



Egg production and quality characteristics of laying hens fed diets supplemented with dry caper (*Capparis spinosa*) leaf powder

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Received: 23-06-2016

Accepted: 09-03-2017

DOI: 10.18805/ijar.B-556

ABSTRACT

An in vivo experiment was conducted to determine the effect of dietary inclusion of dry Caper (*Capparis spinosa*) leaf powder (DCL) on egg production and egg quality of laying hens at the beginning of the laying period. Four groups of commercial hens (ATAK-S) were fed with diets containing 0, 5, 10 and 15 g DCL/kg. The results demonstrated that dietary DCL had no effect on feed intake, feed efficiency, egg mass (EM) and egg production parameters ($P>0.05$). Hen-day egg production (HDP) value of the groups was 69.52%, 64.97%, 68.02% and 71.72% respectively. Trolox equivalent antioxidant capacity (TEAC), ferric reducing antioxidant power (FRAP) and total phenolics amount of DCL were 51.06 ± 2.300 mmol trolox/kg, 50.10 ± 3.488 mmol TEAC/kg and 2.78 ± 0.050 g gallic acid equivalents (GAE)/kg respectively. Based on the results from the current study, the dietary DCL with levels up to 15 g/kg neither reduced productivity performance traits nor improved egg quality characteristics.

Key words: Antioxidant activity, Dry caper leaf powder, Egg quality, Productivity performance, Laying hens.

INTRODUCTION

Keeping farm animals healthy is vital to obtain healthy animal products. In the last decade, the use of additives of natural origin in animal nutrition has been invigorated (Frankic *et al.*, 2009). Numerous earlier researches focused on the clarification of the biochemical structures and physiological functions of various aromatic plants, their extracts and essential oils. Thus various alternative feeding strategies are being considered and incorporated in the diets of laying hen to improve the gut health of the birds, enhance productive performance, and improve egg quality (Olobatoke and Mulugeta, 2011; Kaya *et al.*, 2013; Lokaewmanee *et al.*, 2014).

Caper belonging to Capparaceae family, are the fruits of perennial shrubs of the genus *Capparis*. Caper flower buds, root, fruits and young shoots are used as foodstuffs. It is a plant of tropical/subtropical and arid areas. Caper has several uses for its especially medicinal and aromatic properties (Özcan *et al.*, 2010). Caper is one of the most common aromatic plants found in the Mediterranean Basin. The main producer of Caper plants are countries from the

Mediterranean Basin such as Turkey, Morocco, Spain, Greece, France and Italy where *Capparis spinosa* is predominant and the most important commercial species (Tlili *et al.*, 2009; Yildirim and Gürkan, 2010).

Chemical experiments on caper have claimed that different parts of the plant are rich source of many beneficial chemical compounds particularly glucocapperin, rutin, spermidine, quercetin, kaempferol, stigmaterol, campesterol, tocopherols and carotenoids (Tlili *et al.*, 2010). Results from recent studies has reported that caper possesses antioxidant activities due to the high concentration of phenolic compounds, tocopherols and carotenoids (Germano *et al.*, 2002) and some medicinal properties such as immunostimulant (Ghule *et al.*, 2006), antitumoral (Wu *et al.*, 2003), hypolipidemic (Eddouks *et al.*, 2005) and antibacterial (Halkier, 1999). Caper leaf and its other parts such as buds, flower, root and fruits have not been used in laying hen even though it has a great potential to be used as feed additive in livestock diets due to its multifunction properties. Considering the above benefits, addition of different DCL levels may cause enhancement of the activity

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Table 1: Ingredients and nutrient composition of basal diet (g/kg)

Ingredients	20–32 weeks	Calculated nutrients composition	
Maize	294	ME (MJ/kg)	11.72
Wheat	277	Crude protein	172.0
Soybean meal (47% CP)	92	Calcium	36.0
Barley	30	Available phosphorus	4.0
Sunflower meal (36% CP)	80	Lysine	8.0
Full-fat soybean	100	Methionine	4.5
Soybean oil	24	Methionine+Cystine	7.6
Sodium bicarbonate	1	Analyzed nutrients composition	
Dicalcium phosphate	14	Dry matter	898.0
Marble powder	83	Crude protein	168.0
Salt	2	Ether extract	36
Vitamin–mineral premix1	3	Crude fibre	67
		Ash	136

1 Each kg of vitamin–mineral premix contained: vitamin A, 4800000 IU; vitamin D₃, 1200000 IU; vitamin E, 12000 IU; vitamin K₃, 1600 mg; vitamin B₁, 1200 mg; vitamin B₂, 2400 mg; vitamin B₃, 12000 mg; vitamin B₅, 4000 mg; vitamin B₆, 2000 mg; vitamin C, 20000 mg; vitamin B₁₂, 6 mg; biotin, 20 mg; folic acid, 400 mg; choline, 120000 mg; Cu, 2000 mg; Fe, 24000 mg; Mn, 32000 mg; Se, 60 mg; Zn, 24000 mg; Co, 200 mg; I, 800 mg.

of the endogenous antioxidative enzymes, an improvement in digestive tract performance and improve the egg production in laying hens. Therefore, the aim of the present study was to test whether dietary supplemented DCL influenced egg production and egg quality of laying hens at the beginning of the laying period.

MATERIALS AND METHODS

Experimental design and diets: This study was approved by the Animal Experimentation Ethics Committee (Process no. 307/2011 HADYEK–039) of Gaziosmanpasa University. Prior to the experimental period, the birds were given two weeks adaptation period before the trial began and the trial lasted 12 weeks. Totally eighty commercial ATA–S (Turkish native hybrid) brown layers at the age of 20 weeks were randomly allocated to one of four treatments with four replicates each containing five hens in each group. The experiment was conducted with completely randomized design. Five birds were kept in wire cages (450 cm² per bird) in a windowed poultry house at a light regimen of 16 h light and 8 h dark. Laying hens had *ad libitum* access to water and diets throughout the experimental period. All hens were healthy over the experimental period. Three portable temperature and relative humidity loggers were used to monitor indoor temperatures and relative humidity at a level equal to animal height. The average temperature, relative humidity and light intensity of the entire experimental period were 15.74±0.129 °C, 48.15±6.410% and 14.76±3.828 lux respectively.

Layers in each treatment were fed with one of four experimental diets including one basal diet as control and three corn-soybean meal based diets with addition of 5 g, 10 g and 15 g/kg of a DCL. Organically grown herb of caper (*Capparis spinosa*) was harvested in Tokat province, Turkey

and the leaves were separated from the boughs. The raw material consisted of leaves that were distributed uniformly as a thin layer on the trays and dried in an oven at 40°C for 24 h. Dried plant material was mechanically ground 1 mm diameter particle size with a laboratory mill to obtain a homogenous powder.

The diets were isonitrogenous and isocaloric and were formulated to meet or slightly exceed the nutrient requirement of laying hens according to NRC (1994). Diet samples were analyzed in duplicate for the contents of dry matter (method 945.15), crude protein (Kjeldahl N×6.25, method 990.03), ether extract (method 945.16), crude fiber (method 962.09) and ash (method 967.05) using the AOAC procedures (2000). The ingredients and calculated nutrient level of the basal diet are shown in Table 1.

Egg production performances and egg quality characteristics: Hens were weighed individually at the beginning and the end of the experiment. Feed consumption was recorded weekly and calculated as g/hen/day. Viability were visually observed and recorded daily throughout the entire experimental period. The value of feed efficiency was calculated as feed/egg (g/g).

The first egg was obtained after 23 weeks age in all the groups. Eggs from each replicate were collected twice a day (morning time; 09:00–11:00 and afternoon time; 14:00–16:00) and weighed using electronic balance at the same time every day to calculate HDP and HHP, egg weight and EM from 20 to 32 weeks of age. In addition, age at point-of-lay, first egg weight, age (day) at 10, 20, 30 and 50% egg production were recorded. Thirty two eggs were randomly collected from each experimental group on the last day of every 2 weeks to assess egg quality. Immediately after sampling, eggs were assayed for quality. The egg quality traits were specific gravity, eggshell breaking strength, shell thickness and albumen pH. All eggs were weighed

Table 2: The formulas of some egg quality traits

Criteria	Abbrev.	Equation
¹ Hen-day production, %	HDP	(Number egg produced / Number live hens) x 100
¹ Hen-house production, %	HHP	(Number egg produced / Number live hens in initial experiment) x 100
² Shape index, %	SI	(Egg width /Egg length) x 100
² Yolk index, %	YI	(Yolk height/yolk diameter) x 100
² Albumen index, %	AI	[Albumen height/(long diameter of albumen+short diameter of albumen / 2)×100]
³ Egg surface area (cm ²)	ES	3.9782 × egg weight ^{0.70}
⁴ Shell weight (g)	SW	(2.0341xEgg weight)-[(2.1014xEgg weight)/Specific gravity]
⁵ Haugh unit (score)	HU	100 log (Albumen height+7.57-1.7 W ^{0.37})
⁶ Egg mass (g/hen/day)	EM	(weekly egg number in replicate x average egg weight)/100

¹North (1984), ²Doyon *et al.* (1986), ³Nordstrom and Ousterhout (1982), ⁴Harms *et al.* (1990) ⁵Roush (1981), ⁶Um and Paik (1999)

Table 3: Body weight, feed intake and feed efficiency of laying hens

Parameters	DCL ¹ in diet (g/kg)				SEM ²	P
	0	5	10	15		
Initial body weight (g)	1373.85	1377.70	1376.15	1371.15	8.71	ns
Final body weight (g)	1752.59	1753.29	1756.64	1815.90	15.43	ns
Body weight gain (g)	369.46	376.38	349.90	444.75	17.18	ns
Viability (%)	85.00	85.00	86.67	100.00	5.12	ns
Feed intake (g/hen/day)						
20–24 week	84.99	88.42	88.64	89.44	2.18	ns
24–28 week	129.61a	99.28b	103.55b	116.95ab	3.94	**
28–32 week	132.79	141.45	144.08	140.58	2.06	ns
Overall	114.52	108.43	108.34	115.66	2.21	ns
Feed intake (g/hen/12weeks)	9619.48	9108.03	9100.59	9715.05	185.34	ns
Feed efficiency (g feed/g egg)						
20–24 week	6.62	7.82	5.79	5.81	0.69	ns
24–28 week	3.56	3.42	3.00	2.93	0.19	ns
28–32 week	2.41	2.68	2.72	2.79	0.07	ns
Overall	3.56	3.79	3.69	3.53	0.10	ns
Feed efficiency (g/feed/dozen egg)						
20–24 week	7.96	8.18	6.66	6.91	0.65	ns
24–28 week	2.35	2.16	1.94	1.95	0.12	ns
28–32 week	1.70	1.93	1.98	2.07	0.06	ns
Overall	2.42	2.56	2.49	2.46	0.07	ns

¹ Dry caper leaf powder; ²Standard error of the mean; ** P<0.01; ns: not significant
Means within a row followed by the different superscripts differ significantly

individually. Egg specific gravities were determined by using graded salt solutions ranging from 1.069 to 1.099 with gradations of 0.003 described by Hamilton (1982). After that, shell breaking strength was measured using shell strength device with a spiral pressure system (Fujihara, Saitama, Japan). Subsequently, the eggs were broken one by one on a glass plate with a waiting period of 5 min to measure the albumen and yolk heights using a tripod micrometer, the long and short diameters of albumen, and diameter of yolk were measured using the digital caliper in sensitivity of 0.001 mm. The formulas of some egg quality traits are presented in Table 2. Shell thickness was recorded as an average of three measurements taken at the equator, blunt edge and pointed edge of the egg without membrane using the caliper.

Dry caper leaf powder antioxidant activity and total phenolics assay: Total antioxidant activity was investigated using Ferric Reducing Antioxidant Power (FRAP) assay according to the method described by Oyaizu (1986). Antioxidant capacity assay was carried out using a Perkin Elmer Lambda 35 UV/Vis spectrophotometer by the improved ABTS•+ method according to the procedure described by Re *et al.* (1999). A total phenolic constituent of DCL was performed employing the literature methods involving Folin–Ciocalteu reagent and gallic acid as standard (Slinkard and Singleton, 1977).

Statistical analyses: The data were analyzed by SPSS 16.0 software for Windows (Inc. Chicago, IL. USA). The differences between groups were determined by one-way

Table 4: Effects of dietary DCL levels on egg productivity performance traits of laying hens

Parameters	DCL ¹ in diet (g/kg)				SEM ²	P
	0	5	10	15		
Age at point-of-lay (day)	157.00	159.00	157.00	157.00	0.33	ns
Egg production age (day)						
10%	157.00	158.00	157.00	157.00	0.25	ns
20%	157.00	159.00	157.00	157.00	0.50	ns
30%	157.00	160.00	157.50	158.00	0.61	ns
50%	169.50	161.00	160.00	161.50	2.55	ns
First egg weight (g)	47.99	45.61	48.75	42.99	0.99	ns
Egg weight (g)						
20–24 week	47.66b	48.61ab	50.12a	50.10a	0.38	*
24–28 week	54.99	52.71	54.09	55.51	0.42	ns
28–32 week	58.80b	59.96ab	60.90a	61.67a	0.40	*
Overall	56.34	56.21	56.45	57.97	0.35	ns
Egg mass (g/day/hen)						
20–24 week	18.33	17.66	21.55	20.6	1.48	ns
24–28 week	40.09	29.36	34.93	40.08	2.03	ns
28–32 week	55.09	52.78	53.00	51.13	0.97	ns
Overall	39.05	36.50	38.42	41.50	1.19	ns
Oviposition time						
Morning egg weight (g)	57.32	57.53	58.28	59.50	0.41	ns
Afternoon egg weight (g)	53.04	53.91	52.31	54.21	0.41	ns
Hen–house egg production (%)						
20–24 week	36.82	36.36	42.27	41.36	2.95	ns
24–28 week	60.71	53.93	59.64	72.32	3.48	ns
28–32 week	79.11	78.93	63.04	83.04	5.14	ns
Overall	64.48	61.49	58.21	71.72	3.24	ns
Hen–day egg production (%)						
20–24 week	38.49	36.36	42.27	41.36	2.95	ns
24–28 week	73.14	55.63	64.43	72.32	3.56	ns
28–32 week	93.82	88.02	87.09	83.04	1.98	ns
Overall	69.52	64.97	68.02	71.72	2.17	ns
Morning hen–day egg yield (%)	53.82	41.77	47.64	50.75	2.60	ns
Afternoon hen–day egg yield (%)	15.70	23.19	20.38	20.97	1.14	ns

¹Dry caper leaf powder; ²Standard error of the mean; * P<0.05; ns: not significant

Means within a row followed by the different superscripts differ significantly

ANOVA test. All values were presented as arithmetic means and standard error of the mean; the levels of significantly different was set at P<0.05.

RESULTS AND DISCUSSION

Two methods were used in assessing the antioxidant activities in vitro because a single antioxidant activity test would present limited information about the power of the antioxidant in question. Both TEAC and FRAP assay have been proved as a good parameters for the determination of antioxidant quality of dry *Capparis spinosa* leaf powder. The total phenolic content, TEAC and FRAP value of DCL were 2.78±0.050 g gallic acid equivalents (GAE)/kg, 51.06±2.300 mmol trolox/kg and 50.10±3.488 mmol TEAC/kg respectively.

The productivity performance traits of treatment groups are briefly described in Table 3 and 4. Supplementation of diets with DCL had no significant effects

on body weight, body weight gain, viability, total feed intake, overall feed intake and feed efficiency when compared with the control diet (P>0.05). On the other hand, contrary to expectations, there were significant differences in terms of ES in seven week among the groups (P<0.05). As a result of a random selection of eggs, this difference might be due to the selection of low egg weight in 5 g DCL/kg group. In addition, these contradictions in the findings might have been due to differences in the experimental approach.

All parameters used for evaluation of external and internal egg quality are shown in Table 5. The result demonstrated that levels of DCL did not affect overall SI, specific gravity, shell breaking strength, egg shell thickness, albumen pH, AI, YI, ES, SW and HU (P>0.05).

The results from this study indicated that the *Capparis spinosa* leaf powder possess a strong antioxidant/free radical scavenging effectiveness and ferric reducing

Table 5: Effects of dietary DCL levels on means of external and internal egg quality characteristics during 12 weeks the laying period

Parameters	DCL ¹ (g/kg)				SEM ²	P
	0	5	10	15		
Shape index (SI)	76.25	76.49	76.17	75.38	0.22	ns
Specific gravity (g/cm ³)	1.091	1.091	1.091	1.091	0.000	ns
Breaking strength (kg/cm ²)	2.51	2.20	2.22	2.46	0.08	ns
Egg shell thickness (µm)	351.3	349.6	350.2	346.7	2.80	ns
Egg surface area (cm ²)	66.98	66.22	66.16	67.39	0.38	ns
Albumen pH	8.69	8.70	8.69	8.67	0.01	ns
Albumen index	12.35	13.15	12.60	13.52	0.41	ns
Yolk index	45.78	45.76	46.14	45.88	0.23	ns
Shell weight (g)	6.10	5.97	6.03	6.19	0.06	ns
Haugh unit (score)	96.43	96.37	96.76	97.82	0.71	ns

¹Dry caper leaf powder; ²Standard error of the mean; ns: not significant

abilities, which is probably due to the presence of polyphenolic compounds. *Capparis spinosa* dry leaf powder might be therefore a good candidate for functional animal foods as well as plant-based pharmaceutical products. Moreover when consumed as plant leaf, DCL may contribute to total intake of dietary antioxidants.

In the current literature, the amount of information regarding the effect of adding DCL to poultry diet has been limited, and there have been insufficient study regarding the effects of dietary DCL in laying hens.

The results of productivity performance implied that nutrient composition of diets was same. It indicated that DCL could be used as an ingredient up to 15 g/kg in a diet without any adverse effect on production performance in laying hens. On the other hand, a positive correlation of feed intake with feed efficiency and EM were noticed by Sterling *et al.* (2003). It might be understood clearly that when overall feed intake was evaluated together with overall egg production, egg weight and EM, overall feed efficiency did not undergo significant changes among the groups. Hens consuming the 5 g and 10 g DCL/kg diets exhibited a significant depression in feed intake during 24-28 weeks, but this did not influence hen-day egg production or egg weights. Given that higher levels of DCL supplement did not yield similar depressions, it could be inferred that the effect was transient and likely not specifically related to the dietary treatment. However feed consumption is positively related to egg production proved Jones *et al.* (2000) and Saleh (2013).

Although feed intake had the lack of significant treatment between treatments in 20-24 and 28-32 weeks, dietary DCL supplementation progressively increased average daily egg weight for 10 and 15 g/kg supplementation levels at 20-24 and 28-32 weeks period when compared to the standard commercial diet. The increase in egg weight could be explained with an appetizing effect of caper due to numerically increment in daily feed intake values.

Nevertheless, this increment was not reflected significantly in the overall period. In that regard, it could be hypothesized that the caper containing biologically active substances may increase weight of the egg or a potential limitation of the current study relates to the sample size used in relation to the variability observed in the production data. The current study contributes to this limited database by providing new information on the responses of laying hens consuming diets containing DCL. It is clear from Table 3 that no differences in age at point-of-lay, age at 10, 20, 30 and 50% egg production, first egg weight, EM, egg weight according to oviposition time, HHP and HDP were observed in laying hens fed the different experimental diets ($P > 0.05$). The lower egg production and EM between 20-24 weeks might be related to the all hens unexpectedly lay the 22-23 weeks. In general, all parameters was similar in the graded DCL supplemented group compared with the control group, except the egg production with 15 g DCL/kg group is higher than the control, although the augmentation did not reach statistical significant.

Egg weight and eggshell quality characteristics vary according to the oviposition time. These results are consistent with those of several previous studies indicated that eggs laid early in the morning were heavier than eggs laid during the later periods of the day (Tumová and Ebeid, 2005; Tumová *et al.*, 2007). Egg weight was greater in afternoon eggs in comparison with eggs laid in the morning. Similarly, it could be also indicative of the fact that morning HDP values were higher than that of afternoon HDP in all groups.

In the current study, specific gravity data correspond with the findings of Lim *et al.* (2003), who determined the medium specific gravity ranged from 1.085 to 1.095 in 21-wk-old ISA brown laying hens.

Albumen quality is an important indicator of egg freshness and also significant for the egg processing

industry (Jin *et al.*, 2011). Silver sidesand Villeneuve (1994) claimed that pH is a useful means for describing changes in albumen quality over time during storage. However, albumen pH is not affected by DCL groups and can be used to measure egg freshness. Overall albumen pH is ranged from 8.56 to 8.84 which is in agreement with previous data reported for ATAK-S laying hens by Sekeroglu *et al.* (2010) and Yildirim *et al.* (2013; 2014).

Hens fed with diet supplemented with DCL had numerically highest values of HU score, but lowest values of shell breaking strength compared to control group ($P>0.05$). Egg thickness, SW, AI and YI parameters were similar among all groups between 20 to 32 weeks of age. Overall, the data from the current study supported the contention that laying hen diets can contain up to 15 g DCL/kg without affecting measures of hen productivity or specific measures of egg and eggshell quality.

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CONCLUSION

Results of this study showed that *Capparis spinosa* with total phenolic compounds confirms its nutritional and medicinal value and it could be used as a potential natural antioxidant source in commercial egg production. Overall dietary supplementation of DCL to the laying hens' diets had no adverse effects on laying performance, health status of the hens, external and internal egg quality measurements. Hence, the studies on the supplementation of DCL in poultry diet have been still limited and current findings could be considered as the first report presenting the effect of DCL on egg production, egg quality of laying hens of local breed.

ACKNOWLEDGEMENT

The authors are thankful to the staff for their technical support during the course of experiment from Department of Animal Science, Faculty of Agriculture, University of Gaziosmanpasa, Tokat, Turkey.

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