C for 24 h. Empty body weight was calculated by measuring weight differences between full and empty gastrointestinal tract and bladder.

Carcasses were divided after 24 h of refrigerated storage⁽¹³⁾. Head was cut at the level of atlas, forequarter was obtained by cutting between 6th and 7th ribs, thoracic cage was determined by cutting in last rib, and loin was obtained between 6th and 7th lumbar vertebra cutting transversally to vertebral column to finally obtain hind leg. Hind legs were detached in fat, bone and meat. All these parts were weighed separately.

Meat colour was measured on loin surface between last rib and 6th lumbar vertebra at room temperature (22 °C) using a portable colorimeter i-Lab S560 (Microptix, Wilton, Maine, USA). Values were recorded in terms of CIE L*a*b* colour space using a standard illuminant D65 and observer of 2° as indicated in American Meat Science Association meat colour measurement guidelines⁽¹⁴⁾, pH was determined using a pH meter suitable for meat samples (HI99163 model, Hanna instruments, Cluj-Napoca, Romania). Water holding capacity (WHC) was expressed as percentage of water loss⁽¹⁵⁾. Cooking losses were measured in loins. Samples were put in a plastic bag and cooked at 80 °C until meat internal temperature reaches 68 °C using a check temp digital thermometer (Hanna Instruments, Portugal). Cooked samples were cooled at room temperature and weighed, cooked losses was calculated by weight differences before and after cooking and expressed as a percentage. Cold samples were used to determine texture profile analysis (TPA) by cutting cubes (1 cm each side) and then compressing to 50 % perpendicular to the muscle fibre direction using 1 mm/s of crosshead speed⁽¹⁶⁾. After that hardness, cohesiveness, springiness and chewiness were calculated using Texture Pro CT software on a Brookfield CT3 texture analyser (Brookfield, Middleboro, MA, USA).

An analysis of variance was carried out with obtained data following the general linear model procedure, with feeding treatment being the fixed factor, using the SAS Institute software⁽¹⁷⁾. When statistical differences were found ($P<0.05$), a Tukey comparison test was used.

Daily gain weight, total weight gains and feed conversion rate during the fattening period in rabbits are shown in Table 2. Daily gain during first and fourth weeks of growing period showed statistical differences among treatments ($P<0.05$). Highest total weight gains in fattening rabbits ($P<0.05$) was found in treatments C and 25CP ($1,175 \pm 131$ and $1,190 \pm 186$, respectively), moreover, control group was different ($P<0.05$) to 25RL group. Feed conversion rate values during the fattening period ranged from 2.21 and 2.78 for 25CP and 50CP treatments, respectively. However, 50CP group had a greater feed conversion rate to control group (2.78 ± 0.58 and 2.22 ± 0.24 for 50CP and Control group, respectively). The lowest daily gain during first week (39.15) was found in 25RL group and highest (52.22) in control treatment, but in fourth week highest daily gain (39.76) was observed in 25CP group and the lowest was in 50RL treatment. Finally, the highest total daily gain weight (41.98) was detected in control group and lowest was (33.35) in 25RL group. The highest weight gain (1,175 g) was observed in control group and the lowest (934 g) in the 25 RL group. *Ruta graveolens* use as a complete plant at 25 g.kg^{-1} in fattening rabbits was similar to control group in daily weight gain and feed conversion rate every week during fattening period.

Table 2: Effects of *Ruta graveolens* dietary supplementation on performance of fattening rabbits (Mean \pm SD)

	Treatments				
	C	25RL	50RL	25CP	50CP
LW35d	629.54 \pm 15.06 ^e	686.68 \pm 20.03 ^d	765.62 \pm 95.4 ^c	821.63 \pm 22.14 ^b	889.54 \pm 19.80 ^a
LW63d	1763.18 \pm 150.0 ^b	1632.50 \pm 95.4 ^{ab}	1803.75 \pm 169.49 ^{ab}	1928.48 \pm 255.59 ^a	1816.98 \pm 222.19 ^a
DGW1 (g)	52.22 \pm 8.42 ^a	39.15 \pm 7.92 ^b	44.71 \pm 9.89 ^{ab}	50.00 \pm 11.54 ^{ab}	45.31 \pm 7.80 ^{ab}
DGW2 (g)	40.63 \pm 6.51	33.31 \pm 11.09	44.47 \pm 6.55	41.74 \pm 9.07	37.06 \pm 12.65
DGW3 (g)	39.44 \pm 6.88	29.92 \pm 9.45	34.49 \pm 8.09	38.49 \pm 11.64	36.82 \pm 8.72
DGW4 (g)	35.63 \pm 5.69 ^a	33.05 \pm 12.28 ^{ab}	17.96 \pm 17.93 ^b	39.76 \pm 7.44 ^a	25.87 \pm 17.42 ^{ab}
DGWT (g)	41.98 \pm 4.70 ^a	33.35 \pm 3.39 ^b	35.40 \pm 4.34 ^{ab}	42.49 \pm 6.65 ^a	36.27 \pm 7.42 ^{ab}
TWG (g)	1175.55 \pm 131.68 ^a	934.09 \pm 95.17 ^b	991.42 \pm 121.74 ^{ab}	1190.00 \pm 186.41 ^a	1015.55 \pm 207.85 ^{ab}
FCR	2.22 \pm 0.24 ^b	2.48 \pm 0.28 ^{ab}	2.48 \pm 0.30 ^{ab}	2.21 \pm 0.32 ^b	2.78 \pm 0.58 ^a

C= Control, 25RL= 25 g.kg⁻¹ of rue leaves; 50RL= 50 g.kg⁻¹ of rue leaves; 25CP= 25 g.kg⁻¹ of complete plant; 50CP= 50 g.kg⁻¹ of complete plant; LW35d= Live weight at 35 d of age. LW63d= Live weight at 63 d of age. DGW1-4= Daily gain weight during 1-4 fattening weeks. DGWT= Daily gain weight during all fattening period. TWG= Total weight gained. FCR= Feed conversion ratio.

^{abc} Different superscripts in the same row indicate significant differences ($P<0.05$).

So far, there is few information about rue used as a dietary supplementation in rabbits. The use of 2.5 g.kg⁻¹ of rue in fattening rabbits increase carcass weight and meat proportion⁽¹⁸⁾. Some researchers, did not find a significant difference in productive parameters of rabbits fed diets with *Trametes maxima*⁽¹⁹⁾. Moreover, the supplementation with *Silybum marianum* at the levels of 5 or 10 g kg⁻¹ into the diet of rabbits, found a similar performance productivity⁽²⁰⁾. At the same time, *Lythrum salicaria* supplemented in diets to growing rabbits did not find an increase in growth performance⁽²¹⁾. *Ruta graveolens* has been reported to have antioxidant, antibacterial and anticancer activity *in vitro*^(8,9). A similar growth performance to control group was shown in this study, especially when animals ingest the complete plant. The stems, leaves, and flower of rue are used in human traditional medicine as anthelmintic, antiparasitic, antidiarrheic and antimicrobial properties⁽⁷⁾.

Effects of dietary supplementation with *Ruta graveolens* on rabbit's carcass quality are shown in Table 3. Empty body weight was significantly different among groups ($P<0.05$). Rabbits supplemented with 25CP (1,780.33 ± 216.84 g) and 50CP (1,821.78 ± 265.10 g) were heavier than those supplemented with 25RL (1,496.33 ± 74.36 g); however, there were no significant differences ($P>0.05$) between control group (1,645.00 ± 13.36 g) and rabbits supplemented with rue complete plant (25CP=1,780.33 ± 216.8 and 50CP=1,821.78 ± 265.1 g) producing lighter carcasses ($P<0.05$) than control group (control=1.94 vs 50CP=0.56). These findings are supported by the increase of live weight and weight gained in treatment of 50CP. The use of rue to supplement diets to grow rabbits could be used to promote carcass quality. However, previous researchers did not find differences in carcass traits when diets were supplemented with other plants. Carcass traits of rabbits fed with *Silybum marianum* were similar among treatments⁽²⁰⁾. Likewise, another research group that replaced conventional ingredients with *Amaranthus dubius* at levels up to 32 % did not observe a negative impact in carcass traits, suggesting that this plant should be a potential substitute of conventional ingredients in rabbit diet formulations⁽²²⁾. By other hand, the supplementation with *Lythrum salicaria* to growing rabbits, did not find differences in carcass traits, suggesting that this plant could be used in rabbit diets during the fattening period⁽²¹⁾.

Table 3: *Ruta graveolens* dietary supplementation effects on carcass traits of fattening rabbits (Mean \pm SD)

Variable	Treatments				
	C	25RL	50RL	25CP	50CP
Carcass hot weight	976.11 \pm 46.69 ^{ab}	897.77 \pm 58.90 ^b	961.87 \pm 118.62 ^{ab}	1061.11 \pm 125.04 ^a	1097.22 \pm 182.62 ^a
EBW, g/kg	1645.00 \pm 13.36 ^{ab}	1496.33 \pm 74.36 ^b	1617.88 \pm 187.04 ^a	1780.33 \pm 216.84 ^a	1821.78 \pm 265.10 ^a
Hot dressing, %	59.44 \pm 2.29	60.10 \pm 4.59	59.40 \pm 1.92	59.66 \pm 2.02	60.01 \pm 2.13
CCY, %	57.48 \pm 2.01	58.05 \pm 3.92	56.95 \pm 2.162	57.14 \pm 1.872	56.25 \pm 2.74
Viscera	250.40 \pm 2.11	278.20 \pm 3.46	288.40 \pm 5.55	287.70 \pm 2.44	274.4 \pm 7.13
Heart	3.40 \pm 0.10	3.00 \pm 0.10	3.30 \pm 0.10	0.31 \pm 0.69	0.39 \pm 0.94
Lungs	7.60 \pm 1.90	8.40 \pm 3.20	7.80 \pm 2.16	8.60 \pm 1.99	7.30 \pm 1.03
Spleen	0.60 \pm 0.10	0.70 \pm 0.20	0.60 \pm 0.09	0.60 \pm 0.06	0.70 \pm 0.30
Liver	44.7 \pm 10.83	43.50 \pm 10.40	47.00 \pm 7.84	49.30 \pm 6.76	38.80 \pm 11.08
Kidneys	6.70 \pm 0.90	6.30 \pm 0.74	6.40 \pm 1.18	6.90 \pm 1.16	7.20 \pm 0.86
EGTW	258.28 \pm 46.94 ^b	260.95 \pm 36.12 ^b	294.93 \pm 34.50 ^{ab}	310.85 \pm 49.04 ^{ab}	317.30 \pm 30.13 ^a
Bladder	2.10 \pm 0.50	2.70 \pm 1.11	1.70 \pm 0.47	2.20 \pm 1.08	2.80 \pm 1.19
Kidney fat	8.00 \pm 1.60	7.60 \pm 2.40	6.80 \pm 3.24	8.30 \pm 4.22	9.60 \pm 2.47
Scapular fat weight	2.40 \pm 0.70	2.10 \pm 0.80	1.90 \pm 0.69	2.20 \pm 1.13	3.20 \pm 1.79
Head	57.9 \pm 0.41	63.40 \pm 7.90	61.30 \pm 5.53	59.40 \pm 5.43	57.6 \pm 8.79
Forepart weight	139.30 \pm 6.90	137.60 \pm 11.00	138.00 \pm 6.16	140.80 \pm 7.04	139.90 \pm 6.32
IPW	53.10 \pm 4.70	57.10 \pm 9.50	54.70 \pm 8.07	52.80 \pm 4.80	54.00 \pm 7.61
Hind part weight	111.80 \pm 7.80	107.20 \pm 12.60	106.50 \pm 10.37	110.70 \pm 11.58	112.10 \pm 9.70
Legs	203.70 \pm 8.70	204.80 \pm 12.80	199.90 \pm 7.83	196.10 \pm 11.26	196.60 \pm 9.08
Meat	152.80 \pm 10.10	153.40 \pm 11.10	147.40 \pm 7.58	150.00 \pm 8.49	147.20 \pm 14.56
Bone	45.40 \pm 7.30	46.50 \pm 7.00	48.70 \pm 9.46	41.10 \pm 6.55	44.60 \pm 6.84
Dissectible fat	1.94 \pm 0.12 ^a	1.44 \pm 0.97 ^a	0.81 \pm 0.62 ^{ab}	0.75 \pm 0.68 ^b	0.56 \pm 0.59 ^b

C= Control, 25RL= 25 g kg⁻¹ of rue leaves; 50RL= 50 g kg⁻¹ of rue leaves; 25CP= 25 g kg⁻¹ of complete plant; 50CP= 50 g kg⁻¹ of complete plant.

EBW= empty body weight; CCY= chilled carcass yield; EGTW= empty gastrointestinal tract weight; IPW= intermedia part weight.

^{abc} Different superscripts in the same row indicate differences ($P < 0.05$).

Colour, pH, WHC and other variables of meat from rabbits fed with *Ruta graveolens* supplemented diets are presented in Table 4. Significant differences ($P<0.05$) were found in all measured traits, apart from WHC and hardness. *Ruta graveolens* leaves affected ($P<0.05$) the colour of rabbit meat, providing lower L^* (25RL= 55.3) and b^* (50RL= 8.22) values, and complete plant resulted in lower a^* values (50CP= 0.61) compared to control group (1.23). Likewise, pH decreased when rabbits were fed diets supplemented with 25 g kg^{-1} of rue complete plant. In the results of texture profile analysis, hardness did not shown differences ($P>0.05$) among groups, but resilience, cohesiveness, springiness and chewiness were different between control and *Ruta graveolens* groups. Resilience and cohesiveness were higher in meat of rabbits fed with *Ruta graveolens* leaves, while springiness was higher in 25RL (3.05), 50RL (4.00) and 25CP (2.35) to control group (0.60).

Table 4: *Ruta graveolens* dietary supplementation effects on meat quality of fattening rabbits (Mean \pm SD)

Variable	Treatments				
	C	25RL	50RL	25CP	50CP
L^*	57.90 \pm 3.21 ^a	55.30 \pm 3.60 ^b	58.07 \pm 3.91 ^a	58.21 \pm 3.05 ^a	56.72 \pm 2.84 ^{ab}
a^*	1.23 \pm 1.75 ^{ab}	1.65 \pm 1.43 ^a	1.18 \pm 1.61 ^{ab}	1.29 \pm 1.17 ^{ab}	0.61 \pm 1.40 ^b
b^*	9.74 \pm 2.04 ^a	9.36 \pm 2.00 ^{ab}	8.22 \pm 2.45 ^b	8.96 \pm 2.23 ^{ab}	9.42 \pm 2.19 ^{ab}
pH	5.85 \pm 0.11 ^a	5.81 \pm 0.04 ^{ab}	5.77 \pm 0.03 ^{ab}	5.75 \pm 0.05 ^b	5.80 \pm 0.19 ^{ab}
WHC, %	21.12 \pm 5.19	20.02 \pm 4.86	18.85 \pm 6.19	18.06 \pm 5.69	19.48 \pm 5.26
Hardness, N	8.31 \pm 2.17	9.76 \pm 3.13	9.36 \pm 3.30	10.18 \pm 4.37	10.59 \pm 3.62
Resilience	3.05 \pm 0.17 ^b	5.08 \pm 0.77 ^a	4.94 \pm 0.42 ^a	2.63 \pm 0.46 ^b	2.75 \pm 0.04 ^b
Cohesiveness	0.24 \pm 0.02 ^c	0.48 \pm 0.08 ^{ab}	0.54 \pm 0.01 ^a	0.44 \pm 0.08 ^b	0.22 \pm 0.01 ^c
Springiness	0.60 \pm 0.13 ^c	3.05 \pm 1.17 ^b	4.00 \pm 0.52 ^a	2.35 \pm 0.62 ^b	0.57 \pm 0.12 ^c
Chewiness	12.81 \pm 0.76 ^b	19.47 \pm 5.91 ^a	22.42 \pm 4.59 ^a	8.75 \pm 1.97 ^b	13.56 \pm 2.79 ^b

C=Control, 25RL= 25 g kg^{-1} of rue leaves; 50RL= 50 g kg^{-1} of rue leaves; 25CP= 25 g kg^{-1} of complete plant; 50CP= 50 g kg^{-1} of complete plant.

^{abc} Different superscripts in the same row indicate differences ($P<0.05$).

Other research did not find differences in meat quality with dietary supplementation of plants fed to growing rabbits. Meat quality did not affect fed rabbits with *Silybum marianum*⁽²⁰⁾. Replace conventional ingredients for *Amaranthus dubius* at levels up to 32 %, there was not negative impact in meat quality, suggesting this plant could be a potential substitute of conventional ingredient in rabbit diets formulation⁽²²⁾. Similar results were obtained using *Lythrum salicaria* to growing rabbits, did not find differences in carcass traits and suggested that this plant could be used in rabbit feeding during the fattening period⁽¹⁹⁾.

The pH has been associated with the colour. Low pH values (4.49) were found in the *Longissimus dorsi* muscle in rabbits fed with *Amaranthus dubius*⁽²¹⁾. While L* values around to 54 and pH around to 5.6 were reported when rabbits were fed with purple loosestrife⁽²⁰⁾. Colour and pH of rabbit meat can be affected by age, breed, muscle type, sex, feeding, ante mortem and post mortem conditions and others factors^(23,24).

Ruta graveolens inclusion in diets of fattening rabbits can be considered as a viable feeding alternative to maintain high production parameters and obtain similar carcass traits and meat quality characteristics to conventionally fed rabbits.

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