Effect of a dietary essential oil mixture on performance of laying hens in the summer season

M. Çabuk[1#], M. Bozkurt[2], A. Alçiçek[3], A.U. Çatlı[2] and K.H.C. Başer[4]
[1] Department of Poultry Science, Vocational School of Celal Bayar University, Akhisar, Manisa-Turkey
[2] Poultry Research Institute, Erbeyli, Aydın-Turkey
[3] Department of Animal Science, Agricultural Faculty of Ege University, Bornova, Izmir-Turkey
[4] Anadolu University, Faculty of Pharmacy, Department of Pharmacognosy, 26470 Eskişehir-Turkey

Abstract
The experiment was conducted to evaluate the effects of dietary supplements of either an essential oil mixture (EOM) or a mannan oligosaccharide (MOS) as alternatives to an antibiotic feed additive (avilamycin) for layers kept under hot summer conditions. Four hundred and eighty 54-week old Nick-Brown hens were assigned to four dietary treatments. Each treatment consisted of four replications of 10 cages (three hens per cage). The treatment groups were: 1) Control: Basal diet without additive; 2) Basal diet plus antibiotic (10 mg avilamycin/kg feed); 3) Basal diet plus 1 g MOS/kg feed; 4) Basal diet plus 24 mg EOM/kg feed. Performance of laying hens was affected by dietary treatments. Dietary supplementation of EOM and MOS significantly increased egg production compared with control and antibiotic groups. There were no significant differences in feed consumption between treatments. The EOM significantly improved feed conversion ratio above that of the control group. Egg weights were significantly different between treatments. Laying hens consuming MOS produced significantly lower egg weights than the other groups, while egg weights in the EOM, antibiotic and control groups did not differ significantly. Cracked-broken egg rate was decreased by dietary addition of EOM, MOS and antibiotic compared with the control. Number of deaths among hens was significantly affected by dietary treatments with the number of deaths in the MOS treatment being significantly lower than in the other treatments. The performance of laying hens during the summer season could be maintained with inclusions of EOM and MOS in the diet.

Keywords: Essential oils, mannan oligosaccharide, antibiotic, egg production, laying hen, summer season
[#] Corresponding author. E-mail: metin.cabuk@deu.edu.tr

Introduction
It is generally accepted that high environmental temperatures affect poultry performance negatively in terms of feed intake, live weight gain, feed efficiency (Donkoh, 1989; Siegel, 1995), egg weight (Peguri & Coon, 1991) and eggshell quality (Deaton *et al*., 1981; Grizzle *et al*., 1992). Furthermore, decreased eggshell thickness was observed by Ahvar *et al.* (1981), with a resultant poorer eggshell quality and a reduced shelf life. Therefore, egg production and eggshell quality of laying hens often deteriorate in hot climatic regions, such as in Turkey. Under these conditions heat stress is particularly a problem when hens are kept in conventional, naturally ventilated houses, which have proven to be ineffective in many regions of Turkey. Because of the high cost of cooling animal buildings, dietary manipulation is normally applied to alleviate the negative effects of high environmental temperature on performance of laying hens. For the past four decades, antibiotics have been used as feed additives to improve growth and egg performance, and to protect animals from pathogenic microorganisms. The general ban of the use of feed antibiotics as microbial performance promoter that is expected to be introduced more widely throughout the world in coming years, has increased the urgency for research on botanical feed additives (Best, 2000). Plant extracts and spices as single compounds or as mixed preparations can play a role in supporting both performance and health status of animals (Bakhiet & Adam, 1995; Gill, 1999; Alçiçek *et al*., 2003; 2004; Çabuk *et al*., 2006). Beneficial effects of plant extracts in animal nutrition include the stimulation of appetite and feed intake, the improvement of endogenous digestive enzyme secretion and the activation of immune responses and antioxidant actions (Baratta *et al*., 1998; Jamroz *et al*., 2003; 2005; Botsoglou *et al*., 2005). Some herbs also have anti-heat stress effects (Liu-Fengtlua *et al*., 1998). Dietary supplementation with mannan oligosaccharides (MOS) offers another alternative to antibiotic growth promoters. Mannan oligosaccharides,

derived from yeast cell walls, are indigestible in monogastric animals and can be utilized by lactic acid bacteria. Mannan oligosaccharides also bind the fimbria of pathogenic bacteria to prevent them from attaching to and colonizing on the small intestinal mucosa. Adhered bacteria are subsequently washed out of the small intestine with the flow of intestinal content (Ofek *et al.*, 1977). The effects of MOS on broiler performance were studied by Santin *et al.* (2001), Hofacre *et al.* (2003) and Waldroup *et al.* (2003). However, the effects of essential oils and MOS on performance of laying hens have not been studied extensively, particularly not under hot climatic conditions.

Therefore, this experiment was conducted to evaluate the effects of dietary supplements of either an essential oil mixture (EOM) or MOS as alternatives to an antibiotic feed additive (avilamycin) for layers kept under hot summer conditions. The following response variables were analyzed to investigate treatment effects: egg production, egg weight, egg output, cracked-broken egg rate, feed conversion ratio (FCR), feed consumption, death rate and body weight.

Materials and Methods

Table 1 Ingredients and chemical composition of the basal experimental layer diet (as fed)

	Layer diet (g/kg)
Ingredients	
Maize	580.7
Soyabean meal (46 % CP)	183.4
Sunflower meal	100.0
Meat and bone meal	50.0
Fish oil	9.7
Salt	2.5
Ground limestone	68.1
Dicalcium phosphate	1.1
Vitamin premix*	2.5
Mineral premix**	1.0
DL-methionine	1.0
Total	1000
Composition (analysed)	
Dry matter	887.6
Crude protein	173.9
Ether extract	36.8
Crude fibre	46.5
Crude ash	119.8
Starch	386.6
Sugar	29.8
Calcium	35.3
Total phosphorus	6.7
Metabolisable energy (MJ/kg)(calculated)	11.6
Lysine (calculated)	8.8
Met.+Cys. (calculated)	6.9

*Supplied per kilogram of diet**:** 12 000 IU vitamin A; 2 400 IU vitamin D_3; 30 IU vitamin E; 2.5 mg vitamin K_3; 3 mg vitamin B_1; 7 mg vitamin B_2; 20 mg niacin; 8 mg calcium D-pantothenate; 4 mg vitamin B_6; 0.015 mg vitamin B_{12}; 1 mg folic acid; 0.045 mg D-biotin; 50 mg vitamin C; 125 mg choline chloride
** Supplied per kilogram of diet**:** 80 mg manganese; 80 mg iron; 60 mg zinc; 5 mg copper; 0.2 mg cobalt; 0.5 mg iodine; 0.15 mg selenium

Four hundred and eighty 54-week old Nick-Brown hens were assigned to four dietary treatments. Each treatment consisted of four replications of 10 cages (three hens per cage), amounting to 120 birds per treatment group. The trial was conducted in an open-sided, naturally ventilated layer house during the summer season of 2004, between April and August, in Aydın, western Turkey, situated between 36^{o} and 38^{o} northern latitude and between 26^{o} and 28^{o} eastern longitude. Individual body weights of all hens were taken at the onset and termination of the experiment to determine average body weights and changes in body weight. The ingredients and chemical composition of the basal experimental diet are presented in Table 1.

The experimental diets were as follows: 1) Control: Basal diet without EOM, antibiotic or MOS; 2) Basal diet plus 10 mg avilamycin/kg feed (Kartal Kimya Co,Turkey); 3) Basal diet plus 1 g MOS/kg feed (Bio-Mos, Alltech, Inc., İzmir-Turkey); 4) Basal diet plus 24 mg essential oils/kg feed (Herba Ltd. Co. İzmir-Turkey). The appropriate amount of feed additive (antibiotic, MOS or EOM) for 1000 kg of feed was premixed with a carrier of 1 kg in order to mix the relevant treatment diet. EOM consisted of six different essential oils derived from selected herbs: Oregano oil (*Origanum* sp.), laurel leaf oil (*Laurus nobilis* L.), sage leaf oil (*Salvia triloba* L.), myrtle leaf oil (*Myrtus communis*), fennel seeds oil (*Foeniculum vulgare*), and citrus peel oil (*Citrus* sp.). Diets (in mash form) and water were provided for *ad libitum* consumption. A photoperiod of 17 hours/day was maintained. Egg production and broken-cracked eggs were recorded daily from 54 to 74 weeks of age. During this period random samples of 60 eggs/treatment/day were collected on two consecutive days every week. Hence, 9600 eggs were weighed to determine average egg weight. Feed intake was recorded on a weekly basis. Feed conversion was calculated as the ratio of g of feed consumed per g of egg weight produced. Egg output was calculated by multiplying egg weight by number of eggs. The magnitude of production variables such as feed intake and egg production was adjusted for hen mortalities. Death of hens was recorded daily. Nutrient content of the diets was determined by proximate analysis (Naumann & Bassler, 1993). The experimental diets were analysed for starch, sugar, total Ca and P according to the VDLUFA method (Naumann & Bassler, 1993). Egg weight, egg output, FCR and feed consumption were analyzed with the general linear model procedure of SAS (1987). Significant treatment effects were detected by Duncan's multiple range tests. Egg production, cracked-broken eggs and number of deaths were analyzed by means of the chi-square test. Percentage of egg production, cracked-broken eggs and death rates was also calculated.

Results

The effects of dietary treatment on laying hen performance are shown in Tables 2 and 3. Supplementing MOS and EOM to the basal diet increased ($P < 0.05$) egg production above the control. The addition of EOM and MOS to the basal diet also led to a significantly higher ($P < 0.05$) egg production than the addition of the antibiotic. However, egg production was similar in the EOM and MOS treatments. There were differences between treatments in cracked-broken egg rate ($P < 0.01$). All additive treatments

Table 2 Number of eggs produced, cracked-broken eggs and deaths in laying hens receiving a supplemental antibiotic, a mannan oligosaccharide (MOS) or an essential oil mixture (EOM)

Variables	Control	Antibiotic	MOS	EOM	Expected egg number***	Chi-sq contribution	Chi-sq statistics
Total number of eggs produced, (140 days)	12692 [b]	12945 [b]	13378 [a]	13379 [a]	13099	26.35**	11.34
Egg production, %	75.5	77.1	78.9	79.6	-	-	-
Number of cracked-broken eggs, (140 days)	457 [a]	298 [c]	348 [b]	386 [b]	372	36.17**	11.34
Cracked-broken eggs, % of total number of eggs	3.6	2.3	2.6	2.9	-	-	-
Number of deaths	9 [a]	7 [a]	1 [b]	3 [a]	5	8.0*	7.81
Death rate, %	7.49	5.83	0.83	2.50	-	-	-

[a-c] Means within rows with different superscripts differ at $P < 0.05$
* $P < 0.05$; ** $P < 0.01$
*** Average egg production of hens in the four treatment groups at 140 days

significantly reduced the incidence of broken-cracked eggs compared with the control. Antibiotic treated hens produced fewer ($P < 0.05$) broken-cracked eggs than the EOM and MOS treated hens. The number of broken-cracked eggs did not differ between EOM and MOS treated hens. MOS treatment significantly reduced death rate compared with the control, EOM and antibiotic treatments. The death rate of laying hens did not differ in the EOM, antibiotic and control treatments.

Addition of EOM, MOS and antibiotic to the layer diet did not affect the daily feed intake of the laying hens significantly. However, in terms of FCR there were significant differences between the treatment groups (Table 3). EOM was the only additive to significantly improve feed efficiency compared to the control, but there were no significant differences in feed efficiency between the EOM, MOS and antibiotic treatments. Egg weight produced, differed ($P < 0.01$) between treatments. The supplementation of MOS to the diet significantly decreased egg weight compared to other treatments (EOM, antibiotic, and control), which did not differ among themselves. On the other hand, egg output was not affected by treatments ($P > 0.05$), although there was a tendency towards higher egg outputs in all three additive treatments.

Table 3 Feed intake, feed conversion ratio, egg weight and egg output in laying hens receiving a supplemental antibiotic, a mannan oligosaccharide (MOS) or an essential oil mixture (EOM)

Variables	Control	Antibiotic	MOS	EOM	Pooled s.e.m.	Probability
Feed intake, g/hen/day	103.9	103.9	102.9	104.5	0.61	0.3377
Feed conversion ratio, g feed/g egg	2.09^{a}	2.05^{ab}	2.01^{ab}	1.99^{b}	0.02	0.0021
Egg weight, g	65.6^{a}	65.7^{a}	65.0^{b}	65.5^{a}	0.15	0.0001
Egg output, g	49.5	50.6	51.3	52.1	0.85	0.3212

$^{a-b}$ Means within rows with different superscripts differ at $P < 0.05$

Results on initial and final body weights are shown in Table 4. Initial body weights of hens at 54 wk of age did not differ between treatments ($P > 0.05$). Body weight loss of the hens in all treatments commenced by 64 wk of age, probably reflecting deleterious effects of the high environment temperature during trials. However, there were no significant differences in body weight of the hens at 64 wk of age. The final body weights of the hens differed significantly between treatments. At 74 wk antibiotic, MOS and EOM treated hens had higher ($P > 0.05$) final body weights than the hens in the control.

High temperatures were observed during this experiment (Table 5), particularly in July and August (Tables 2 and 4).

Table 4 Body weight of the laying hens receiving a supplemental antibiotic, a mannan oligosaccharides (MOS) or an essential oil mixture (EOM)

Parameters	Control	Antibiotic	MOS	EOM	Pooled s.e.m.	Probability
Body weight, g (54 wk of age)	1926	1930	1913	1930	12.6	0.7398
Body weight, g (64 wk of age)	1884	1882	1863	1871	13.9	0.6877
Body weight, g (74 wk of age)	1601^{b}	1649^{a}	1669^{a}	1653^{a}	16.81	0.0263

$^{a-b}$ Means within rows with different superscripts differ at $P < 0.05$

Table 5 Mean house temperatures (°C) during study

Months of the experiment	Time of day			
	09:00	13:30	16:00	Mean
April	16.7	20.4	20.3	19.13
May	26.3	34.2	35.2	31.90
June	27.2	35.1	36.0	32.77
July	29.4	36.2	36.8	34.13
August	29.1	36.6	37.8	34.50

Discussion

Supplementation of EOM and MOS to the basal diet significantly increased egg production. Dietary supplementation with EOM also improved ($P < 0.01$) FCR. A similar result was observed by Ather (2000), who reported that the addition of essential oils of a polyherbal feed additive to a broiler breeder diet showed better average egg production compared to their control. A recent study by Deying *et al.* (2005) found that a diet supplemented with herbal medicine (*Ligustrum lucidum* and *Schisandra chinensis*) significantly improved egg production and FCR of laying hens. In addition, Chukwu & Stanley (1997) observed that the addition of MOS to the diet significantly improved egg production and FCR in laying hens kept at ambient temperatures ranging from 23 °C to 32 °C. High environmental temperatures may create opportunities for infectious agents to overwhelm the immune system and exaggerate negative attributes of heat stress (Deying *et al.*, 2005). Eventually, nutrient utilization, feed digestibility and energetic efficiency deteriorated due to unbalanced intestinal microflora (Zuprizal *et al.*, 1993; Bonnet *et al.*, 1997). Jamroz *et al.* (2005) also demonstrated increased pancreatic and intestinal lipase activity in broilers fed diets with added plant extract. In contrast to our result, Botsoglou *et al.* (2005) found that the addition of rosemary, oregano and saffron to a layer diet had no significant effect on egg production, feed intake and FCR. Supplementation of EOM and MOS to the layer diet decreased the number of cracked-broken eggs significantly. Hence, adding EOM and MOS to layer diets may be a practical way of decreasing cracked-broken egg rates in terms of commercial feed additive. Contrasting results have been reported by Deying *et al.* (2005) who reported that a diet supplemented with herbal medicine had no beneficial effect on cracked-broken egg rate.

Supplementation of MOS to the diet decreased the egg weight significantly compared to the other treatments. On the other hand, birds consuming EOM, antibiotic and control diets had similar egg weights. Contrary to our results, Bassei (1994) and Jamroz *et al.* (1998) observed that the supplementation of an antibiotic to a layer diet increased egg weight. Although death rate of hens in the present study tended to be lower in all three additive groups compared to the control, it was only reduced significantly by MOS supplementation compared to the other treatments. Decreased body weight and increased death rate of the control hens appeared to be related to high ambient temperatures. A similar effect on death rate of broilers was observed by Waldroup *et al*. (2003) who compared MOS and antibiotic feeding programs in the summer months of June to August.

The results of the present study support the beneficial effect of MOS and EOM in layer diets by reducing the negative effects of high ambient temperatures. Thus, the performance of laying hens during the summer season can be maintained with dietary inclusions of EOM and MOS.

Conclusions

The observed results in the summer season indicated that the inclusion of 24 mg EOM/kg in a layer diet significantly improved egg production, FCR and reduced cracked-broken egg ratio of laying hens over a period of 20 weeks. The supplementation of 1 g MOS/kg to the layer diet also had beneficial effects on egg production, cracked-broken egg ratio and death rate. The EOM and MOS could be considered as potential natural feed additives for laying hens, because these additives meet the needs of producers for increased layer performance and the consumers' demands for environmentally friendly egg production. However, further and detailed evaluations are required to establish the effect of essential oils and MOS in diets on the performance of laying hens during the hot summer season.

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