

The effect of feeding stearidonic acid enriched soya oil to broilers on the fatty acid composition and sensory characteristics of chicken meat

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Introduction Enriching chicken meat with long chain n-3 polyunsaturated fatty acids (LC n-3 PUFA) is a viable means of increasing population intakes (minimum recommended 450 mg/d, SACN, 2004) of these essential fatty acids. Feeding broilers fish oil to achieve this causes problems with taint in the meat, whereas feeding an oil rich in C18:3 n-3 (LNA) does not result in the deposition of LC n-3 PUFA (Rymer and Givens, 2006). Stearidonic acid (C18:4n-3) is further down the conversion pathway of LNA to LC n-3 PUFA and appears to be converted much more efficiently by humans to LC n-3 PUFA than LNA (James *et al.*, 2003). Feeding birds an oil rich in C18:4n-3 may therefore result in more enrichment of poultry meat with n-3 PUFA other than LNA, without the taint associated with feeding fish oil. The objective of this experiment was to determine what the effects on meat fatty acid composition and sensory characteristics were when broilers were fed an oil (SDA) derived from soyabean genetically modified to produce high concentrations of C18:4 n-3.

Methods Male, day old Ross 308 chicks (120) were reared for 14 d in a single group on a common starter diet containing conventional soyabean oil (CON). At 15 d, the birds were weighed and randomly allocated to one of 24 pens (five birds per pen). Pens were blocked in groups of three, and within each block, pens were randomly allocated to one of three treatment groups (eight replicate pens per treatment). Birds were fed identical grower (days 15-28) and finisher (29-50 d) diets supplemented with either CON oil, fish oil (FISH) or SDA. Diets were based on wheat, soyabean meal, sunflower seed meal and maize gluten meal; crude protein contents were 261, 275 and 230 g/kg and AME contents 12.2, 13.2 and 13.1 MJ/kg in the starter, grower and finisher phases respectively. Birds were weighed at 41d and sacrificed in blocks between 41 and 50d. Samples of skinless breast and leg meat were taken from all birds, composited by pen for analysis of fatty acid composition and composited by treatment for sensory analysis (quantitative descriptive analysis with ten assessors scoring meat between 0 and 100 for a range of descriptors), using either freshly cooked meat (breast and leg meat) or reheated meat (leg meat only). The effect of broiler diet on the meat's fatty acid composition and sensory characteristics were determined by analysis of variance.

Results There was no significant effect of treatment on bird performance between 15 and 41 d. FISH reduced LNA and increased LC n-3 PUFA content of meat (Table 1). SDA increased LC n-3 PUFA content compared with CON and total n-3 content (after deduction of LNA, to give an estimate of potential LC n-3 PUFA supply) was considerably higher with both FISH and SDA compared with CON. Broiler diet had no significant effect on the meat's appearance, or on the aroma or flavour of breast meat. However, fishy aromas, flavours and aftertastes were detected in the leg meat of FISH fed birds, particularly when the meat was reheated (Table 2). Significant fishy notes were also detected in the reheated leg meat of birds fed SDA.

Table 1 Effect of broiler diet on the meat n-3 fatty acid composition (mg/100 g tissue)

Fatty acid	Skinless breast meat			SEM	Sig	Skinless leg meat			SEM	Sig
	SDA	CON	FISH			SDA	CON	FISH		
LNA	116	80	31	9.8	**	231 ^a	175 ^a	89 ^b	21.2	*
LC n-3 PUFA	87 ^b	31 ^a	209 ^c	15.0	*	141 ^b	29 ^a	430 ^c	29.9	*
Tot. n-3 less LNA	318 ^b	34 ^a	222 ^b	26.1	*	583 ^b	39 ^a	466 ^b	47.3	*

*: P<0.05, **: P<0.01. ^{a,b} Mean values within a row without a common superscript differ significantly (P<0.05)

Table 2 Effect of broiler diet on the sensory scores (0-100) of leg meat

Fishy attribute	Freshly cooked meat			Reheated meat			LSD
	SDA	CON	FISH	SDA	CON	FISH	
Aroma	18.3 ^{ab}	12.7 ^a	29.1 ^c	23.6 ^{bc}	9.1 ^a	51.5 ^d	10.7
Flavour	20.1 ^{abc}	11.8 ^{ab}	23.0 ^{bc}	29.0 ^a	10.7 ^c	51.3 ^d	11.2
Aftertaste	14.5 ^{abc}	12.1 ^{ab}	22.5 ^c	18.8 ^{bc}	9.2 ^a	45.9 ^d	9.4

^{a,b,c,d} Mean values within a row without a common superscript differ significantly (P<0.0001)

Conclusions Feeding broilers SDA produced meat with nutritionally significant concentrations of LC n-3 PUFA or its immediate precursors (recommended intake of LC n-3 PUFA 450 mg/d, SACN, 2004) while reducing the taint detected when birds were fed fish oil.

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Bayesian estimation of posterior means of heritability for weight at week 8 in Iranian indigenous chickens using an animal model

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Introduction Indigenous poultry breeds are of a major importance to supply the rural population with eggs and meat in many developing countries of the world (Esmaelkhanian *et al.*, 2004). A successful animal breeding plan is a continuous process of elimination and searching in a way that breeder could be able to manipulate genetic variation for changing a population to optimise desired phenotypes (Williams *et al.*, 2002). As highlighted by Chambers (1990) animal's body weight, which could be easily measured, is probably the most frequently used indicator of growth at a specific age. In practical animal breeding programmes, the breeding objective is usually genetic improvement of production and reproduction traits for a population under consideration. Knowledge of genetic parameters is of crucial importance to be implemented for predicting breeding value of candidate animals. The main objective of this study was to estimate heritability of body weight at week 8 for Iranian indigenous chickens through Bayesian statistical approach using an animal model.

Materials and methods In this study a total of 6440 weight records at week 8 belonging to Iranian indigenous chickens were used to obtain Bayesian estimation of posterior means of heritability. All records were collected in a centre of indigenous chicken breeding located in Khorasan province, Mashhad. In the data file, the average weight at week 8 was 571.46 g. An animal linear model was applied for genetic analysis. In the model, the fixed environmental effects of sex and hatch number (4 hatches) were included. Furthermore, the weight of chicks (w) at day one (average 35.03 g) was fitted in the model as linear covariate. Random additive genetic effect of chicks (animal) was also put in the model. All data were from the first of generation. The number of chicks with records, sires and dams were 6440, 81 and 747 respectively. A Bayesian estimation approach was used to obtain posterior means of additive genetic as well as environmental variance components for weight at week 8. The variance components were estimated implementing MTGSAM software (Van Tassell and Van Vleck, 1995) through carrying out 100,000 Gibbs sampling in which the first 5,000 rounds were assumed as burning in period. The convergence criterion was set to be 0.0001. The statistical linear model was as follows:

$$y_{ijk} = \mu + sex_i + hatch_j + b * w_{ijk} + animal_k + e_{ijk}$$

Results Estimated additive genetic and environmental variance components were presented in Table 1. The posterior means of additive genetic ($\sigma_A^2 = 2331$) and environmental ($\sigma_E^2 = 3565$) variance components were 2331 and 3565 respectively. Posterior means of heritability of weight at week 8 was found to be approximately 0.4 which is in the range of body weight heritability estimates reported by (Chambers, 1990).

Table 1 Bayesian estimates of additive genetic, environmental and phenotypic variance components for weight at 56 d

Trait	No. Records	Variance components (g ²)			Heritability
Weight at 56 d	6440	$\sigma_A^2 = 2331$	$\sigma_E^2 = 3565$	$\sigma_P^2 = 5896$	0.4

Conclusion Heritability is an important elemental key to make genetic progress in a population. As the heritability of a trait increases the magnitude of selection response increases over a generation. The relatively high estimate of heritability obtained for weight at 56 d for Iranian indigenous chickens indicates that there is a great genetic potential for the population to be improved based upon genetically selecting individuals with high weight performance. However, as many traits are simultaneously considered in poultry breeding, multi traits animal models are needed to be used for obtaining an accurate estimate of genetic parameters.

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Effects of chronic progesterone injection on performance, plasma hormones and ovarian morphology of feed-satiated and fed restricted broiler breeder hens

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Introduction Single injection of chronic dose of P₄ in laying hens during the preovulatory open period has been shown to have positive effects on inducing a preovulatory LH surge and ovulation (Johnson *et al.*, 1985). However, chronic injection of P₄ has been shown to increase baseline concentrations of P₄ and result in arrested laying and disrupted distribution of hierarchical follicles in turkeys (Bacon and Liu, 2004). The current study was designed to examine the effects of a simulated progesterone surge (by injection of chronic dose of P₄) on broiler breeder hens performance, ovary morphology, plasma metabolites and hormones concentrations in either feed-satiated or feed-restricted hens.

Materials and methods A total of 64 Cobb 500 hens were fed either restricted or *ad libitum* from 27 to 38 wk of age. Fourteen laying hens from each group were selected to conduct P₄ injection assay. Half of the birds in each group (n=7) were injected daily with 2.5 mg P₄/kg BW and the remaining birds were used as control. The P₄ injections were given subcutaneously, at the base of the neck, daily (at 0900 h) for 21 d. Settable and abnormal eggs were recorded daily. Blood sample were taken just before initiation of injections, 10-d and 20-d after initiation of injection. Plasma samples were analyzed for glucose, cholesterol, triacylglycerol (TAG), P₄, estradiol (E₂), testosterone, T₃ and T₄ concentration. Ovulation rate was calculated as (normal eggs + soft shell eggs + 2×double yolk eggs). The experimental birds were housed in individual cages. The experimental design was completely randomized with factorial arrangement using individual broiler breeder hens as experimental unit. Performance data as well as plasma metabolites and hormones data were analyzed as repeated measures using Proc Mixed of SAS software (SAS Institute, Cary, NC).

Results and discussion Although egg production declined following P₄ injection (Fig.1), ovulation rate remained at high levels in injected birds in the first week after initiation of P₄ injection (Fig 2). Progesterone injection in feed-satiated and feed-restricted birds resulted in ovary regression; the ovary of these birds having no hierarchical follicles. Progesterone injection increased incidence of holding hard-shelled eggs in the uterus. Progesterone injection depressed plasma E₂ concentration (Fig 3) in both *ad libitum* and restricted fed hens. Hens with free access to feed had significantly lower plasma E₂ levels compared to restricted fed hens. Our results revealed that whereas injecting chronic dose of P₄ induced frequent ovulation early in the injection period in both feed- satiated and feed-restricted breeder hens; however this higher ovulation rate did not result in more settable egg production. Restricted fed and *ad libitum* fed laying breeder hens respond in a similar way to chronic dose of P₄ injection.

Figure 1 Settable egg production (%) of hens

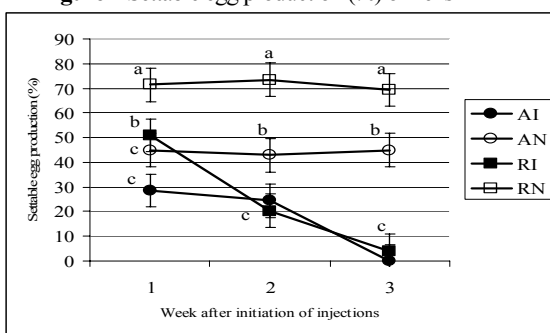


Figure 2 Ovulation rate (%) of hens

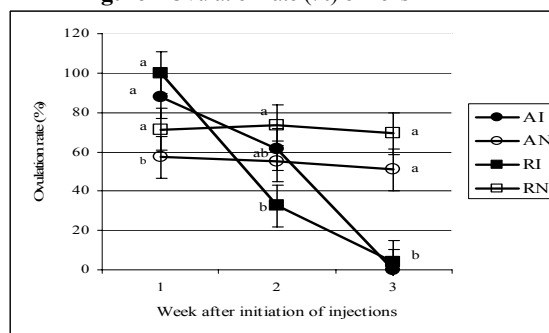
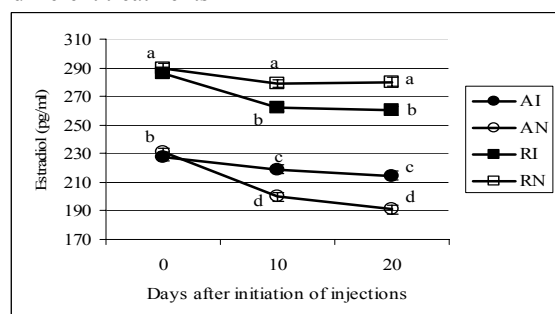


Figure 3 Plasma estradiol concentration of hens from different treatments



A: *ad libitum*, R: Restricted, I: Injected, N: Non-injected.
Data are means ±SEM. a-c Data points with different letter are significantly different at the age of the hens (P<0.05).

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Effect of medicinal plants and organic acid on growth performance of Ross broilers

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Introduction The prophylactic use of antibiotic (as growth promoters) in animal feeds has made intensive farming possible and improved feed conversion. In the presence of low levels of an antibiotic, resistant cells survive and grow which produces an antibiotic-resistant population. Consequently, the use of antibiotic for broilers has been limited. Therefore, a number of studies on alternative products that can aid promotion of growth, improved feed utilization, and maintenance of gut health have taken place (Hernandez *et al.*, 2004). Herbs and organic acid have received attention as an alternative to antibiotics (Craig, 1999; Ricke, 2003). The main aim of the present research was to evaluate the efficiency of alternative antibiotic growth promoters on Ross broiler performance.

Materials and methods A total of 240 Ross male broiler chicks were fed in a completely randomized design with 4 replicate pens (15 birds per pen). Four dietary treatments used: T₁= Control diet based corn and soybean meal without supplementation, T₂= Control diet+ antibiotic (Virginiamycin) 15 ppm, T₃= Control diet+ medicinal plants (commercial mixture of medicinal plants, Digestrom) 450g/ton diet T₄= Control diet+ organic acid (commercial mixture of propionate, Formycine) 450g/ton diet. The broilers were raised on a battery house under commercial condition and offered ad libitum feed and water throughout the 42 d study. The initial room temperature was set at approximately 32°C and reduced by 2 to 3°C weekly. The performance traits were body weight, average daily weight gain, average daily feed intake, feed to gain ration (or feed conversion ratio). The traits were measured for on individual chicks in the experiments. The data were analysed by a general linear model (GLM) by SAS programme. Multiple comparisons among different treatments were undertaken by Duncan multiple range test.

Results The results of analysis of variance for growth performance traits are shown in Table 1.

Table 1 Pair-wise comparisons* of different treatments for growth performance traits

Treatment	T ₁	T ₂	T ₃	T ₄	± SEM
Body weight, (g)					
21 d	593.99 ^b	697.27 ^a	615.51 ^b	609.89 ^b	10.21
42 d	1824.62 ^c	2045.03 ^a	1994.74 ^{ab}	1959.50 ^b	20.74
Average daily gain, (g)					
1 to 21 d	26.84 ^b	31.15 ^a	27.31 ^b	26.94 ^b	0.46
21 to 42 d	57.83 ^b	64.72 ^a	65.66 ^a	63.78 ^a	0.77
1 to 42 d	42.08 ^c	47.93 ^a	46.48 ^b	45.36 ^b	0.47
Average daily feed intake, (g)					
1 to 21 d	43.76 ^a	43.86 ^a	42.56 ^c	42.95 ^b	0.08
21 to 42 d	120.64	120.36	120.60	120.66	0.19
1 to 42 d	82.20 ^a	82.11 ^a	81.85 ^b	81.80 ^b	0.11
Feed: gain ratio					
1 to 21 d	1.70 ^a	1.42 ^c	1.59 ^b	1.59 ^b	0.02
21 to 42 d	2.10 ^a	1.88 ^b	1.85 ^c	1.91 ^b	0.02
1 to 42 d	1.90 ^a	1.65 ^c	1.75 ^b	1.72 ^b	0.01

*Treatments with similar letters were not statistically different from each other

Conclusions Supplementation with virginiamycin, medicinal plants and organic acid significantly ($p < 0.05$) improved body weight, average daily gain and feed:gain ratio broilers during the first 42 days when compared with treatment 1. The results obtained in the present research also revealed that for the traits of body weight as well as average daily gain and feed intake for the treatments 3 and 4 had lower values than those of the treatment 2. It can be, therefore, suggested that a higher level of medicinal plants and organic acid to be used to detect an accurate influence of the antibiotic alternatives on growth performance of broilers.

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Effects of vitamin E and C supplementation on performance and immune response of broiler chicks

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Introduction Vitamin E (VE) is known for its role as an antioxidant, protecting unsaturated bonds of cellular membrane phospholipids against free radical attack. VE also has been shown to be a requirement for normal development and function of the immune system. Although poultry are renal synthesizers of ascorbic acid, synthesis is inadequate under stressful conditions such as high environmental temperature, high humidity, high egg and meat production rate and parasite infestation. The main objective of this study was to assess the effects of various levels of dietary VE supplementation and Vitamin C (VC) in drinking water on performance, humoral and cell mediated immune response.

Materials and methods Commercial broiler males (Ross 208) were obtained from a local hatchery. One-day-old chicks were weighed and randomly allotted to dietary and water treatments. The basal corn-soybean meal diet was prepared according to NRC recommendations (1994), except that VE was omitted from the vitamin premix. Thirty-six pens of chicks (12 chicks per pen) were assigned to each of nine dietary treatments consisting of standard commercial starter and grower diets supplemented. The experimental design was a 3*3 factorial arrangement of three levels of dietary VE (0, 50, 75, mg dl-a-tocopherol acetate/kg of feed) and three levels of dietary VC (0, 500 and 1000ppm L-Ascorbic Acid in drinking water). At 26 days of age, three birds per pens were randomly assigned for lymphoblastogenesis assay in vitro. Blood samples (3ml/chick) were withdrawn with a EDTA syringe and diluted with equal volume of PBS. Blood from all birds in each pens was pooled and The peripheral blood lymphocytes were separated from the whole blood. Proliferative responses of lymphocytes were expressed as absorbance as described by Maslak and Reynolds (1995).

Results The results of the present study identified no significant differences in body weight gain in chicks fed supplemental VE to 42d. Proliferation of purified lymphocytes in mitogen-free cell culture medium (control) was affected by the level of dietary VE fed to chicks (data not shown). Chicks fed additional VC (1000ppm) exhibited a significant beneficial effect for body weight gain (42d), daily weight gain and feed consumption (21-42d). Mitogenic responses to PHA and Con A were significantly altered by VE. Proliferation of whole blood lymphocytes in the absence of mitogens was not affected with 50 and 75 IU/kg of added VE compared to the control. Chicks drank 500 and 1000 ppm of VC had significantly greater ($P < 0.05$) in vitro lymphocyte proliferative responses to ConA and PHA than those of chicks drank normal water. proliferation with mitogen and control (without mitogen) of whole blood lymphocytes in the mitogens PHA and Con A was not affected with VC. Lymphocyte proliferation in response to mitogens is correlated with the ability of the host to mount a cellular immune response. It has been suggested that differential reactivity to mitogens reflects either maturational or functional differences in the responsive lymphocytes. It was shown that many factors, including cell preparation, cell culture medium, and duration of the assay, might influence the effects of dietary factors on in vitro responses.

Table 1 Effect of vitamin E and C supplementation on the proliferation lymphocyte in purified lymphocyte and whole blood (OD in 540 nm)

Treatment	Purified lymphocyte			Whole blood		
	Without Mitogen (Control)	Mitogen		Without Mitogen (Control)	Mitogen	
		PHA	Con.A		PHA	Con.A
Vitamin E						
0	0.33 ^b	0.48 ^b	0.44 ^b	0.24	0.42 ^c	0.37 ^b
50	0.46 ^a	0.67 ^a	0.58 ^a	0.24	0.46 ^b	0.4 ^b
75	0.23 ^c	0.36 ^c	0.32 ^b	0.25	0.5 ^a	0.5 ^a
Vitamin C						
0	0.33 ^b	0.47 ^c	0.42 ^c	0.25	0.47	0.41
500	0.38 ^a	0.56 ^a	0.48 ^a	0.24	0.47	0.44
1000	0.32 ^b	0.49 ^b	0.45 ^b	0.25	0.45	0.43
SEM	0.007	0.008	0.006	0.012	0.008	0.008

Conclusions This study demonstrated that dietary VE improve the proliferative response of lymphocytes to polyclonal mitogens. VC supplementation (1000 ppm) improve body weight gain and cell mediated immune response, respectively. Moderate additions of combination VE (50 IU/kg) and VC affected the indices of immune response more than higher levels of supplementation. We hypothesize that moderate and high dietary levels of VE and VC may have different effects on the cellular free radical antioxidant balance, which results in different signal transduction events and activation states of the immune cells.

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Application of a duplex PCR approach for the specific and simultaneous detection of *Bifidobacterium* spp. and *Lactobacillus* spp. in duodenum, jejunum, ileum and cecum of broilers

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Introduction *Bifidobacterium* spp. and *Lactobacillus* spp genera are useful and important microbiota which affect poultry performance. These bacteria are natural inhabitants of the intestine of broilers. These bacteria have a unique hexose metabolism that occurs via the phosphoketolase pathway. *Bifidobacterium* spp. produces the vitamins B1, B2, B6, B12, nicotinic acid and folic acid. Enzymatic hydrolysis by *Bifidobacterium* spp. increases bioaccessibility of lipids and proteins. *Lactobacillus* spp. exhibit properties which include the ability to adhere to cells, persist and multiply and produce acids, hydrogen peroxide and bacteriocidal and bacteriostatic agents. The ability of *Lactobacillus* to inhibit bacterial pathogens including *E. coli* and *Salmonella* species and to influence parasitic infection including *Cryptosporidium parvum*, *Eimeria acervulina*, and *Giardia intestinalis* (Seidavi *et al.* 2008). The aim of this study was to develop a method for the direct and simultaneous detection of *Bifidobacterium* spp. and *Lactobacillus* spp. in the intestine by multiplex PCR.

Materials and methods Bacterial genomic DNA was extracted from the contents of gastrointestinal segments with the Wizard[®] Genomic DNA Purification Kit (Promega Corporation, Madison, WI, USA) with minor modifications. All oligonucleotide primers used in this study were synthesized by Metabion International AG. Sequences of the two PCR primer pairs for duplex PCR and size of expected amplification products are presented in Table 1. The GenBank program BLAST (<http://www.ncbi.nlm.nih.gov/BLAST>) was used to ensure that the applied primers were complementary with the target species but not with other species. Primers were compared with sequences in the GenBank, and none were found to have the exact same sequence as the non-targeted sequence. Different combinations of primer ratios were subsequently tested and the primer combination found to be optimal for duplex PCR was 0.28 μ M of each primer (Bif164-f, Bif662-r, LAA-f and LAA-r). The optimum Taq polymerase amount was determined by performing PCR for 0.5, 1, 1.5 and 2 U of Taq polymerase and 1 U of Taq polymerase produced high amplicon yields. The optimum Mg²⁺ concentration for duplex PCR was obtained by adding 1.2 mM MgCl₂. Finally, the optimum extension time was determined by performing PCR extension reactions for 45, 60, 90 and 120 s and an extension time of 60 s produced high amplicon yields. The PCR amplification mixture (25 μ l) consisted of 1 μ l of 25 ng DNA sample, 0.08 mM of each dNTP, 1.2 mM MgCl₂, 1 \times PCR buffer, 0.28 μ M of each primer (Bif164-f, Bif662-r, LAA-f and LAA-r), 1 U of Taq DNA polymerase and 18.6 μ l ddH₂O. Amplification was performed on a thermocycler which initial denaturation was at 95°C for 3 min, followed by 35 cycles of denaturation at 94°C for 45 s, annealing at 55.5°C for 45 s and extension at 72°C for 60 s, with a final extension at 72°C for 3 min.

Table 1 Primer sequences and size of PCR-amplified gene targets of two types of studied bacteria

Bacteria	Primer	Primer Sequence (5'→3')	Expected product size(bp)
<i>Bifidobacterium</i> spp.	Bif164-f	GGGTGGTAATGCCGGATG	510
	Bif662-r	CCACCGTTACACGGGAA	
<i>Lactobacillus</i> spp.	LAA-f	CATCCAGTGCAAACCTAAGAG	286
	LAA-r	GATCCGCTTGCCCTTCGCA	

Results All gastrointestinal samples tested including duodenum, jejunum, ileum and cecum showed positive results with the PCR assay for both *Bifidobacterium* spp. and *Lactobacillus* spp. There was no cross reactivity among these two closely related microorganisms. All primer pairs showed specificities only for their corresponding target microorganisms. In this investigation, two pairs of oligonucleotide primers were applied to simultaneously detect two different genera of useful bacteria by duplex PCR in a single tube. They are targeted at a species-specific region of the 16S rDNA. The specificity of the primers Bif164-f and Bif662-r for specific detection of *Bifidobacterium* spp. and of LAA-f and LAA-r for specific detection of *Lactobacillus* spp. has been reported. The primers can be used in gut samples with successful results. The genus-specific duplex PCR amplified 16S rDNA of the correct predicted size from the appropriate *Bifidobacterium* spp. and *Lactobacillus* spp. tested, but not from the other bacterial strains. When the two genus specific primer pairs were used individually, the same results were obtained as when the two reactions were run together as a duplex PCR. This technique complemented by microbiological technique to prove the validity of the PCR primers. Culturing of samples proved bacterial samples used in this study. Thus, the specificity of the PCR primers verified, and prevent to the detection of false positive.

Conclusion *Bifidobacterium* spp. and *Lactobacillus* spp. genera represent the probiotic cultures generally used in the food and poultry industry. The high throughput and cost-effective duplex PCR system developed in this study could provide a powerful supplement to conventional methods for more accurate risk assessment and monitoring of these useful bacteria in the poultry gut. Also in future, it must be developed approaches like real-time PCR for quantification of these bacteria.

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Investigation on relative population of *Salmonella* spp. bacteria in duodenum, jejunum, ileum and cecum of poultry using densitometry technique

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Introduction Salmonellosis is an infectious disease caused by *Salmonella* spp. that continues to plague human populations in both developed and third-world countries (Bohórquez, 2007). Conventional culture-based methods of detection that rely on enrichment of the sample and plating onto selective agar media may not be as sensitive as immunologic- or genetic-based rapid methods (Feng, 2001). PCR-based methods offer the advantages of high specificity and sensitivity. Some efforts have been made to reduce the time required and to increase the sensitivity and the accuracy of the methods to quantification of *Salmonella* in poultry samples (Mandrell and Wachtel 1999). This study was conducted in order to determine the relative population of *Salmonella* spp. in the duodenum, jejunum, ileum and caecum of chickens at different ages using densitometry.

Materials and methods Twenty four broilers were raised under conditions identical to those found in commercial broiler operations. DNA was extracted from duodenum, jejunum, ileum and cecum contents. Then PCR primers were optimized for detecting *Salmonella* spp. and all bacteria using specific and universal primers respectively. Amplified products were electrophoresed in 1.4% agarose gels containing ethidium bromide. A pUC Mix Marker 8 was used as molecular size marker. The relative population of *Salmonella* spp. to total gut bacteria was calculated by means of densitometry technique.

Results The population of *Salmonella* spp. relative to total bacteria in the different segments of the broiler gastrointestinal tract and different ages are shown in Figures 1 and 2. The relative populations of *Salmonella* spp. in the lower regions i.e. ileum and caecum were more than in the upper regions i.e. duodenum and jejunum. The highest and lowest relative population of *Salmonella* spp. were obtained at 4 and 30d of ages respectively. The results showed that the relative population of *Salmonella* spp. in duodenum, jejunum, ileum and cecum of broilers in 4d of ages were 0.28, 0.11, 0.16 and 0.91%. Also, relative population of *Salmonella* spp. in duodenum, jejunum, ileum and cecum of broilers in 14d of ages were 0.14, 0.18, 0.15 and 0.36%. Also, The highest and lowest relative population of *Salmonella* spp. in 30 day of ages obtained in ileum and duodenum of broilers (0.17 and 0.0%) respectively. Since these segments function differently and provide different environments, it is expected that different types of bacteria would colonize them. Also it is suggested bacteria population are transferred among various gut segments as animal aged.

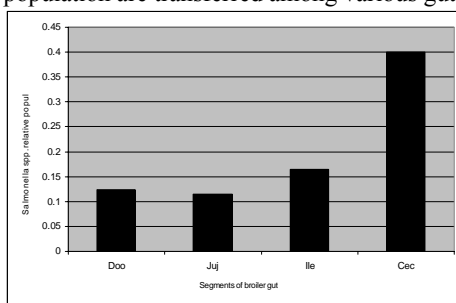


Figure 1

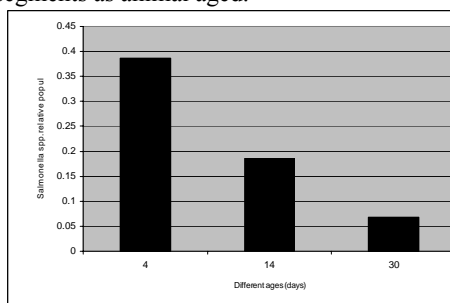


Figure 2

Figure 1 *Salmonella* spp. population relative to total bacteria in different segments of broiler gut (%).

Figure 2 *Salmonella* spp. population relative to total bacteria (average counts for all segments) in different ages (%).

Conclusion Enzmann *et al.* (1999) reported densitometry technique have high efficiency and precise in mitochondrial DNA quantification relative to spectrophotometer approach. Zhang *et al.* (2007) calculated quantities of immunoglobulin proteins, bovine serum albumin, casein, and beta-lacto globulin in dairy products. They confirmed this approach is more precise and efficient than conventional methods. The densitometry based on PCR protocol used in this work quantified the population of *Salmonella* spp. relative to total bacteria in intestinal samples as efficiently. The results from the densitometry protocol were obtained a few hours after sample processing began. Furthermore this reduction in quantification time represents a true advantage compared to the traditional culture and other rapid methods in food and gut microbiology.

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Relative frequency alteration of *Escherichia coli* in broiler intestine

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Introduction *Escherichia coli* is a commensal bacterium of the gut microflora of the chicken. *Escherichia coli* infections are responsible for significant economic losses in the poultry industry world-wide. The pathogenesis and the role of virulence factors are not yet fully elucidated, although considerable progress has been made in recent years to establish the mechanisms of pathogenesis. *Escherichia coli* strains cause a number of diseases in domestic poultry, ultimately leading to disease and death, or to a decrease in egg and meat production or condemning of carcasses. On the other hand, our knowledge about the composition of the gut flora and microbial ecology of the gastrointestinal tract is still limited. Previous investigations have mainly used culture-dependent approaches. Studies on the composition of the intestinal flora of chickens date back to 1901 (Rahner, 1901) and were continued in the 1940s (Shapiro and Sarles, 1949), but comprehensive surveys that attempted to culture as many of the intestinal bacteria as possible were not carried out until the 1970s (Barnes *et al.* 1972; Salanitro *et al.* 1974). Such studies are technically difficult since strict anaerobic conditions have to be maintained during isolation and biochemical differentiation of the bacteria. It is well recognized that many bacteria have not been cultured yet in the laboratory because their growth requirements are still unknown. Recent molecular studies have yielded more detailed insight into the composition of the microbial community of this ecosystem (Zhu *et al.* 2002).

The objective of this study was to develop a PCR based method for rapid quantification of *Escherichia coli* and investigation on its relative frequency in duodenum, jejunum, ileum and cecum of broilers.

Materials and methods DNA from gastrointestinal contents extracted as described by Seidavi *et al.* (2008). The specific detection of the *Escherichia coli* species was based on PCR amplification of the 16S rRNA gene using oligonucleotide primers ECO-f GACCTCGGTTTAGTTCACAGA and ECO-r CACACGCTGACGCTGACCA which PCR product size was 585 bp. Specific bands related to *Escherichia coli* obtained in duodenum, jejunum, ileum and cecum of broilers. Relative frequency alteration of *Escherichia coli* calculated by means of Gel-Proc Analyzer software for each age in different intestine segments and each intestine segment in different ages. Relative frequency is showed alteration proportion of this bacterium in various ages and segments. The linear regression with extrapolation method is selected for investigation on relative frequency alteration of *Escherichia coli*, since this method had the lowest variance. Obtained data from densitometry approach is entered and proceed using excel software.

Results In the rearing period, the highest relative frequency of *Escherichia coli* occurred in the cecum (76.47%) and the lowest relative frequency of *Escherichia coli* in the duodenum (0.0%) Relative frequency of *Escherichia coli* consists 62.72, 0.47 and 36.79% at 4, 14 and 30 day of ages. Other results are summarized in Tables 1 and 2. Today, accessing to a method not based on microbial culture is necessary for quantification of data related to bacteria populations, since PCR can detect target sequences and does not require cell growth. However microbial culture depended to target bacteria growth (Bjerrum *et al.* 2006).

Table 1 Relative frequency of *Escherichia coli* for each age in different intestine segments

Intestine Segment▶	Duodenum	Jejunum	Ileum	Cecum
Age (day)▶ 4	0.0	0.03	33.69	62.67
14	0.0	0.002	0.002	99.43
30	0.0	0.0	0.002	99.71

Table 2 Relative frequency of *Escherichia coli* for each intestine segment in different ages

Age (day)▶	4	14	30
Intestine Segment▶ Duodenum	0	0	100
Jejunum	99.94	0.05	0
Ileum	99.50	0.001	0
Cecum	51.40	0.61	47.97

Conclusion From obtained results is showed that relative frequency of *Escherichia coli* is different in duodenum, jejunum, ileum and cecum of broilers. Furthermore, this bacteria profile is altered at different rearing periods. Thus it is recommended that microbial flora studies perform PCR based methods for investigation on effects of factors such as diet ingredients, diet processing, enzyme, probiotics, environment etc on relative frequency of bacteria.

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Effects of different levels of canola meal on egg quality

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Introduction In recent years there has been increasing interest in cultivating canola in tropical areas. This product is used to supply edible oil for humans, and also as a high quality protein source for poultry (Baker and Chang 1992), soybean meal has been replaced by canola meal and there were some negative and positive effects on egg production, egg quality, egg shell quality (Roth-maier1999) In the present study, investigating the effect of canola meal on egg quality, different levels of soybean meal were replaced by canola meal in laying hen diets.

Materials and methods 96, 60 weeks old layer hens, Hy-line W-36 variety were used in a completely randomized design with 4 replicates in 3 periods (28 days per period) for investigating the soybean meal replacement by canola meal at 6 levels (0, 20, 40, 60, 80, 100%) based on crude protein, on egg quality. The data for Haugh unit, yolk colour, specific gravity, egg weight, egg production and egg taste were analysed by SAS(8.02)

Results The result of this experiment are summarised in table 1. The effects of different levels of canola meal on egg production, Haugh unit, yolk colour and egg taste were not significant ($P>0.05$). The specific gravity for 100% replacement treatment was significantly lower in comparison with other treatments ($P<0.05$). Egg weight has been effected by replacement of high levels of canola meal and it was significantly lower in comparison with control treatment ($P<0.05$).

Table 1 Ingredients of experimental diets

treatment	Cor n %	Soybean meal %	Canola meal %	DCP %	Oyster shell %	Soybean oil %	DLmet %	L- lys%	Salt %
0	67	18.12	0	1.16	10.87	1.93	0.09	0.02	0.31
20	65.5	14.64	4.55	1.14	10.82	2.38	0.08	0.08	0.31
40	64	11.5	9.11	1.12	10.77	2.82	0.08	0.14	0.32
60	62.5	7.67	13.66	1.1	10.72	3.27	0.07	0.2	0.32
80	61	4.19	18.22	1.09	10.68	3.72	0.07	0.26	0.32
100	59.2	0	23.69	1.06	10.62	4.26	0.06	0.33	0.32

Table 2 Effects of dietary treatments on egg weight, egg production, specific gravity, Haugh unit, Yolk colour and Egg taste.

treatment	Egg production	Egg weight	Yolk colour	Haugh unit	specific gravity	Egg taste
0	72.83	62.46 ^{ab}	7.94	82.7	1.08 ^a	5.86
20	73.83	62.7 ^a	8.1	83.43	1.08 ^a	6
40	74.92	61.44 ^{bc}	8.11	84.59	1.079 ^a	5.59
60	73.5	61.14 ^c	8.14	82.02	1.079 ^a	5.59
80	75.2	60.99 ^c	8.07	81.33	1.078 ^{ab}	6.14
100	73.58	61.01 ^c	7.97	84.9	1.075 ^b	5.63
SE	1.29	0.39	0.142	1.27	0.001	0.61

† Means by the same letter within columns are not significantly different at $P=0.05$ (Duncan's).

Conclusions It has been suggested before that high level of sulphur ion in canola meal has negative effects on calcium absorption, and also high levels of phytate in this product could decrease calcium and phosphorous absorption and deposition in egg shell and decrease egg specific gravity. High levels of phytate and crude fibre in this product decrease nutrient digestibility and result in decrease of egg weight (Summers *et al* 1988). This study showed that replacement of soy bean meal by canola meal could have economical saving in laying hen diets but because of mentioned reasons the suitable level for replacement could be debated and needs further investigation.”

Acknowledgements The authors would like to express their special regards for financial support of University of Tehran.

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Modelling and optimising early performance for broiler chicks

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Introduction Several methods have been introduced to estimate the optimum level of dietary nutrients such as metabolisable energy (ME), crude protein (CP), and lysine (Lys) in broiler chicken production. Performance optimisation is usually measured as maximising body weight gain and minimising adjusted feed conversion ratio (Adj FCR). One useful method is to model a system that requires an explicit mathematical input-output relationship. Group method of data handling-type neural network (GMDH-type NN) and genetic algorithm (GA) is used to model and optimise an output in an imprecise environment (Yao, 1999). The purpose of this study was to apply the GMDH-type NN and GA methods to provide an optimised formula for broiler chicken performance based on the dietary level of ME, CP, and Lys.

Materials and methods Six hundred and forty d-old male broiler chicks (Ross 308) were assigned to eighty units of brooder batteries with 8 chicks per unit. The birds were fed a commercial starter diet (3,200 kcal of ME/kg, 23% CP, and 1.3% of Lys) till d 5. A factorial design experiment with four levels of ME (2900, 3000, 3100, and 3200 kcal/kg), four levels of CP (17, 20, 23, and 26 %), and five levels of Lys (1, 1.1, 1.2, 1.3, and 1.4 %) was used to provide eighty dietary treatments. The trial period started on d 5 and continued to d 20. The mean body weight gain (MBWG) and feed consumption of each unit were measured. Eighty data lines consisted of ME, CP, and Lys dietary levels as inputs, and the MBWG or Adj FCR as an output. The experimental data were imported to a GEvoM programme to generate polynomial equations (Ahmadi *et al.*, 2008; Nariman-Zadeh *et al.*, 2005). The GA was used to optimise polynomial equations and to find the optimum nutrient requirements for achieving maximum MBWG and minimum Adj FCR.

Results The corresponding polynomial equations produced to develop the MBWG model with goodness-of-fit criteria, R^2 and root mean square error (RMSE) were as follows:

$$y_1 = 171.272 + 5.732CP + 46.216Lys - 0.771CP^2 - 265.973Lys^2 + 30.207CP.Lys$$

$$y_2 = -0.000013 - 0.019839ME - 0.000002Lys + 0.000039ME^2 + 0.000014Lys^2 + 0.018739ME.Lys$$

$$MBWG = -0.000045 + 0.346757y_1 - 0.173136y_2 - 0.002605y_1^2 - 0.001864y_2^2 + 0.006698y_1.y_2$$

$$R^2 = 0.68 \quad RMSE = 25.5$$

The corresponding polynomial equations produced to develop the Adj FCR model with goodness-of-fit criteria, R^2 and RMSE were as follows:

$$y_1 = 2.319 - 0.060CP + 0.362Lys + 0.0022CP^2 + 0.321Lys^2 - 0.054CP.Lys$$

$$y_2 = 0.0000010 + 0.0014879ME + 0.0000006Lys^2 - 0.0000003ME^2 + 0.00000002Lys^2 - 0.0000056ME.Lys$$

$$Adj \ FCR = 45.087 + 2.640y_1 - 58.845y_2 + 0.692y_1^2 + 19.850y_2^2 - 2.424y_1.y_2$$

$$R^2 = 0.58 \quad RMSE = 0.08$$

The results of optimization, ME, CP, and Lys requirements for maximization of MBWG or minimization of Adj FCR are shown in Table 1.

Table 1 Optimum dietary nutrient requirements for broiler chicken performance during d 5 to 20

	ME (kcal/kg)	CP %	Lys %
MBWG	3158	25.87	1.37
Adj FCR	3036	24.97	1.38

Conclusion This method of data mining may be used to model and optimise broiler performance based on dietary nutrients. More data of this type is needed to more accurately model and optimise performance.

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Modifying egg fatty acid content by supplementation of laying hen diets with palm olein oil (POO)

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Introduction Palm oil is the most abundant of all oils produced globally. It is very high in saturated fatty acids specifically palmitic acid, but other fatty acids (monounsaturated (MUFA) and polyunsaturated) are presented at low concentrations. In the processing plant some high amount of oleic acid with some other unsaturated fatty acids are extracted and marketed as Palm olein oil, and used to reduce blood or egg cholesterol (Rivelles *et al.*, 1994). The objective of this study was to determine the optimum level of dietary palm olein oil required to enrich the mono-unsaturated fatty acid content of yolk, egg cholesterol and antibody titre.

Materials and methods Eighty four 26-wk-old white Hy-line W-36 laying hens were randomly assigned to 4 diets, with 3 replicates of 8 layers each. The experiment was conducted over a period of 12 wks. The mash diets were provided iso-caloric and iso-nitrogenous (ME= 2820kcal/kg, Cp= 16.2%, Lys=0.84% Na=0.18%, Ca=3.70% and AP=0.39%) and contained 0, 1.5, 3, and 4.5% of Palm olein oil (POO). At the end of experimental period, 2 hens per replicate were randomly selected, weighed, and blood was withdrawn from wing vein.. Blood cholesterol and antibody titre against Newcastle disease (ND), Infectious bronchitis disease (IBD) were measured. One egg was randomly selected from each replicate hens, yolk was separated and stored in -20 C for later analyses. The yolk fatty acid contents were determined via the Folch method by GC apparatus. The data were analysed with SAS version 6.12.

Results The results of yolk and blood cholesterol and antibody titres, are presented in Table 1. The results of egg yolk fatty acid content and ratios of SFA/PUFA and omega-6/omega-3 are shown in Table 2.

Table 1 The effect of dietary palm olein oil on antibody titer, yolk and blood cholesterol of hens.

Performance	Dietary palm olein oil content (%)				SEM
	0.0	1.5	3.0	4.5	
Yolk cholesterol(mg/g)	12.95	13.20	13.55	13.75	0.65
Blood cholesterol(mg/dl)	143.00 ^b	157.67 ^{ab}	161.00 ^a	162.33 ^a	9.96
IBD titre	6812	6075	5590	5520	742.78
ND titre	8.67	8.33	8.33	8.67	0.67

^{a-b} Values within a row with no common superscripts differ significantly(P<0.05).

Table 2 The effect of dietary palm olein oil on yolk fatty acid contents and the ratio of SFA/PUFA and omega-6/omega-3 fatty acids.

Fat content (%)	Dietary palm olein oil content (%)				SEM
	0.0	1.5	3.0	4.5	
Fatty acid					
Palmitic acid	34.92	34.13	32.78	29.99	1.65
Stearic acid	8.89	8.48	8.50	8.66	0.32
Oleic acid, n-9	39.96 ^c	40.06 ^c	43.13 ^b	46.00 ^a	0.707
Linoleic acid, n-6	10.24	10.49	10.51	10.39	0.42
Linolenic acid, n-3	0.50	0.56	0.47	0.46	0.154
Arachidonic acid, n-6	1.34	1.46	1.36	1.23	0.127
Eicosapentanoic acid, n-3	0.157	0.123	0.107	0.102	0.06
Docosahexaenoic acid, n-3	0.031	0.024	0.023	0.029	0.002
∑ n-3 fatty acids	0.69	0.62	0.60	0.58	0.005
∑SFA ¹	43.24	41.35	41.14	38.95	2.28
SFA/PUFA ²	0.81	0.79	0.78	0.77	0.08
ω -6 / ω -3	18.01 ^b	19.43 ^a	19.63 ^a	19.75 ^a	0.28

a-c Values within a row with no common superscripts differ significantly(P<0.05).

1- SFA: saturated fatty acid, 2- PUFA: polyunsaturated fatty acid

Conclusion The different levels of dietary POO did not (P> 0.05) affect the saturated fatty acid (meristic, palmitic and stearic acid), omega-7 (palmitoleat), omega-6 (linoleate and arachidonate) and omega-3 (Linolenate, eicosapentenoate and Docosahexaenoate) content of egg yolk. The oleic acid (major fatty acid in the omega-9 family) was increased (P<0.05) as the level of POO increased in the diet. The SFA and SFA/PUFA ratio did not change with the level of dietary palm olein oil. The increase in dietary POO caused an increase in the ratio of omega-6/omega-3 fatty acid and blood cholesterol (P<0.05). The yolk cholesterol, ND and IBD titre was not affected by dietary POO (P>0.05). It is concluded that the dietary POO level may affect the omega-9 fatty acid and the ratio of n-6/n-3 fatty acids of egg yolk.

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Analysis of inbreeding, effective population size and inbreeding depression in Iranian native fowls (Mazandaran Province)

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Introduction Mating of related individuals produces an inbred progeny, with an inbreeding coefficient that is one-half the additive relationship between its parents. One of the consequences of inbreeding is reduction of the mean phenotypic value or inbreeding depression, particularly for reproductive and fitness characters. Another consequence of inbreeding is reduction in additive genetic variance within lines or increasing homozygosity and higher risk for the incidence of lethal or deleterious recessive alleles. In recent years many studies show that inbreeding reduces reproduction and fitness capacity, for example Inbreeding depression in reproductive and productive traits has been reported by Flock *et al* (1993), Smith *et al* (1998), Klemetsdal (1998), Thompson *et al* (2000a,b) and Nwagu *et al* (2007).

The purpose of this study was to investigation recent trends in inbreeding in Iranian Native Fowls and relationship between increase in inbreeding and decreases in economic traits including body weight, age at first egg, egg production and egg weight.

Materials and methods A pedigree file of total 50,508 including 10458 cocks and 40050 hens (was born during 1988 to 2004) was investigated for the occurrence of inbreeding. Also body weight (BW), age at first egg (AFE), egg production (EP) and egg weight (EW) were analyzed for study the inbreeding effect on these traits. Inbreeding coefficients was included in the animal model as a linear covariate. The following animal models were employed:

for body weight: $y_{ijk} = \mu + s_i + g_j + h_k + bX_{ijk} + a_{ijk} + e_{ijk}$, for age at first egg: $y_{jk} = \mu + g_j + h_k + bX_{jk} + a_{jk} + e_{jk}$,

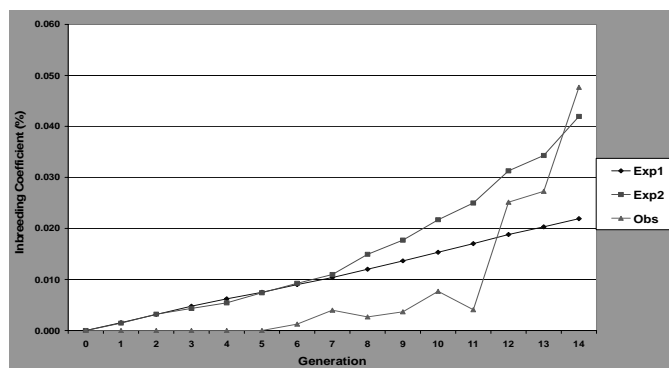
for egg weight: $y_{jk} = \mu + g_j + h_k + bX_{jk} + a_{jk} + e_{jk}$ and for egg production: $y_{jk} = \mu + g_j + h_k + bDIP_{jk} + bX_{jk} + a_{jk} + e_{jk}$, where y was observation of ijk individual, μ , s_i , g_j , h_k , DIP_{jk} , a_{ijk} , e_{ijk} were overall mean, sex, generation, hatch, days in production as covariate for egg number, random additive genetic effect and random error effect respectively, b and x_{ijk} were coefficient of partial linear regression and inbreeding coefficient deviation of ijk individual. Computations were performed using the DFREML package programmes of Meyer (2001). On the other hand the expected inbreeding according to the effective population size was predicted and compared with observed inbreeding during 14 generations. The first expected inbreeding was according to the number of parents (equation 1) and second one was according to the family size variance (equation 2):

$$\text{equation 1: } N_e = \frac{4N_m N_f}{N_m + N_f} \quad \text{equation 2: } N_e = \frac{4N}{V_i + 2}$$

Results The overall mean inbreeding during 14 generation was 0.01246, so that a large proportion of population were inbred (26153 or 51.78%) with mean inbreeding of 0.02387. On the other hand the linear regression coefficients of BW, AFE, EN and EW on inbreeding was -0.457 g, 0.122 day, -0.218 number and -0.006 g per 1% increase of inbreeding respectively that its effect was significant only on egg number ($p < 0.001$).

Table and chart 1 Comparison of observed and expected inbreeding according to the effective population size: Obs: Rate of inbreeding according to the pedigree, Exp1: expected rate of inbreeding according to the number of parents and Exp2: rate of inbreeding according to the family size variance.

Gen	N_m	N_f	Ne1	Exp1	Ne2	Exp 2	Obs
0	94	574	323	0.000	344	0.000	0.000
1	89	617	311	0.002	284	0.001	0.000
2	87	659	307	0.003	445	0.003	0.000
3	99	654	344	0.005	456	0.004	0.000
4	111	717	384	0.006	250	0.005	0.000
5	95	683	334	0.008	269	0.007	0.000
6	102	705	356	0.009	290	0.009	0.001
7	87	588	303	0.010	124	0.011	0.004
8	85	656	301	0.012	178	0.015	0.003
9	84	609	295	0.014	123	0.018	0.004
10	81	637	287	0.015	149	0.022	0.008
11	79	555	277	0.017	77	0.025	0.004
12	94	645	328	0.019	160	0.031	0.025
13	86	634	303	0.020	63	0.034	0.027
14				0.022		0.042	0.048



Conclusions Inbreeding affected significantly only egg production with less heritability (0.16). It is clear that the expected inbreeding according to the number of parents (0.022) was less than observed (0.042). It means that this method can not predict inbreeding correctly because of non random mating. However, inbreeding according to family size variance was closer to observed inbreeding. This is because information from relatives in selection schemes is used (BLUP Animal Model) giving high co-selection probabilities, family size variance and increased rates of inbreeding during generations.

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White mulberry (*Morus alba*) fruit waste and its use in broiler nutrition

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Introduction The mulberry belongs to the genus *Morus* of the family Moraceae. There are 24 species of *Morus* and one subspecies, with at least 100 known varieties. One of the most important points about mulberry fruit is its sensitivity to environmental conditions during harvesting time and also the methods of harvesting. Because of these sensitivities, there is a lot of waste in the production of mulberry fruit. In the animal feed industry, agricultural by-products are used in animal and poultry nutrition. However, there is no information about the use of mulberry fruit waste in poultry nutrition; therefore this experiment was carried out to investigate the effect of mulberry fruit in broiler nutrition.

Materials and methods According to the result of previous experiment, the metabolizable energy of mulberry fruit waste is similar to corn grain (AMEN= 13.62MJ/Kd DM), but its crude protein content is half of corn grain (CP=4.4%). As the price of corn grain is 2.5-3 times higher than mulberry fruit waste, we can use this material as a source of energy for poultry. A total of 400 day-old chicks were randomly assigned to 16 litter floor pens (4 replicate per treatment). Experimental groups consisted of 0, 5, 10 and 15 % white mulberry that were inserted in broiler rations. The diets were formulated according to Cobb 500 nutritional recommendations for broiler chickens. Feed and water were provided for ad libitum consumption. In the experimental period (7-42 days) feed consumption (g/d), mortality (%), weight gain (g/d) and feed conversion were measured weekly. Finally at the age of slaughter (42d) 2 chicks were selected randomly from each replicate and carcass quality, carcass weight, breast, leg, abdominal fat, liver, kidney and heart weights were measured.

All data were subjected to one-way analysis of variance. Statistical significances among treatment means were determined by the method of new multiple range test of Duncan (1995) when the F value was deemed significant at 0.05.

Results Using different levels of white mulberry fruits waste had no significant effect on feed intake, but the amount of feed intake in all white mulberry fruits waste group was higher than the control. Weight gains in white mulberry fruit waste groups were lower ($P<0.05$) than controls. Though not measured, it seems that the diets which included mulberry waste had a higher bulk density, and this maybe one of the reason of decreasing weight gain without changing in feed intake. Carcass, leg, breast, heart and abdominal fat were not affected by the level of mulberry fruit waste. Kidney weight percentage was increased by increasing level of mulberry fruits waste supplementation. This increasing in kidney can be related to high content of K in mulberry fruits waste. Increasing concentration of white mulberry fruit waste was associated with an increase in liver weight (%) which may be related to increase of liver activity.

Conclusion In conclusion mulberry fruit waste can be used in poultry diets as an energy source (because of its low price in comparison with corn). In this experiment the use of mulberry had negative effect on performance so we recommended that further study evaluate the mechanism of this effect and determined the best level of supplementation and in poultry nutrition specially, layer hens.

Table 1 The effect of different levels of mulberry fruit waste on broiler performance

White mulberry fruits waste (%)	Feed Intake(g/d)	Weight gain(g/d)	Feed Conversion	Liveability (%)	BW42	Production Index
0	107.32	52.55a	2.04c	96	2139a	239.44 a
5	110.49	42.29b	2.24b	98	2008a	209.03 b
10	111.43	48.88b	2.28b	98	1995a	204.35b
15	110.95	43.40c	2.56a	96	1776b	158.81c
SEM	0.87	0.95	0.05	0.68	39.42	8

Table 2 The effect of different levels of white mulberry fruit waste on broiler carcass characteristics

White mulberry fruits waste (%)	Carcass (%)	Leg (%)	Breast (%)	Kidney (%)	Liver (%)	Heart (%)	Abdominal fat (%)
0	64.33	20.85	21.59	0.557b	2.47c	0.48	1.08
5	64.36	19.80	21.57	0.625ab	2.65bc	0.49	1.04
10	63.52	20.58	21.15	0.711a	2.99ab	0.54	0.913
15	66.67	19.81	20.99	0.969a	3.25a	0.55	1.02
SEM	0.97	0.24	0.23	0.02	0.09	0.02	0.11

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Effect of various levels of full-fat sunflower seed on performance of broiler chickens

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Introduction Sunflower (*Helianthus annuus*L.) is one of the most widely cultivated oilseeds in the world and ranks third in importance as a source of vegetable oil. As an alternative to fats and oils, full-fat oilseeds such as soybean seed are used to replace the supplemented fats and oils in broiler diets. However, soybean seed has anti-nutritional factors such as trypsin inhibitors, which need further processing, thus increasing the cost of soybean seed. Among the various oilseeds available on the market, full-fat sunflower seed (FFSS) contains more ether extract (EE) and is available at a relatively low price. This experiment was conducted to study the effect of FFSS (that FFSS had 18% CP, 38% EE, 14.3% CF, and 3868 kcal/kg ME) on performance of broiler chickens.

Materials and methods In this study, 176 day-old male broiler chickens (Ross strain) were allocated to four treatments with four replicate (11 in each replicate) in a completely randomized design to evaluate the effect of FFSS on the performance of chickens for 7 weeks. Treatments were 0, 7, 14 and 21 percent of FFSS for starter (1-21 days) and grower phases (22-49 days). The diets of starter phase calculated to contain 20.86% CP and 3000 kcal of ME per kg of diet. They also contained 18.75% CP and 3000 kcal of ME per kg of diet for the grower phase. Body weight gain and feed consumption were recorded. Data for all parameters were subjected to an analysis of variance, using the general linear model procedure of SAS (SAS Institute, 2004).

Results Feed intake increased significantly ($p < 0.05$) when increasing levels of FFSS was incorporated in the diet during the experiment except for 43-49 days of age (Table). Weight gain also increased significantly in the different stages of our experiment ($p < 0.05$). Except for 1 to 21 and 1 to 49 days of age, feed conversion ratio (FCR) improved significantly ($p < 0.05$).

Table 1 Effect of full-fat sunflower seed on performance parameters of broiler (1-49 days of age)

	Feed Intake (g/b)				Weight Gain (g/b)				Feed Conversion Ratio			
	1-21	22-42	43-49	1-49	1-21	22-42	43-49	1-49	1-21	22-42	43-49	1-49
FFSS (g/kg)												
0	823.6 ^b	2446 ^b	1019	4125.1 ^b	417 ^b	1117 ^b	350 ^b	1741 ^b	1.97	2.54 ^a	2.91 ^a	2.39
70	874.7 ^{ab}	2793 ^a	1089	4757.4 ^a	451 ^{ab}	1099 ^b	456 ^{ab}	1935 ^{ab}	1.94	2.48 ^a	2.38 ^b	2.45
140	903.6 ^{ab}	2950 ^a	1168	4985.6 ^a	503 ^a	1199 ^{ab}	450 ^{ab}	2096 ^a	1.79	2.19 ^b	2.59 ^{ab}	2.38
210	932.1 ^a	2809 ^a	1139	4762.9 ^a	497 ^a	1311 ^a	492 ^a	2155 ^a	1.90	2.14 ^b	2.31 ^b	2.22
SE	29.47	24.40	85.35	5.93	10.01	20.97	24.83	0.069	0.121	0.281	0.10	

^{a,b}Within the same column, means with different letters are significantly different ($p < 0.05$).

Conclusions FFSS was proven as a good source of CP and ME in broiler diets. The results from the current experiment indicated that substitution of FFSS for corn, soybean meal up to 210 g/kg of diet had positive effect on performance parameters. These results are in accordance with those obtained by Elzubeir and Ibrahim (1991), who reported that unprocessed sunflower seed can be given to broilers at up to 225 g/kg of the diet with no adverse effects on performance. In contrast with these findings, Dagher *et al* (1980) observed that feeding 150 and 250 g kg⁻¹ FFSS to broilers depressed both body weight gain and feed intake. One possible explanation for this disagreement of results is that different sunflower varieties or cultivars varying in chemical composition were used in the experiments.

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Determination of apparent metabolizable energy of full-fat sunflower seed in broiler chickens

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Introduction Sunflower (*Helianthus annuus* L.) is one of the most widely cultivated oilseeds in the world and ranks third in importance as a source of vegetable oil. In our experiment, full-fat sunflower seed (FFSS) had 180 g/kg CP, 380 g/kg EE, and 143 g/kg CF. Among the various oilseeds available on the market, FFSS contains more ether extract (EE) and is available at a relatively low price. This high EE content contributes to a high ME per unit or high energy density of feed. The increased production and availability of hybrid FFSS coupled with its oil content make FFSS a potentially desirable ingredient in poultry feeds. In the last few years, unextracted whole seed has been used as a feed ingredient in poultry diets. This experiment was conducted to determine the apparent metabolizable energy (AME_n) of FFSS in broiler chickens.

Materials and methods One-day old male chicks of Ross strain were housed in floor pens, exposed to light for 24 h/d, and fed a standard broiler diet for 2 wk. Feed and water were provided *ad libitum*. FFSS was incorporated into the basal diets at 3 concentrations (70, 140, and 210 g/kg). The 4 experimental diets, which contained 3 g/kg chromium oxide as an indigestible marker, were evaluated in a balance trial to determine the ME content. On d 10, 80 birds were placed at random in 16 cages giving 4 replicates per dietary treatment. On d 15, the birds were starved for 4 hours and then received the experimental diets from 15 to 21 d of age. During the last 3 d, excreta samples from each cage were collected and stored at -20°C. After being thawed, excreta were homogenized, dried, and ground through a 1-mm screen. Diets and excreta were analysed for dry matter, CP, chromium oxide, and gross energy. Apparent metabolizable energy was calculated as follows:

ME (MJ/kg) = dietary gross energy × [1 - (diet Cr2O3 / excreta Cr2O3) × (excreta gross energy / diet gross energy)].

The correction of AME to zero nitrogen retention (AME_n) was based on a factor of 34.4 kJ/g of retained N (Hill and Anderson, 1958). The AME_n value of FFSS was calculated using the following equation: AME_n = (AME_n T - α × AME_n B) / b, where T is the test diet, α is the proportion of the basal diet in the test diets, B is the basal diet, and b is the proportion of FFSS in the test diets. Statistical analyses were performed by using the GLM procedures of SAS software (SAS Institute, 2004). Data were subjected to ANOVA to identify variation produced by inclusion level of FFSS; regression analysis was also used to establish dietary changes as a function of inclusion level of FFSS.

Results Table shows AME_n data (MJ/kg) for the experimental diets and for FFSS using the difference technique described above. Increasing inclusion rate of FFSS increased the AME_n of the diets numerically but this effect was not significant. The AME_n (MJ/kg) of FFSS, calculated by difference (Table) ranged from 13.7 to 14.3 MJ/kg. The AME_n values obtained for the diets were regressed against the level of FFSS in the basal diet to estimate the AME_n content in FFSS. The equation derived by fitting a linear model was the following: y = 12.394 + 0.0018X (R² = 0.80). An estimate of the AME_n of FFSS, obtained by extrapolation of this equation gave a value of 14.22 MJ/kg.

Table 1 Apparent metabolizable energy (AME_n)¹ of diets with increasing levels of full-fat sunflower seed (FFSS), and of FFSS determined by difference and regression analysis

Level of FFSS (g/kg)	AME _n of diets (MJ/kg)	AME _n of FFSS (MJ/kg)
0	12.42	-
70	12.51	13.74
140	12.60	13.68
210	12.82	14.31
SEM	0.164	

¹AME_n determinations were made based on 16 cages of 1 bird each.

Linear regression equation: y = 12.394 + 0.0018X; R² = 0.8 where y = AME_n (MJ/kg) and x = dietary inclusion level of FFSS (g/kg)

In this experiment, the energy value obtained for FFSS was lower than the 18.70 MJ/kg and 17.67 MJ/kg reported by Rodriguez *et al* (1998) and Rodriguez *et al* (2005), respectively. This difference may be related to crude fat content because the sunflower seed they used had a higher amount of crude fat (473 and 444 g/kg respectively).

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Growth performance of broilers consuming wheat- or barley-based diets with or without enzymes

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Introduction Cereal grains, such as wheat, have been regarded as one of the most affordable ingredients to supply dietary energy for fast growing broilers. However, broilers compete with humans for wheat to satisfy their nutritional needs and also they cannot effectively utilise wheat due to the limited ability of their gut enzymes to utilise dietary fibre. The efficiency of wheat utilisation can be increased by adding exogenous enzymes into cereal based diets for these birds (Annisson and Choct, 1993). These enzymes can also reduce fermentation in the small intestine and so help maintain the gut health. Therefore, we compared the effect of adding a commercial fibrolytic enzyme to wheat and barley-based diets on the utilisation of either diet by broilers from 0 to 35 days of age.

Materials and methods Two sets of four iso-nitrogenous (213-221 g CP/kg) diets with 12.6-13.6 MJ ME /kg were prepared by mixing (per kg diet) up to 650g coarsely ground wheat (A) or barley (C) with other ingredients in the absence (A and B) or presence of an enzyme (E= β xylanase) (A+E= B and C+E=D). Other ingredients were soybean meal, sunflower oil, sodium chloride, calcium mono phosphate, calcium carbonate and vitamin mineral premix. For the B and D diets, the enzyme was thoroughly mixed first with a portion of the diet which was then remixed with the relevant diet. One hundred and fifty six day old female Ross 308 chicks were transported from a commercial hatchery to the University Farm. The chicks were identified by using leg rings, weighed and divided into 12 experimental units of 13 chicks each where each unit was balanced for initial chick weight. Each chick unit was randomly housed in a circular pen on concrete floors covered with wood shavings in a pre-heated room at 35°C. This room was set for recommended light durations and each pen was equipped with a brooder, feeder and water container. Each of the 4 diets was allocated to 3 pens of 13 chicks each according to a completely randomized design. The brooding temperature was decreased weekly by 3°C until it reached 22°C. These chicks were offered *ad libitum* water and relevant starter diets for 21 days and grower cum finisher diets for another 14 days to the age of 35 days. Weekly body weight per chick and feed consumption per pen was recorded and feed efficiency calculated. Chick health was monitored and feed samples collected for their chemical composition. For this paper, only broiler growth data involving both the starter and finisher diets were analysed using ANOVA on Minitab to compare the effect of these diets on the broiler body weight at each week. The means of these diets in each week were compared by using Tukey test at P<0.05.

Results The mean body weights (BW) of broilers at various days of age are summarized in Table 1. The birds continued to increase their diet intake with the increase in their age and so maintained a healthy growth. The BW at day 0 did not differ (P>0.05) between diets confirming that the initial bird distribution was uniform across the diets. While the diets caused significant differences in BW at 7, 14, 21 and 35 days of age (P<0.05), they did not differ at 28 days of age (P>0.05). The impact of wheat and barley based diets was variable depending upon the age of these birds. The mean BW weight of birds consuming wheat based diets was greater than those on barley based diets showing significance at days 7 and 14 (P<0.05). While, the overall effect of enzymes on BW was non significant at all days of age, the broilers had greater BW for the presence than the absence of enzyme in barley-based diets at 21 to 35 days of age (P<0.05 for diet C v D).

Table1 Mean body weight (g) of broilers fed wheat or barley based diets with or without enzymes at different days

Diets	Days →					
	0	7	14	21	28	35
A (Wheat)	35.7	164 ^{ab}	413 ^{ab}	808 ^a	1309 ^a	1829 ^{ab}
B (A+E)	36.0	175 ^b	422 ^b	790 ^a	1290 ^a	1819 ^a
C (Barley)	36.5	158 ^a	387 ^{ac}	746 ^b	1231 ^b	1680 ^c
D (C +E)	36.2	154 ^a	402 ^a	805 ^a	1300 ^c	1849 ^b

E= Enzyme; The figures with different superscripts in each column were significantly different at P<0.05

Conclusions The birds had greater body weights for wheat than barley based diets at most days of age. However, enzyme addition appeared to have greater effect on body weights when barley plus enzyme was compared with barley without enzyme at 21, 28 and 35 days of age. The study shows a potential in enzymes to improve the utilisation of barley based diets in order to improve bird performance and so reduce the cost of broiler production.

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Effects of antibiotic and probiotic supplementation to diets containing fat on broiler performance

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Introduction The use of supplementary fat in commercial poultry diets has been wide-spread since the 1960s. In addition to their value as a dense source of energy, supplemental fats are an excellent source of essential fatty acids and enhance the absorption of fat soluble vitamins. The efficiency of nutrient digestion in poultry largely depends on the microorganisms which live naturally in its digestive tract (Apajalahti *et al.* 2003). It has been reported that intestinal microflora influences the absorption of fats (Pesti, 2002). Today, antibiotics and probiotics are used for manipulating the gut microflora in poultry production and act as growth-promoting agents. Thus, using these growth promoters (GP) will probably affect nutrients absorption, especially of dietary fats. The aim of this study was to evaluate the effects of supplementary antibiotics and probiotics in diets containing different levels of fat on broiler performance

Materials and methods Three hundred and sixty one-day old Ross broiler chicks were used in a 2×3 factorial arrangements with 2 levels of soy oil (3 and 6% of diet) and 3 levels of growth promoter (No-GP, antibiotic and probiotic), with 4 replicates and 15 birds per replicate in a randomized complete block design. Flavomycin (400g/t) and protexin (500g/t) were added to diet as growth promoter. Diets were iso-energetic and iso-nitrogenous and sand were used as inert to diets containing high level of oil to adjust their metabolisable energy. Feed and water were provided ad-libitum and a 24h lighting program was followed throughout the 42 days experiment. Feed intake and Body weight gain were recorded weekly and feed consumption ratio was calculated. Blood samples were collected randomly from two birds per each replicate at 35 days of age. The blood samples were assayed for cholesterol, HDL, LDL, VLDL and triglycerides. All collected data were analysed using General Linear Models procedure of SAS (SAS, 2004), and means were compared by Duncan multiple range test.

Results The effects of fat levels, growth promoters on broiler performance and blood biochemical properties are presented in Table 1. Addition of probiotics to diets decreased feed intake and weight gain (P<0.05). Birds fed diets supplemented with probiotics had the lowest blood cholesterol and LDL. The probiotic effectiveness on broiler performance was related to levels of fat in diets and diets containing probiotic + 6% fat resulted the lowest weight gain (P<0.05).

Table 1 The effects of treatments on performance and blood biochemical properties of broiler chicks

Variables	Performance			Blood biochemical properties(mg/dl)				
	FI(g)	WG(g)	FCR	TG	Cholesterol	HDL	LDL	VLDL
Fat (%)								
3	3815.3 ^a	2203.1 ^a	1.75 ^b	71.9	162.1 ^b	85.2 ^b	66.8 ^b	14.2
6	3674.6 ^b	2022.6 ^b	1.82 ^a	74.1	197.7 ^a	94.4 ^a	86.7 ^a	14.8
SE	24.7	43.6	0.02	4.4	2.2	1.6	1.4	0.9
GP								
No-GP	3840.6 ^a	2110.0 ^b	1.76	71.6	179.8 ^b	60.3 ^{ab}	72.8 ^b	14.2
Antibiotic	3893.4 ^a	2221.5 ^a	1.81	69.0	195.3 ^a	96.0 ^a	91.4 ^a	13.7
Probiotic	3500.9 ^b	2007.0 ^c	1.79	78.4	164.6 ^c	82.9 ^b	66.1 ^c	15.6
SE	53.5	30.3	0.02	5.3	2.8	1.9	1.7	1.1
Fat(%) × GP								
3 × No-GP	4028.6 ^a	2233.9 ^a	1.77 ^b	74.2 ^{ab}	164.6 ^d	87.1 ^b	60.4 ^d	14.7 ^{ab}
3 × Antibiotic	3883.9 ^a	2216.4 ^a	1.75 ^b	77.2 ^{ab}	174.8 ^{cd}	93.3 ^{ab}	83.0 ^b	15.3 ^{ab}
3 × probiotic	3533.5 ^b	2159.1 ^a	1.73 ^b	64.3 ^b	146.9 ^e	75.2 ^c	57.0 ^d	12.8 ^b
6 × No-GP	3652.6 ^b	1986.1 ^b	1.8 ^b	68.9 ^b	195.1 ^b	93.6 ^{ab}	85.2 ^b	13.8 ^{ab}
6 × Antibiotic	3902.7 ^a	2226.7 ^a	1.77 ^b	60.8 ^b	215.9 ^a	98.8 ^a	99.8 ^a	12.1 ^b
6 × probiotic	3468.4 ^b	1854.9 ^c	1.89 ^a	92.4 ^a	182.2 ^c	90.7 ^{ab}	75.2 ^c	18.5 ^a
SE	75.7	42.9	0.03	7.5	3.9	2.7	2.4	1.6

^{a-d} Mean values with different superscripts on same column are significantly different (P<0.05).

Conclusions It was concluded that the supplementing diets containing fat with probiotics may have negative effects on broiler performance.

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Can medicinal plants with antimicrobial properties be replacement for antibiotics in broiler production?

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Introduction Antibiotics have been used for 50 years to enhance growth performance and to prevent disease in poultry production. Recently, most of the antibacterial growth promoters have been banned because the feeding of antibiotics is risky due not only cross-resistance but also to multiple resistances. Plants (especially herbs) have been used as food and medicinal purposes and some of them have played a significant role in maintaining human health and improving the quality of human life for thousands of years. Herbs or products containing plant extracts, essential oils or main components of the essential oil are among the alternative growth promoters that are already being used in practice. (Acamovic and Brooker, 2005; Ocak *et al.*, 2008). There is evidence suggesting that herbs, spices, and various plant extracts have appetizing, digestion-stimulating and antimicrobial properties. But, there is only limited evidence about whether the inclusion as a solid herb material would have the growth promoting effects in live birds. The aim of this study was to evaluate the antibacterial effects of dietary dry peppermint (*Mentha piperita L.*), Cumin (*Cuminum L.*), Milfoil (*Achellia millefolium L.*) and poley (*Teucrium Polium L.*) on the performance of broiler.

Materials and methods Three hundred and sixty one-old broilers (Arbor Acres) were randomly assigned to 6 treatment diets, with 4 replicates and 13 birds per replicate in a completely randomized design. The diets were isocaloric and iso-nitrogenous and contained 15, 3, 2 and 2 g/kg of dried herb of cumin, mint, Milfoil and poley, respectively. Two treatments with (flavomycin, 400g/t) and without antibiotic were considered as control groups. The diets were fed as mash and chicks were permitted free access to feed and water during the 42 days of experimental period. Continuous lighting was provided during the experiment. Body weight gain and feed intake were recorded weekly and feed conversion ratio was calculated on pen weight basis. All collected data were subjected to an analysis of variance, using the General Linear Model procedure of SAS (SAS Institute, 2004) and means were compared by Duncan's multiple range test.

Results The effects of treatments on broiler performance are presented in Table 1. Supplementation diets with antibiotic significantly ($p < 0.05$) improved body weight gain, and FCR in broilers during the 42 days rearing period when compared with other treatments.

Table 1 Effects of dietary inclusion medicinal plants and antibiotic on WG (g), FI (g) and FCR of broiler chicks.

Traits	Treatments						SEM
	Control	Antibiotic	Cumin	Mint	Milfoil	Poley	
Starter							
FI	397.6	404.6	396.1	399.6	387.7	385.5	5.75
WG	309.1 ^{ab}	319.1 ^a	308 ^{ab}	313.6 ^{ab}	297.2 ^{bc}	290 ^c	5.76
FCR	1.28	1.26	1.28	1.27	1.30	1.33	0.026
Grower							
FI	1196 ^c	1242.3 ^a	1200.7 ^{bc}	1222 ^{ab}	1209.3 ^{bc}	1195.9 ^c	7.53
WG	760.1 ^{bc}	776.4 ^a	753.5 ^c	767.4 ^{ab}	712.4 ^d	712.9 ^d	4.41
FCR	1.57 ^b	1.60 ^b	1.59 ^b	1.59 ^b	1.69 ^a	1.67 ^a	0.015
Finisher							
FI	2033.1 ^d	2163.5 ^b	2156.9 ^b	2196.4 ^a	2093 ^c	2157.5 ^b	10.94
WG	980.6 ^c	1030.1 ^a	982.2 ^c	1005.3 ^b	899 ^d	869.2 ^e	6.41
FCR	2.07 ^d	2.10 ^d	2.19 ^c	2.18 ^c	2.35 ^b	2.48 ^a	0.013
Total							
FI	3532.5	3589.8	3570.1	3601.1	3548.2	3504.5	38.15
WG	1936.7 ^c	2043.6 ^a	1982 ^{bc}	2008.5 ^b	1857.5 ^d	1821.3 ^e	11.06
FCR	1.79 ^b	1.75 ^b	1.80 ^b	1.79 ^b	1.91 ^a	1.92 ^a	0.019

^{a-d} Means in each row with different superscripts are significantly different ($P < 0.05$).

Conclusions It is concluded that medicinal plants had significant effect on broiler performance, but they can not be replacement for antibiotics. Mint leaves had most positive effects on weigh gain and feed conversion ratio than other studied medicinal plants. Further studies are needed to investigate the effects of different levels of mint, cumin, Milfoil or Poley on colonization and proliferation of microorganisms in the broiler intestine, and its performance.

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Effects of enzyme and probiotic supplementation to diets containing wheat on broiler performance

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Introduction Enzymes and probiotics are often used in feeding of poultry in intensive rearing systems. Beneficial effects of probiotics were observed in toxin neutralization, prevention of development and multiplication of specific bacteria, change in microbial metabolism and immunity stimulation. Administration of useful microorganisms as probiotic into diet usually cause enhancement of organic acids production such as lactic acid that in turn can reduce gastrointestinal pH and subsequent prevention of pathogen microorganisms such as salmonella to colonize the alimentary tract (Fuller, 1992). Exogenous enzymes have been shown to alleviate the adverse effects of high viscosity of digesta in the small intestine and to improve digestion (Petersen *et al.* 1999). Pervious studies demonstrate that simultaneously using probiotics and enzymes in broiler diets, improve their growth performance (Midili, and Tuncer, 2001). The objective of the present study was to examine the influence of probiotic (protexin) and enzyme (Natozyme) supplementation on the performance of broilers fed diets containing wheat.

Materials and methods Seven hundred and twenty male broiler chicks (Ross 308) were used in a 2×2×3 factorial arrangements with 2 levels of wheat (0 and 30% of diet), 2 levels of probiotics (0 and 300 g/ton) and 3 levels of enzyme (0, 100 and 200 g/ton), with 4 replicates and 30 birds per each replicate in a completely randomized design. Feed and water were provided ad-libitum and a 23:1 light: dark program was followed throughout the 6-week experiment. Feed intake and body weight gain were recorded weekly and feed consumption to weight gain ratio (FCR) was calculated. At the end of the experiment (week 6), two birds from each replicate were randomly selected, slaughtered and the weights of liver, caeca and abdominal fat were measured. All data were analysed using General Linear Model's procedure in SAS (SAS Institute, 1994) and means were compared by Duncan's test at 5% of probability

Results The effect of wheat, protexin and enzyme on broiler performance are shown in Table 1. The results showed that 30% wheat in diets decreased feed intake and body weight gain, significantly ($P < 0.05$). Supplementation diets with Protexin significantly ($p < 0.05$) improved feed intake, body weight and FCR, but addition of enzyme to diets had not significant effects on broiler performance. Relative weight of the abdominal fat significantly decreased in bird fed diets containing protexin, but protexin increased caeca weight ($P < 0.05$). The interaction effect between enzyme × probiotic × wheat on body weight gain and FCR was significant. Supplementation wheat-soy based diets with probiotic and enzyme improved body weight gain and FCR, significantly ($P < 0.05$).

Table 1 Effect of different levels of enzyme and probiotic and wheat on the performance of broiler chickens

Factor	FI(g)	BWG(g)	FCR	Abdominal fat%	Liver%	Caeca%
Wheat (%)						
0	4747 ^a	2224 ^a	2.12 ^a	1.05	1.29	0.49
30	4725 ^b	2114 ^b	2.34 ^b	1.10	1.34	0.46
Enzyme(g/ton)						
0	4702	2007	2.33	1.07	1.33	0.47
100	4760	2061	2.21	1.06	1.25	0.45
200	4728	2055	2.27	1.09	1.27	0.51
Probiotic(g/ton)						
0	4677 ^b	2007 ^b	2.36 ^b	1.16 ^a	1.31	0.44 ^b
300	4798 ^a	2131 ^a	2.21 ^a	0.99 ^b	1.32	0.51 ^a
SEM	6.01	8.71	4.97	14.47	12.87	19.55

^{a, b} Mean values with different superscripts on same column are significantly different ($P < 0.05$).

Conclusion It is concluded that the simultaneously using of probiotics and enzyme in wheat – soy based diets is a good way to obtain more economical benefits in broiler production.

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Effect of different levels of digestible lysine on performance, and blood parameters of male and female broilers in the starter period

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Introduction Lysine is the reference amino acid (AA) in the ideal AA ratios for chickens. Feed formulation based on digestible AA has been shown to increase weight gain and feed intake and improve body composition in broilers. Amino acid (AA) in most feed ingredients will not be totally digested, and knowledge of such efficiency is important in formulating diets and will be used to eliminate differences in absorption efficiencies due to feedstuff sources. This study was conducted to evaluate the growth performance and blood parameters of broilers fed various levels of DL (Digestible Lysine) supplemented in diets from day 1 to day 18.

Materials and methods Arian male and female broilers (1-18 days) by feeding practical diets formulating on digestible amino acids basis. A total of 240 male and female chicks were used (initial weight of 44±1 g). The birds were distributed in a completely randomized design, using six treatments and four repetitions, with 5 chicks per repetitions. These experimental chickens were kept in thermostatically controlled batteries. The experimental treatments consisted of a lysine-deficient basal diet that was supplement with L-lysine-HCl in order to contain six digestible lysine levels (0.68, 0.8, 0.92, 1.04, 1.16 and 1.28%). These diets were isocaloric, isonitrogenous and equal in electrolyte balance. All diets met an the Illinois recommended ideal amino acid ratio for all other amino acids. Feed and water supplied for *ad libitum* consumption. During the experiment feed consumption (FC), weight gain (WG) and feed conversion ratio (FCR) were measured weekly. Also, blood samples were collected at the end of the starter period. Data were analysed with SAS software in proc ANOVA and Duncan's multiple range test were used to compare treatments means. The results were showed this form: Mean ± SD (Standard Deviation)

Table1 Effects of digestible lysine levels on blood parameters

	DL (%)	Lys (nmol/ml)	Albumin (g/dl)	Uric Acid (mg/dl)	BUN (mg/dl)	Creatinine (mg/dl)
Male	0.68	70±1.10 ^e	0.90±0.02 ^c	15.10±0.07 ^f	6.61±0.07 ^e	0.78±0.00 ^c
	0.80	90±6.35 ^d	1.38±0.35 ^d	15.58±0.08 ^e	6.44±0.08 ^e	0.75±0.02 ^b
	0.92	140±7.34 ^c	1.70±0.00 ^c	16.21±0.09 ^d	5.66±0.17 ^d	0.73±0.01 ^b
	1.04	140±3.77 ^c	1.93±0.01 ^b	16.50±0.18 ^c	5.22±0.15 ^c	0.70±0.01 ^a
	1.16	150±5.23 ^b	2.40±0.01 ^a	17.11±0.17 ^a	4.00±0.20 ^b	0.71±0.01 ^a
	1.28	160±2.82 ^a	2.38±0.01 ^a	16.83±0.07 ^b	3.29±0.06 ^a	0.80±0.01 ^c
	Mean	125.01	1.78	16.22	5.20	0.75
Female	0.68	50±2.00 ^f	0.70±0.01 ^f	12.37±0.10 ^a	5.49±0.10 ^f	0.45±0.01 ^d
	0.80	70±1.09 ^e	1.12±0.01 ^c	12.72±0.17 ^b	5.10±0.14 ^e	0.41±0.01 ^c
	0.92	90±5.21 ^d	1.50±0.01 ^d	13.20±0.10 ^c	4.82±0.05 ^d	0.40±0.01 ^{bc}
	1.04	110±5.53 ^c	1.96±0.01 ^a	13.62±0.20 ^d	4.47±0.10 ^c	0.36±0.10 ^a
	1.16	120±4.12 ^b	1.90±0.00 ^c	14.10±0.12 ^e	4.00±0.10 ^b	0.49±0.01 ^e
	1.28	140±3.99 ^a	1.92±0.01 ^b	13.90±0.04 ^c	3.53±0.09 ^a	0.39±0.00 ^b
	Mean	96.67	1.52	13.32	4.57	0.42

Results Increase of digestible lysine level to 1.28% due to: maximum body weight, plasma lysine level, minimum feed conversion. There was the best of plasma albumin level and carcass N deposition in 1.16% and 1.04% of digestible lysine diet level for male and female respectively in the starter phases.

Table2 Effects of DE levels on feed intake, BW, FCR and carcass nitrogen

	DL (%)	Body Weight(g)	FCR	Feed Intake(g)	N.Carcass (g bird-1 day-1)
Male	0.68	370.71±6.54 ^d	2.08±0.03 ^c	772.92±17.21 ^d	0.86±0.01 ^c
	0.80	380.23±7.62 ^d	2.06±0.17 ^c	782.39±20.10 ^d	1.16±0.01 ^d
	0.92	415.27±12.91 ^c	1.75±0.01 ^d	726.74±23.82 ^c	1.16±0.01 ^d
	1.04	463.10±8.20 ^b	1.51±0.03 ^c	700.30±8.79 ^c	1.19±0.01 ^c
	1.16	485.58±8.14 ^a	1.31±0.03 ^b	637.42±21.50 ^b	1.24±0.02 ^a
	1.28	490.60±9.63 ^a	1.20±0.02 ^a	588.70±12.80 ^a	1.21±0.02 ^b
	Mean	434.25	1.65	701.41	1.37
Female	0.68	342.53±2.87 ^f	2.12±0.01 ^f	724.45±8.23 ^c	0.80±0.01 ^f
	0.80	358.30±8.56 ^e	2.01±0.01 ^e	717.54±18.17 ^c	0.92±0.01 ^e
	0.92	392.66±2.59 ^d	1.86±0.12 ^d	730.35±7.27 ^c	0.98±0.01 ^d
	1.04	405.77±8.47 ^c	1.60±0.02 ^c	649.32±1.73 ^b	1.20±0.01 ^a
	1.16	415.19±3.86 ^b	1.47±0.02 ^b	609.25±4.14 ^a	1.14±0.01 ^c
	1.28	426.02±3.59 ^a	1.41±0.14 ^a	600.68±6.97 ^a	1.16±0.01 ^b
	Mean	390.08	1.743	671.93	1.03

Conclusion This experiment supports the hypothesis that the different digestible lysine levels enhance protein utilization in broilers. All parameters as well were also improved with increase of digestible lysine levels and this suggests an effect of the amino acids utilization. Several possible mechanisms may account for the enhanced growth of chickens in response to additional dietary lysine. These include increased availability of lysine for protein synthesis, stimulated secretion of hormones such as glucagon, insulin and growth hormone which may consequently increase protein synthesis and feed intake.

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Dietary digestible lysine immune responses and carcass nitrogen of broiler chickens in starter period

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Introduction lysine is an essential amino acid that promotes normal growth by helping to maintain the proper protein balance in the body. Most vegetable protein sources used in poultry diet formulations are moderate to low in lysine contents; hence supplementation with lysine is inevitable in growing broilers to ensure rapid growth and optimum efficiency of feed utilization. Adequate supply of nutrients during the starter improves gut development and could result in long term improvements in feed utilization. In order to evaluate the effect of different digestible lysine dietary levels on growth and immune response in starter, this experiment was done.

Materials and methods 240 Arbor Acres broiler chicks (1-18 days) were used in a completely randomized design experiment (6 dietary, 4 replicates and 5 birds per each replicate). The levels of digestible lysine dietary were 0.68, 0.8, 0.92, 1.04, 1.16 and 1.28% (feeding practical diets formulating on digestible amino acids basis). All diets were formulated to meet NRC (1994) requirements and were formulated on a digestible amino acid basis with maintained ratios of dietary essential amino acids to lysine content. These diets were isoenergetic, isonitrogenous and equal in electrolyte balance (Na + K – CL). All diets met an ideal amino acid ratio recommended Illinois for all other amino acids. Feed and water supplied for *ad libitum* consumption. During the experiment feed conversion ratio (FCR) were measured weekly. Also, blood samples were collected to determine antibody titer (Titr), white blood cells(WBC), Lymphocyte (L), Hetrophyle (H), immune index (H/L) and the antibody response to a nonpathogenic antigen sheep red blood cell (SRBC) on day 18. A linear model and Duncan's mean test were used to analyze data by applying SAS software.

Results different digestible lysine levels had a significant effect on FCR and nitrogen deposition. Immune response as well was also improved with increase of digestible lysine levels and this suggests an effect of the on amino acids utilization.

Table 1 Effect of different digestible lysine levels on immune response, FCR and nitrogen deposition

	DL(%)	WBC($\times 10^3/\mu\text{l}$)	H(%)	H/L	Titr(log2)	SRBC(log2)	FCR	N Deposition
Male	0.68	14300 \pm 173 ^f	15 \pm 2.24 ^f	0.37 \pm 0.04 ^f	1.50 \pm 0.01 ^f	1.54 \pm 0.06 ^d	2.08 \pm 0.03 ^e	0.86 \pm 0.01 ^e
	0.8	15200 \pm 189 ^e	19 \pm 1.41 ^e	0.42 \pm 0.02 ^e	1.92 \pm 0.03 ^e	1.70 \pm 0.06 ^c	2.06 \pm 0.17 ^e	1.16 \pm 0.01 ^d
	0.92	19000 \pm 124 ^d	22 \pm 0.43 ^d	0.47 \pm 0.01 ^d	2.52 \pm 0.02 ^d	1.78 \pm 0.05 ^c	1.75 \pm 0.01 ^d	1.16 \pm 0.01 ^d
	1.04	25900 \pm 90 ^c	27 \pm 0.36 ^c	0.58 \pm 0.01 ^c	2.58 \pm 0.02 ^c	2.10 \pm 0.11 ^b	1.51 \pm 0.03 ^c	1.19 \pm 0.01 ^c
	1.16	28000 \pm 184 ^b	33 \pm 1.07 ^b	0.62 \pm 0.02 ^b	2.74 \pm 0.03 ^b	3.12 \pm 0.11 ^a	1.31 \pm 0.03 ^b	1.24 \pm 0.02 ^a
	1.28	41300 \pm 160 ^a	39 \pm 1.27 ^a	0.69 \pm 0.02 ^a	2.88 \pm 0.01 ^a	3.22 \pm 0.10 ^a	1.20 \pm 0.02 ^a	1.21 \pm 0.02 ^b
	Mean	23950	25.83	0.52	2.36	2.24	1.65	1.37
	0.68	16800 \pm 195 ^f	17 \pm 1.18 ^f	0.38 \pm 0.02 ^f	1.45 \pm 0.01 ^f	1.50 \pm 0.06 ^e	2.12 \pm 0.01 ^f	0.80 \pm 0.01 ^f
Female	0.8	19600 \pm 259 ^e	20 \pm 1.18 ^e	0.41 \pm 0.01 ^e	1.70 \pm 0.01 ^e	1.66 \pm 0.06 ^d	2.01 \pm 0.01 ^e	0.92 \pm 0.01 ^e
	0.92	23800 \pm 139 ^d	26 \pm 0.63 ^d	0.52 \pm 0.01 ^d	2.08 \pm 0.01 ^d	1.73 \pm 0.05 ^d	1.86 \pm 0.12 ^d	0.98 \pm 0.01 ^d
	1.04	26600 \pm 135 ^c	30 \pm 0.67 ^c	0.59 \pm 0.02 ^c	2.56 \pm 0.03 ^c	1.94 \pm 0.11 ^c	1.60 \pm 0.02 ^c	1.20 \pm 0.01 ^a
	1.16	32200 \pm 43 ^b	35 \pm 0.76 ^b	0.66 \pm 0.01 ^b	2.74 \pm 0.02 ^b	2.27 \pm 0.11 ^b	1.47 \pm 0.02 ^b	1.14 \pm 0.01 ^c
	1.28	44100 \pm 211 ^a	42 \pm 1.52 ^a	0.73 \pm 0.01 ^a	2.78 \pm 0.03 ^a	2.40 \pm 0.11 ^a	1.41 \pm 0.14 ^a	1.16 \pm 0.01 ^b
	Mean	27183.33	28.33	0.55	2.22	1.92	1.743	1.03

Conclusion The results of this experiment support the hypothesis that the different digestible lysine levels enhance protein utilization in broilers. The influence of Lys was probably mediated by IGF-I(Conconi,2001) High immune response is possibly due to increased protein availability for liver protein synthesis associated with immune response or antibody production. In the present study, increasing the dietary digestible lysine levels from 0.68 to 1.28% of the NRC-recommended requirement significantly increased immune response and nitrogen retention. These results concur with those previously obtained from chickens (Bons *et al.*, 2002). Supplementation of L-lysine also increased their antibody responses, with increasing circulating lymphocytes, monocytes, neutrophils, and humoral response. Understanding the nuances of nutrition and immunity is important for optimizing bird health and productivity, and will be an important contributor towards fulfilling the consumer's conflicting demands for more natural production and better animal welfare. Immune function, especially of lymphocytes, should be monitored when conducting experiments to determine nutrient requirements. Least-cost diets are not usually optimal for immunity because they deliver too much energy and are marginal in some nutrients. Experiments to date indicate that concentrations of some nutrients that give optimal growth and efficiency of feed utilisation are inadequate for immunity. Among amino acids, the dietary concentrations that support maximal growth performance appear to be adequate for immunity for lysine, arginine, isoleucine and valine; but sulphur amino acids may be an exception (Klasing, 2007).

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Effect of prebiotic on performance of broiler chicks in low protein diets

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Introduction In view of the severe restriction or total ban on the use of antibiotics as growth promoters and therapeutic agents in poultry industry, probiotics and prebiotics have been suggested as alternatives to antibiotics (piray *et al*, 2007). Prebiotic are known as "a nondigestible feed ingredient" that beneficially affects the host by selectively stimulating the growth or activity of a limited number of bacteria in the colon (Gibson and Roberfroid, 1995). Prebiotics has a significant effect on body weight gain and feed to gain ratio (piray *et al*, 2007). The objective of this research was to study the effectiveness of adding prebiotic on broiler growth performance in low protein diets.

Materials and methods Two hundred forty day of hatch mixed sex broiler chickens of the Ross strain-308 were housed on litter-floor and allocated randomly to each of 6 treatments with 4 replicates (pens) each in CRD design. The environmental condition was according to the Ross guide book. All groups received starter diet contains 23% crude protein from days 1-10. Three groups received an experimental diet formulated to meet Ross guide nutrient requirements for crude protein (21% in grower diet from days 11-28 and 19% in finisher diet from days 29-42) and other nutrients with three levels of (0.0, 1.5 and 3.0 g /kg into the basal diets) Aspergillus meal prebiotic (Fermacto). It is the feed additive, derived from Aspergillus mycelium. The other 3 groups received a diet deficient in crude protein (19% in grower diet from days 11-28 and 17% in finisher diet from days 29-42) with the same three level of prebiotic. Feed and water were provided ad- libitum. Feed intake and body weight gain of chickens were recorded weekly and feed to gain ratio calculated as the unit of ate feed per unit of body weight gain (g/g). GLM proc of SAS was used for statistical analysis and least square means.

Results In finisher period (29-42 days) supplementation of Fermacto increased feed intake only in diet containing standard protein with 3 g/Kg Fermacto. However, difference between diet containing standard protein with 3 g/Kg Fermacto and three low protein diets was significant ($p < 0.05$). Addition of Fermacto into low protein diets did not improved feed intake in comparison to control. Addition of Fermacto into both standard and low protein diets increased weight gain but differences were not significant. Among low protein diets, highest value of weight gain observed in diet with 3 g/Kg Fermacto. Chicks fed low protein diet with 3 g/kg Fermacto did not show significant difference in weight gain in comparison to standard protein diets. Chicks fed low protein diet with 3 g/kg Fermacto had comparable feed conversion ratio (FCR) value with standard protein diets. Using Fermacto in standard protein diet had not significant effect on FCR but addition of 3 g/Kg Fermacto into low protein diet improved FCR significantly ($p < 0.05$).

Table 1 Mean feed intake, weight gain and FCR in chickens fed two levels of protein with three levels of Fermacto in finisher period.

Diets	Feed intake, g	weight Gain, g	FCR	Body weight, g
SP ¹ + 0 g/Kg Fermacto(control)	2000 ^{ab3}	1126 ^{abc}	1.78 ^{bcd}	2711 ^{ab}
SP+1.5 g/Kg Fermacto	1958 ^{bc}	11277 ^{ab}	1.74 ^{cde}	2614 ^{bc}
SP+ 3 g/Kg Fermacto	2060 ^a	11583 ^a	1.78 ^{bc}	2782 ^a
LP ² + 0 g/Kg Fermacto	1943 ^{bcd}	1026 ^{de}	1.90 ^a	2533 ^c
LP+1.5 g/Kg Fermacto	1946 ^{bcd}	1066 ^{bcd}	1.83 ^{ab}	2589 ^{bc}
LP+ 3 g/Kg Fermacto	1956 ^{bcd}	1101 ^{abcd}	1.78 ^{bcd}	2604 ^{bc}
SEM	0.0295	0.0271	0.0264	54

¹ Standard protein ² Low protein

³Means in each column with different superscripts differ statistically ($p < 0.05$).

Conclusions Generally, using Fermacto at high level (3g/kg) in standard and low protein diets improved weight gain and FCR of chicks numerically and statistically respectively. FCR and weight gain values were more affected in low protein diets containing Fermacto. This findings show potential using of Fermacto in some countries which could not formulate diets at protein level recommended by Ross company.

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Effect of probiotic on lipid profile of broiler chicks

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Introduction Research on poultry genetics, feeding and management for BWG and FCR resulted in fast growth but decreased the quality of poultry products as modern fast growing broilers have been found to contain higher amount of abdominal fat (Chambers *et al*, 1981). Most recently considerable attention has been paid to test the potency of growth promo ants on altering lipid metabolism, because, World Health Organization suggest that excess fat deposition is undesirable in human body. Recent report suggested that feeding of chicory beta fructans; a probiotic reduced the serum cholesterol and abdominal fat of broiler chicken. The present study was undertaken to study the effect of Aspergillus meal probiotic (Fermacto) on abdominal fat, serum total cholesterol, HDL and triglyceride levels in low protein diets.

Materials and methods Two hundred forty day of hatch mixed sex broiler chickens of the Ross strain-308 were housed on litter-floor and allocated randomly to each of 6 treatments with 4 replicates (pens) each in CRD design. Three groups received an experimental diet formulated to meet Ross guide nutrient requirements for crude protein (21% in grower diet from days 11-28 and 19% in finisher diet from days 29-42) and other nutrients with three level of (0.0, 1.5 and 3.0 g/kg into the basal diets) Fermacto. Fermacto is the feed additive, derived from Aspergillus mycelium. The other 3 groups received a diet deficient in crude protein (19% in grower diet from days 11-28 and 17% in finisher diet from days 29-42) with the same three level of Fermacto. A day prior to slaughter, blood samples were randomly collected via wing vein from 5 males of each treatment at 48 days of age. After slaughter, abdominal fat were measured in the dressed carcasses. Serum samples were analyzed for total cholesterol, HDL cholesterol and triglycerides in Imam Reza hospital biochemical lab. GLM proc of SAS was used for statistical analysis and obtaining least square means.

Results In recent study, supplementation of Fermacto decreased total cholesterol in diet containing standard protein with 1.5 g/kg Fermacto significantly ($p<0.05$). With addition of 1.5g/kg Fermacto to standard protein diet, the cholesterol value decreased from 148 to 125.8 mg/dl. However, in the diets containing low protein, addition of Fermacto had no effect on cholesterol level in comparison to control diet. Triglycerides level did not affected by supplementation of Fermacto in diets containing low protein. However, supplementation 3 g/kg Fermacto in standard protein diet decreased triglycerides level in comparison to control diet ($p<0.05$). Decreasing of HDL observed only in diet containing standard protein with 1.5 g/kg Fermacto in comparison to control diet ($p<0.05$), but in other treats did not show significantly differences. These observation show that serum lipids affected by Fermacto in standard protein diets but not in low protein ones. There was decreasing trend in percentage of abdominal fat pad with addition of Fermacto to diets containing standard protein. However, the diets containing low protein, addition of Fermacto with 1.5 g/kg decreased significantly ($p<0.05$) abdominal fat pad in comparison to diet containing low protein without Fermacto.

Table 1 Serum lipid and abdominal fat(% of live weight) content of broiler fed different levels of protein and Fermacto

Diets	Total Cholesterol(mg/dl)	Triglycerides(mg/dl)	HDL(mg/dl)	Abdominal fat	Body weight, g
SP ¹ + 0 g/Kg Fermacto(control)	148 ^{a3}	147.2 ^a	118.8 ^a	2.58 ^{ab}	2711 ^{ab}
SP+1.5 g/Kg Fermacto	125.8 ^c	133 ^{abcde}	108 ^{cde}	2.25 ^{abcd}	2614 ^{bc}
SP+ 3 g/Kg Fermacto	135.4 ^{bcde}	126.2 ^{bcde}	117 ^{abc}	2.14 ^{bcde}	2782 ^a
LP ² + 0 g/Kg Fermacto	138.6 ^{abcd}	145.6 ^{ab}	113 ^{abcd}	2.80 ^a	2533 ^c
LP+1.5 g/Kg Fermacto	140 ^{abc}	142 ^{abcd}	112 ^{abcde}	2.12 ^{bdce}	2589 ^{bc}
LP+ 3 g/Kg Fermacto	140.2 ^{ab}	143.4 ^{abc}	118 ^{ab}	2.36 ^{abc}	2604 ^{bc}
SEM	3.9476	6.8946	3.2660	0.2193	54

¹ Standard protein ² Low protein ³Means in each column with different superscripts differ statistically ($p<0.05$).

Conclusions supplementation of Fermacto decreased total cholesterol and abdominal fat pad in broilers fed standard protein diet but at low protein diet, just abdominal fat pad was decreased by Fermacto supplementation; therefore inclusion level of Fermacto and level of protein were important for effectiveness. Use of Fermacto was one of the biological ways to improve economic burden in poultry production with decreasing abdominal fat percentage and improve human health by decreasing cholesterol level in serum.

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Ileal crude protein digestibility, caecal crude protein retention and digesta viscosity in broilers fed enzyme supplemented rice husk

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Introduction Enhanced nutrient digestion and absorption in enzyme supplemented broiler diets could be either through reduced digesta viscosity or weakening of the cage effect elicited by soluble or insoluble Non – Starch – Polysaccharides (NSP) on nutrient digestion and absorption. Inhibitory effect of insoluble NSP on endogenous protease reduces efficient digestion of nutrients embedded in cell wall matrix of grains. Increased amount of undigested and unabsorbed protein reaching the caecum implies available nutrient for fermentation and increased coliform forming units of such bacteria as *clostridium* that ferment proteins, peptides and amino acids. The study aimed to assess the effect of partial replacement of maize with Rice Husk (RH) supplemented with Roxazyme G2 G enzyme on ileal crude protein digestibility, caecal crude protein retention and digesta viscosity in broilers.

Materials and methods Four diets were formulated consisting of a Maize – Soya bean meal (M/SBM) control and 3 treatment diets in which maize was partially replaced with 15 (75g/kg of diet), 30 (150g/kg of diet) and 45% (225g/kg of diet) RH. The RH based diets were supplemented with Roxazyme G2 G at the rate of 200g/tonne. Titanium dioxide (TiO₂) was added as an indigestible marker. Free access to experimental diets was provided to 48 Ross unsexed broilers from day 7 to 35 post hatch. The birds were distributed into 48 cages of 1 bird per cage with two cages representing a replicate. A group of 12 cages were then randomly assigned to each of the experimental diets. The birds were reared in a well ventilated temperature controlled house. Digesta was collected from the ileum (2cm posterior to Merkel's diverticulum and 2cm anterior to the ileal – caecal – colonic junction). Digesta collection, collection of digesta liquid fraction and determination of digesta viscosity was done by the method of Steinfeldt *et al* (1998) with the exception of type of viscometer used. A Roto Visco 1 from HAAKE (Germany) was used instead of the Brookfield type. TiO₂ analysis was determined using the method of Brandt and Allam (1987). Ileal crude protein digestibility coefficient was calculated using the formula as given below. The experiment was arranged as complete randomized design of 6 replicates and 2 birds per replicate. All data collected were subjected to polynomial regression procedure in SAS.

$$\text{Ileal crude protein digestibility coefficient} = 1 - \frac{(\text{conc. of TiO}_2 \text{ in feed} \times \text{conc. of nutrient in digesta})}{(\text{conc. of TiO}_2 \text{ in digesta} \times \text{conc. of nutrient in feed})}$$

Results Ileal crude protein digestibility, caecal crude protein retention (unabsorbed protein leaving the ileum less the protein voided in faeces) and digesta viscosity were similar across dietary treatments. Low digesta viscosity (1.71mPa.s) was recorded in broilers fed maize-soya bean meal control diet, with corresponding low caecal crude protein retention (2.55g/kgDM) and a high ileal crude protein digestibility (0.989). A similar trend was observed in broilers fed 15 (2.06mPa.s, 3.62g/kgDM and 0.984), 30 (1.85mPa.s, 2.70g/kgDM and 0.988) and 45% (1.86mPa.s, 3.59g/kgDM and 0.986) enzyme supplemented rice husk-based diets.

Table 1 Enzyme effect on ileal crude protein digestibility, caecal crude protein retention and digesta viscosity in broilers

Parameters	M/SBM	M/SBM+ 75g/kg RH	M/SBM+ 150g/kg RH	M/SBM+ 225g/kg RH	SEM	P value
	Ileal CP digestibility	0.989	0.984	0.988		
Caecal CP retention (g/kgDM)	2.55	3.62	2.70	3.59	0.40	0.161
Digesta viscosity (mPa.s)	1.710	2.060	1.850	1.860	0.17	0.175

mPa.s: millipascal seconds, CP: crude protein

Conclusion The results show a relationship between ileal crude protein digestibility, caecal crude protein retention and digesta viscosity. The amount of protein reaching the caecum is dependent on the amount absorbed in the ileum which can be influenced by ileal digesta viscosity. The inhibitory effect of high amount of insoluble NSP present in rice husk on endogenous protease could have been minimized by the enzyme utilized via disruption of cell wall matrix.

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Immunomodulatory effects of dietary *Allium cepa* in chicken after immunization

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Introduction Although antibiotics treatment has been used as an efficient technique to control infectious diseases in poultry industry, there are intensive studies to decrease the use of antibiotics because of the increase in microbial resistance. Therefore, enhancing protective immunity with immunostimulators by magnifying the capacity of immune response seems to be the most promising and practical approach. *Alliums* have been reported to possess therapeutic properties often attributed to sulphur-containing compounds (Bonaccorsi *et al.*, 2005). In addition to these compounds, *Allium cepa* is among the richest dietary flavonoids sources; those flavonoids have shown in mammals *in vitro* antimicrobial and anticarcinogenic activities (Block *et al.*, 1992). However, very few studies aimed at examining their effects in poultry have been performed. The objective of this study is to examine if the addition of supplementary *Allium cepa* to a balanced ration enhances the immune response of chickens after immunization.

Materials and methods One-day-old, White Leghorn male chicks were distributed into 3 groups of 8 each. The birds were housed in wire cages, 24 hr light-program and 24°C. After the first week, birds were fed on diets containing powder of *Allium cepa* with 10 or 30 gm/kg of the basal starter diet. Free access to water and feed was allowed. Feed intake and body weight were recorded for growth performance study. On day 14, chickens were immunized intraocularly with Newcastle disease vaccine (NDV-clone 30), and intravenously with *Brucella abortus* (BA), and repeated 14 days later. Blood samples were drawn from wing vein weekly for antibodies determination. Anti-NDV antibody was determined by hemagglutination inhibition (HI) titre whereas anti-BA antibody was determined by agglutination titre. Ratios of CD4⁺ and CD8⁺ T-lymphocytes, and CD4⁺CD8⁺ lymphocytes (mostly B-lymphocytes) in splenocytes were studied. Thus, splenocytes were incubated with FITC-labelled anti-chicken CD4 antibody and PE-labelled anti-chicken CD8 antibody; ratios were determined using flow cytometry. To check *in vitro* co-mitogenic properties of *Allium cepa* extract on B-lymphocyte proliferation, bursocytes were resuspended in RPMI-1640 media to 1×10⁶ cell/ml, and B-lymphocytes proliferation was induced by phorbol 12-myristate 13-acetate (PMA). After 48 hr incubation (39.5°C and 5% CO₂), 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) was added, and finally, light absorbance was measured at 570 nm wave length as described by Bao (1998) with some modifications. Proliferation was expressed as stimulation index (SI = proliferation of B-lymphocytes in culture with PMA/proliferation without PMA). Data were analyzed using single factor ANOVA of Microsoft Excel. P<0.05 was considered significant for all analysis.

Results Average feed conversion ratios of *Allium cepa*-fed chickens had values of 2.15 for low concentration and 2.17 for high concentration which were significantly improved (P<0.01, P<0.001, respectively) compared to control (2.70). Feeding low concentration of *Allium cepa* exerted a significant increase in anti-NDV antibody production started in response to primary immunization (Figure 1A). Anti-BA antibody titre was higher with dietary *Allium cepa* in response to secondary immunization; however, low concentration had stronger and prolonged stimulatory effect (Figure 1B). Supplementary high concentration of *Allium cepa* significantly increased the ratio of CD4⁺CD8⁺ cells (48.6%, P<0.001), while decreased that of CD4⁺ (21.0%, P<0.05) compared with control (43.0% and 25.6%, respectively). Adding 1.3 ~ 6.5 µg/ml as a final concentration of *Allium cepa* extract had a higher (P<0.001) SI of 2.52 compared with that of PMA only (1.86).

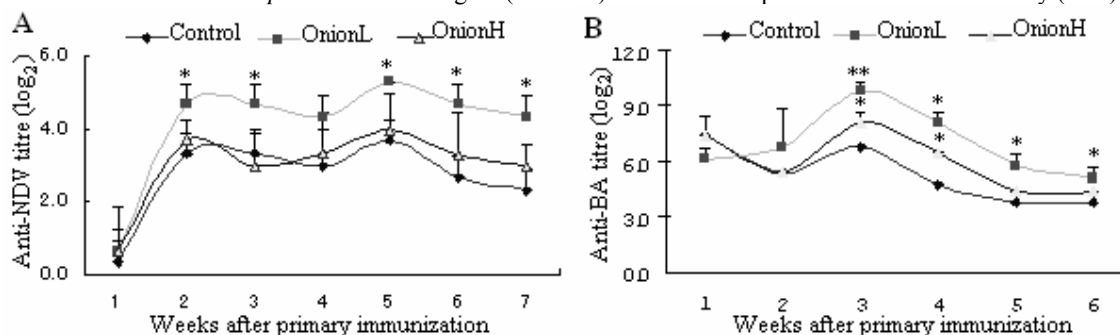


Figure 1 Effect of dietary *Allium cepa* (L, 10 or H, 30 gm/kg diet) on anti-NDV (A) and anti-BA (B) antibody titres (log₂) in chicken. (*P<0.05, **P<0.01)

Conclusion The results of this study clearly showed that dietary supplementation of White Leghorn-type chicken with *Allium cepa* stimulated humoral immune response and growth performance. As well, *Allium cepa* had *in vitro* co-mitogenic effect on PMA-induced B-lymphocytes proliferation.

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Nutritive value of poultry by-product meal from Iran in broiler feeding

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Introduction Recently, production of poultry by – product meal (PBPM) has been increased in the northwest of Iran, East Azarbijan. PBPM has a profile of available essential amino acids and is rich in calcium, phosphorus and vitamin B12 (NRC 1994). The chemical composition and mineral content can vary greatly depending on raw material and processing conditions. In our country, fish meal is a common ingredient as animal protein in poultry diets. However, fish meal is imported at high cost. In recent years, there have been attention paid to PBPM supplementation in diets by East Azarbijan poultry feed industry. Therefore, the present study was conducted to determine the chemical composition, mineral content and to study the effects of different dietary concentration (3, 6 and 9%) of PBPM on broiler performance.

Materials and methods The three composed PBPM samples were collected from rendering units of industrial poultry slaughter-house in East Azarbijan province and were chemically analyzed according to AOAC (1992) procedures. For feed evaluation 240 Ross-308 as hatched broiler chickens were used in this experiment. Four different treatments were formed in the study. First treatment was control with no PBPM. The other treatments were 3.0, 6.0 and 9.0% PBPM supplementation, respectively. Experimental diets were formulated as isonitrogenous and isocaloric (based on Ross308 management guide book) and fed from 22 to 49 days of age. Broiler chickens were assigned randomly to four treatment groups (per treatment/5 pens). Fifteen broilers were housed on litter-floor. Feed and water was available on an *ad libitum* basis. Broilers were weighed individually at 22, 42 and 49d of age. For each pen, feed consumption ratio (FCR) and weight gain were measured on a weekly basis. Feed intake was weighed back on the same day that body weights (BW) were determined. Mortality was not observed in this study. The data was analyzed in a completely randomized design using GLM procedure of SAS. Comparison of means was conducted by Duncan's multiple range tests.

Results The average of chemical composition and mineral contents of the PBPM samples are shown in Table 1, and the effects of supplementation PBPM to broiler diets on various live performances is shown in Table 2. Feed intake and weight gain in first stage of experiment with increasing inclusion level of poultry by- product meal, were decreased ($p<0.05$). But feed intake and weight gain in second stage of experiment due to adaptation were not significant difference with control birds ($P>0.05$). Weight gain in total experiment period was less than control birds ($p<0.05$). Feed conversion ratio in two stage and total experiment period were similar to standard feed conversion ratio in Ross308 guide book and non significant difference with control bird ($P>0.05$).

Table 1 Composition of PBPM samples (% as fed)

Composition	DM	CP	EE	TVN*	Ca	P	Na	K	Cu	Zn	Mn	Fe
Average	95.5	62.12	25.28	5.6	1.3	0.43	0.61	0.53	0.061	0.173	0.042	0.821

*Total volatile nitrogen

Table 2 The effects of PBPM inclusion in broiler diets at different levels on growth performance

Treatments	22-42d				43- 49 d			22-49d		
	Feed Intake (gr)	Weight Gain (gr)	Body Weight (gr)	FCR	Feed Intake (gr)	Weight Gain (gr)	FCR	Feed Intake (gr)	Weight Gain (gr)	FCR
Control	2398 ^a	1390 ^a	1919 ^a	1.72 ^a	1159 ^a	570 ^a	2.04 ^a	4037 ^a	2281 ^a	1.76 ^a
3%	2278 ^b	1346 ^a	1800 ^b	1.69 ^a	1118 ^a	552 ^a	2.02 ^a	3875 ^a	2201 ^b	1.76 ^a
6%	2170 ^c	1269 ^b	1664 ^c	1.71 ^a	1172 ^a	599 ^a	1.96 ^a	3785 ^b	2134 ^b	1.77 ^a
9%	2066 ^d	1177 ^c	1561 ^d	1.75 ^a	1133 ^a	569 ^a	2.00 ^a	3629 ^c	1984 ^c	1.82 ^a
SEM	28.7	22.2	26.5	0.025	33.2	16.4	0.08	44.2	22.7	0.025

In the same column differently superscripted are significantly ($P<0.05$) different.

Conclusions In summary, the results showed that composition of studied PBPM samples were different from NRC (1994). Also, under the conditions of this study, using of different levels of poultry by product meal (3, 6 and 9 percent) did not show any detrimental effect on feed conversion ratio but depressed broiler growth rate age of 22 to 49 days.

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Effects of addition of Natuzyme® to broiler diets containing different levels of canola meal

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Introduction Canola meal (CM) is a suitable protein source in poultry diets, although it contains a number of antinutritive factors including non-starch polysaccharides and phytic acid. These factors can limit inclusion rate of canola meal in poultry diets (Kochoer *et al.*, 2000). The negative correlation between NSP levels and nutritive value of the diet has been demonstrated in poultry (Choct and Annison, 1990). In addition to reducing the availability of phosphorus to birds, phytates are also associated with a number of antinutritional effects, largely because they can chelate divalent cations and reduce protein availability (Ravindran *et al.*, 1995). Successful use of enzymes in cereal-based diets has stimulated interest in the application of enzymes to target the vegetable protein components of poultry diets as well. The current study was, therefore, conducted to investigate the effects of a multi-enzyme on performance and serum thyroid hormone concentrations of broilers fed rations in which soybean meal was replaced by CM.

Materials and methods Eight hundred day-old broiler chicks (Cobb) were randomly allocated to 8 dietary treatments in a 4 x 2 factorial arrangement with 5 replicates per treatment and 20 birds per replicate. Experimental diets had different levels of canola meal (0, 100, 200 and 300 g/kg), with or without enzyme (Natuzyme, Sunnybank, Brisbane, Australia). The supplemental enzyme activities reported by the manufacturer were cellulase 6000 units/g, xylanase 10,000 units/g, amylase 700 units/g, glucoamylase 700 units/g, phytase 500 units/g, pectinase 70 units/g, proteases 3000 units/g and lipase 30 units/g. The diets were isoenergetic and isonitrogenous and were provided *ad libitum* throughout a 42 d experiment. Feed intake and weight gain were determined and feed conversion ratio was calculated. At 42 d age, 2 birds were randomly selected from each replicate and blood samples were collected for measuring T3 and T4 concentrations. T3 and T4 concentrations in the sera were determined by RIA according to the procedure of Kloss *et al.* (1994). Data were subjected to the GLM procedure for ANOVA (SAS, 2001). Mean separation was accomplished using Duncan's multiple range tests.

Table 1 Effects of canola meal and multi enzyme on performance and serum thyroid hormone levels of broilers

Item		1-21 d			1-42 d			Serum T3 (µg/ml)	Serum T4 (µg/dl)
		Feed intake(g)	BW gain (g)	Feed:gain (g/g)	Feed intake(g)	BW gain (g)	Feed:gain (g/g)		
Enzyme	CM								
No	0	924 ^b	688 ^{ab}	1.34 ^c	3725	2221 ^{ab}	1.68 ^{bc}	1.33	0.63
	10	921 ^b	606 ^c	1.52 ^{ab}	3726	2075 ^{bc}	1.79 ^{ab}	1.21	1.36
	20	952 ^{ab}	653 ^{abc}	1.46 ^{abc}	3711	2039 ^{bc}	1.82 ^a	1.33	0.4
	30	969 ^{ab}	618 ^{bc}	1.57 ^a	3696	1935 ^c	1.91 ^a	1.36	0.83
Yes	0	965 ^{ab}	710 ^a	1.36 ^{ab}	3690	2268 ^a	1.62 ^c	1.37	1.44
	10	991 ^a	637 ^{abc}	1.55 ^a	3707	2104 ^{ab}	1.76 ^{abc}	1.41	0.64
	20	971 ^{ab}	640 ^{abc}	1.51 ^{abc}	3705	2103 ^b	1.75 ^{abc}	1.25	0.48
	30	956 ^{ab}	643 ^{abc}	1.48 ^{abc}	3681	2087 ^{bc}	1.76 ^{abc}	1.24	0.66
S.O.V									
Enzyme	*	ns	ns	ns	ns	*	*	ns	ns
CM	ns	**	*	ns	ns	**	*	ns	ns
Enzyme × CM	*	*	*	ns	ns	*	*	ns	ns
SEM		14.6	16.2	0.02	37.1	35.3	0.03	0.06	0.23

^{a-c}Means with different superscripts are significantly different. ^{ns}non-significant *P<0.05 **P<0.01

Results Canola meal had no effect on feed intake. Body weight gain was significantly reduced when canola meal was added into the diets (P<0.01). No effect of enzyme addition was observed on BW gain and feed:gain during 1 to 21 d. Enzyme addition significantly increased body weight gain during 1-42 d (P<0.05). Feed conversion ratio was highest from 1 to 42 days when 30 g/kg canola meal was added to the diets. Enzyme supplementation significantly (P<0.05) improved feed conversion ratio during 1- 42 d. The interactions between enzyme and canola meal for body weight gain and FCR were significant. The effect of enzyme addition on weight gain and feed:gain was numerically greater in diets which had higher levels of CM. Different levels of added canola meal and enzyme had no significant effect on concentrations of serum thyroid hormones.

Conclusions The results of this study showed that inclusion of CM at or above 100 g/kg depressed weight gain and FCR without affecting the feed intake. Enzyme supplementation had the potential to reduce the unfavourable effects of canola meal.

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The effects of multi-enzyme addition on performance of broiler chicks

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Introduction Today the use of enzymes is common in practical poultry nutrition. Enzyme supplementation usually result in numerous beneficial effects, such as increased utilization of nutrients (e.g. fat & protein), improved AME values, increased growth rate, improved feed conversion ratio, decreased viscosity of intestinal digesta, reduced incidence of sticky excreta and improved litter conditions (Broz and Ward., 2007). The use of multi enzymes to improve the digestibility of corn-soybean meal-wheat diets for broilers is less well documented than wheat and barely diets. Therefore, the aim of the present study was to examine the effects of a multi-enzyme supplementation on the performance of broiler chicks fed on corn-soybean meal-wheat diets.

Materials and methods A total of 150 one-day-old mixed-sex broiler chicks (Cobb 500) were used in a completely randomized design with 3 replicates with 25 chicks in each replicate. The birds were randomly allocated to 6 pens. The main ingredients of diets included corn, soybean meal and wheat. The amount of corn, soybean meal and wheat in starter (1-10 d) and grower (11-28 d) diets were about 437, 356, 150 and 374, 314, 250 g/Kg diet respectively. Diets were formulated according to Cobb 500 rearing guideline and contained 2950 Kcal/Kg metabolizable energy and 22% crude protein during starter, and 3000 Kcal/Kg metabolizable energy and 21% crude protein during grower period. Feed and water were provided *ad libitum* during the experiment. Temperature was maintained at 32° C for the initial 3 d and then gradually reduced according to normal management practices, until a temperature of 22° C was achieved. The experiment lasted 4 weeks. The experimental diets contained 2 levels of a dietary NSP degrading enzyme (0, 0.05%; Endofeed W produced from *Aspergillus niger*, with minimum activity of 2250 u/g xylanase and 700 u/g Beta-glucanase). According to the manufacturer, the allowance for this enzyme product was 0.05% and it also contained activities of other enzymes, including cellulase, protease, α -amylase and α -galactosidase. The enzyme complex was added to the chicken diet as a dry powder from hatching to the end of the experiment. Chickens were weighed weekly (from 1 to 28 d) to determine their performance. Data were analyzed by analysis of variance using the GLM procedure (SAS institute, 2001). Differences among means were compared by Duncan's multiple range test (1955).

Results Data obtained from this experiment indicated that enzyme supplementation significantly improved body weight gain and feed conversion ratio (Table 1). These results are in agreement with the findings of previous studies (Wang *et al.*, 2005; Gao *et al.*, 2007). Adding enzyme to broiler diets may improve performance by two mechanisms: increasing feed intake and improving nutrient digestibility. Both mechanisms might be induced, at least partially, by reduction of gut viscosity which decreased retention time of digesta in the gut, allowing more feed consumption and therefore improving growth and feed conversion ratio (Lázaro *et al.*, 2003). The results showed that enzyme supplementation to corn-soybean meal-wheat based diet can improve weight gain whilst maintaining intake, which is suggestive of improved digestibility of a limiting nutrient.

Table 1 Effect of multi-enzyme supplementation on broilers performance during 1 to 28 d of age

Diets	Feed intake (g)	Weight gain (g)	FCR (g:g)	Energy efficiency ratio ¹ (%)	Protein efficiency ratio ² (g:g)	Growth ratio ³
1 (without enzyme)	2094.44	1389.95 ^b	1.51 ^a	22.10 ^b	3.142 ^b	34.74 ^b
2 (with enzyme)	2095.05	1435.68 ^a	1.46 ^b	22.80 ^a	3.245 ^a	35.89 ^a
s.e.m	13.05	7.13	0.008	0.13	0.016	0.18

^{ab} Means with different superscripts in each column are significantly different (P<0.05)

¹Calculated as weight gain×100 divided by total Kcal ME intake. ²Calculated as weight gain divided by protein intake.

³Calculated as BW at 28 d of age – BW at 1 d of age / BW at 1 d of age.

Conclusions Results of the present study indicated that supplementation of enzyme at the level of 50 mg/Kg diet significantly improved feed conversion, energy and protein efficiency and growth ratio of Cobb 500 broiler chicks during starter and grower period of rearing (1 to 28 d of age).

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Enrichment with long chain omega-3 fatty acids and sensory evaluation of chicken meat

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Introduction N-3 fatty acids are essential for normal growth and development, and may play an important role in prevention of coronary artery disease, hypertension, diabetes, arthritis, other inflammatory and autoimmune disorders and cancer in humans (Simopoulos, 1999). Fatty acid profiles of broiler meat may be modified by adding fish oils to the diet (Lopez-Ferrer *et al.*, 2001). When meat is enriched with PUFA, particularly n-3 long-chain fatty acids (C_≥20), all sources of added vegetable oils seem to be less effective than marine oils (Bou. R *et al.*, 2004). The purpose of this experiment was to study the effect of dietary fish oil on fatty acid composition of thigh and breast meat in broiler chickens.

Materials and methods Four hundred and fifty d-old Ross 308 male, broiler chickens were allocated to 6 dietary treatment with 5 replicates of 15 birds each. The birds were reared in a controlled environment house and had *ad libitum* access to water and feed. All dietary nutrients were provided as to meet AVIAGEN recommendations. Khazar Kilka fish oil was added at the levels of 0, 10, 20, 30, 40 and 50 g/kg in diets, fed from 28 to 42d of age. Performance criteria were measured during the experimental period. One bird with an average live body weight of each replicate group was selected and slaughtered on day 42 d The left thigh and left side of the breast were excised and stored at -20 C for later analysis. Samples (including skin) were analyzed for n-3 fatty acids with GC and sensory evaluation after each sample was individually cooked in boiling water. Data were analysed by using GLM procedure of SAS (9.1)

Results Weight gain and Feed Conversion Ratios were similar (P>0.05) for all treatments (table1). The Eicosapentaenoic acid (EPA) and Docosahexaenoic acid (DHA) contents of thigh or breast tissues were increased significantly (P<0.05) as the level of dietary fish oil increased. Effects were not significant for LA(linoleic acid) and ALA(alpha linolenic acid). Sensory evaluation showed that panellists did not identify the fishy smell of cooked thigh or breast meat of birds fed diets with 0, 10 or 20 g/kg fish oil. The concentration of DHA in breast and thigh meat increased from 0.046 (mg/g) and 0.086 (mg/g) to 0.166 (mg/g) and 0.27 (mg/g), respectively, when dietary fish oil increased from 0 to 20 g/kg,. Significant linear relationships were found between the levels of fish oil and EPA in the breast and thigh (R² = 0.59 ; R²= 0.71 respectively) and between the fish oil and DHA in the breast and thigh (R² = 0.71 ; R² = 0.72, respectively).

Table 1 Broiler chickens performance and concentration of some n-3 FAs in breast and thigh meat (mg/g in meat) of birds given diets including fish oil

Treatments	WG (g/b/d)	FCR	Thigh				Breast				Scores ¹	
			C18:2	C18:3	C20:5	C22:6	C18:2	C18:3	C20:5	C22:6	Thigh	Breast
control	69.8	2.24	5.4	0.35	0.028 ^b	0.086 ^c	0.54 ^b	0.02b	0.014 ^b	0.046 ^c	1.10	1.12
1% fish oil	71.4	2.27	6.2	0.13	0.051 ^b	0.16 ^{bc}	0.87 ^{ab}	0.04 ^{ab}	0.042 ^b	0.145 ^{bc}	1.27	1.17
2% fish oil	69.2	2.48	6.1	0.42	0.086 ^{ab}	0.27 ^{bc}	0.99 ^{ab}	0.10 ^a	0.04 ^b	0.166 ^{bc}	1.25	1.25
3% fish oil	72.1	2.29	4.6	0.32	0.16 ^{ab}	0.38 ^{ab}	0.67 ^{ab}	0.03 ^{ab}	0.04 ^b	0.240 ^{ab}	2.32	2.25
4% fish oil	67.6	2.53	4.2	0.43	0.23 ^a	0.53 ^a	0.99 ^{ab}	0.07 ^{ab}	0.09 ^a	0.340 ^a	3.25	3.87
5% fish oil	68.0	2.44	3.2	0.30	0.16 ^{ab}	0.58 ^a	1.10 ^a	0.06 ^{ab}	0.08 ^a	0.290 ^a	4.02	4.25
SEM	1.78	0.04	0.09	0.11	0.05	0.08	0.16	0.04	0.01	0.04		
P value	NS	NS	NS	NS	0.003	0.003	NS	NS	0.006	0.001		

^{abc} Values in column with no common superscripts differs significantly (P<0.05)

¹ Flavour scores using a 5 point scale: 5=very poor,4=poor, 3=indifferent, 2=acceptable, 1=typical chicken flavour

Conclusion The levels of long chain fatty acids (EPA and DHA) were linearly increased in the chicken meat as the levels of dietary Khazar Kilka fish oil increased from 0 to 5%. The panellists identified the fishy smell of the meat when more than 20 g/kg fish oil was included in the diet.

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Determination of chemical composition and metabolizable energy of wastes of spaghetti, pasta, biscuit, crisp, chickpea pre-cleaning and chickpea screening plants

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Introduction Based on the official statistics of industries and mines organization about 1000 ton wastes of spaghetti, pasta, biscuit, crisp, and about 7500 ton waste of chick pea pre-cleaning plants are produced annually in East Azerbaijan province of Iran. In our country there is little researches on wastes. In one study, ileal amino- acid digestibility of wheat, autoclaved wheat and spaghetti by-products for broiler chicks was determined (Zaghari 2006). The aim of the present study was to determine metabolizable energy and chemical composition of wastes of spaghetti, pasta, biscuit, crisp, chickpea pre-cleaning and chickpea screening plants.

Materials and methods After classified random sampling from 10% of spaghetti, pasta, biscuit, crisp, chickpea pre-cleaning and chickpea screening plants, the samples were ground and mixed. 24 adult laying-type cockerels (Hy-Line W36, 35-week-old) with mean body weight of 2000 g ± 100 were randomly grouped into six groups of four replicates. All birds were kept in individual battery cages (25 cm × 35 cm × 50 cm in dimension) and fed commercial diets prior to the experiment. The wastes of spaghetti, soup pasta, rice pasta, crisp, biscuit, pre-cleaning chickpea, and chickpea screening were mixed in the ratio of 15% to basal diet. Then, in order to determine metabolizable energy (TME, TMEn) of waste, 30 grams of mixed feed were force-fed to 4 adult Leghorn-type roosters, according to the method described by Sibbald (1986). Excreta voided from each bird following the feeding procedure was collected quantitatively, for 48 h. Birds had free access to water, and when not on experiment, to a commercial diet. All the birds remained healthy and survived the experimental procedure. The chemical analysis of wastes and collected samples of excreta was carried out according to the standard methods of analysis (AOAC,1990). Gross energy of wastes and individual samples of excreta was measured by a bomb calorimetry.

Results Summarized in table1, are data showing chemical composition of studied wastes. Maximal crude protein, ash and crude fiber value was obtained with chickpea pre-cleaning.

Table 1 Chemical composition (g/kg fresh weight) of wastes

wastes	Dry Matter	Ash	Crude Protein	Crude Fat	Crude Fiber	NFE	NDF	ADF	Gross Energy (MJ/kg)
spaghetti	915	12	127	24	2	805	17	1	18.79
pasta	909	7	141	65	2	755	14	3	18.74
biscuit	948	20	90	170	51	649	111	17	20.37
crisp	935	15	92	42	5	826	131	3	18.23
chickpea pre-cleaning	923	73	302	87	178	320	323	224	19.38
chickpea screening	919	60	300	78	78	450	351	96	19.79

Table 2 Metabolizable energy of experimental wastes (MJ/kg)

wastes	AME	AMEn	TME	TMEn
spaghetti	14.41 ^d ±0.17	14.8 ^c ±0.18	15.77 ^d ±0.17	15.23 ^c ±0.18
pasta	15.69 ^c ±0.19	16.24 ^b ±0.25	17.06 ^c ±0.19	16.67 ^b ±0.25
biscuit	16.26 ^a ±0.09	16.78 ^a ±0.26	17.26 ^a ±0.09	17.22 ^a ±0.22
crisp	15.98 ^b ±0.13	16.25 ^b ±0.20	17.34 ^b ±0.13	16.69 ^b ±0.20
chickpea pre-cleaning	10.89 ^e ±0.13	10.34 ^f ±0.16	10.96 ^f ±0.22	11.64 ^d ±0.17
chickpea screening	9.6 ^f ±0.22	10.34 ^f ±0.16	10.96 ^f ±0.22	10.77 ^f ±0.16
SEM	0.088	0.098	0.088	0.098

Among the wastes, biscuit had the highest ME, presumably related to its highest fat content. Statistical analysis showed that there were significant differences ($p < 0.05$) between ME types of wastes.

Conclusions These results show that experimental wastes especially chickpea, biscuit and spaghetti are rich sources of protein and energy with some values even exceeding those in corn and wheat. Further work is required to test their suitability in diets for poultry.

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The effect of different lightning programs on reproductive performance of native turkeys

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Introduction Lighting is very important for turkey production, because their life period is longer than poultry (Nixey 1994). One of the important effects of lighting is to change the time of sexual maturity in pullets. Classen *et al.* (1994) concluded that constant light causes an increase incidence of leg problem and metabolic disorders. With lighting programs and lighting intensity, time of sexual maturation can be modified. Because there is little information about lighting programs in native turkey production in Iran, the aim of this research is determining the best lighting program for native turkey production.

Materials and methods The effect of 2 lighting program in growth period and 7 lighting program in production period on native turkey's reproductive performance was surveyed. Turkeys received (7L:17D) and (14L:10D) lighting programs by 36 weeks respectively. 400 female and 80 male turkeys were used on completely randomized design with four repeat, each contain 50 female and 10 male turkeys. The survey of lighting program continued in production period (37- 66 weeks). The groups receiving 7h light during the growth period received 4 different lighting program (12L:12D), (14L:10D), (16L:8D) and intermittent (1L: 3d) for 66 weeks. The group receiving 14h light during the growth period received 3 different lighting programs (14L:10D), (16L:8D) and (1L:3D). This period of experiment was carried on completely randomized design with four repeat, each contain 10 female and 2 male turkeys. Sexual maturity age, egg production, fertility, and hatchability percent and feed intake for every 1day's chick production were recorded.

Results The largest hatchability was in 7L: 17D in growing period and then 14L:10D in production period group, that was significant differences with 14L:10D in growing period and (1L:3D) in production period group ($p<0.05$). The least feed intake for every 1 day's chick production was in 14L:10D in growing and production period group, but there wasn't significant differences between treatments ($p<0.05$). The largest produced chick was in 7 and 14 lights respectively for growth and production period group, but there wasn't significant differences between treatments ($p<0.05$).

Table 1 Number of chicks produced from 100 female turkey, amount of feed intake(kg) for every 1day old chick production and mean of maturity age

Lighting program	Number chicks produced from 100 female turkey in every day	Fertility	Hatchability	Feed intake(kg) for chicken production	FI(kg) every chick production	for 1d maturity age(day)
Growth period	Production period					
7L:17D	1L:3D ¹	7.05	64 ^c ±20	77 ^{ab} ±18	54.93	7.79
7L:17D	12L:12D	9.30	72 ^b ±19	80 ^{ab} ±14	30.29	3.27
7L:17D	14L:10D	11.94	84 ^a ±12	85 ^a ±21	28.32	2.37
7L:17D	16L:8D	10.55	77 ^{ab} ±12	79 ^{ab} ±19	42.52	4.03
14L:10D	1L:3D	8.19	72 ^{bc} ±26	71 ^b ±15	22.45	2.74
14L:10D	14L:10D	10.18	62 ^{bc} ±19	85 ^a ±15	22.48	2.21
14L:10D	16L:8D	10.80	78 ^{ab} ±17	81 ^{ab} ±17	28.94	2.68

1-1L:3D=Intermittent program (1h light:3h dark). 2-7L:17D=continuous program(7h lightness:17h dark)

Conclusion The best FCR was for the groups received 14h L in growth period and 14h D in production period and also in the total production period, the highest egg production percent was for the groups received 7h L in growth period and 14h L in production period

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Effects of butyric acid, mannanoligosaccharide (MOS) and avilamycin on performance and small intestine morphology of broiler chickens

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Introduction The subtherapeutic use of antibiotics in animals has been under scientific and public scrutiny as antibiotic growth promoters (AGP) have been linked to the development of antibiotic resistance in bacteria, which poses a threat to human health (Smith *et al.*, 2003). Short chain fatty acids such as butyrate are considered as potential alternative to AGP, in addition to its bactericidal activity; butyrate appears to play a role in development of the intestinal epithelium (Leeson 2005). Prebiotics (e.g. mannanoligosaccharides, MOS) are nondigestible feed ingredients that can selectively stimulate growth or metabolic activity of a limited number of intestinal microorganisms (Gibson and Roberfroid, 1995). This study was, therefore, conducted to investigate effectiveness of mannanoligosaccharides and butyric acid as potential alternatives to AGP in broilers.

Materials and methods Two hundred and eighty eight one-day-old male broiler chicks (Arbor Acres Plus) were raised over a 42 d experimental period. Dietary treatments were: 1) Negative control, antibiotic free diet; 2) Positive control diet (containing 15mg/kg of avilamycin); 3) Negative control with MOS (Bio-Mos, Alltech Inc., Nicholasville, KY, USA, 2g/kg of the diet) and 4) Negative control with butyric acid (BabyC4, SILO, Industria Zootecnica, Florence, Italy, 3 g/kg of the diet to 21 days). Chicks were randomly assigned to 4 replicates per treatment and 18 birds per replicate. Dietary treatments were fed during starter (0-14 d), grower (14-28 d) and finisher (29- 42 d) periods. All diets within each period were prepared with the same batch of ingredients and had the same nutrient composition. For intestinal morphometric examination, two birds per replicate (8 birds per treatment) were euthanized at day 28 and 2-cm segments of the midpoint of the jejunum were removed and fixed in 10% buffered formalin. Serial sections were cut at 5 µm and placed on glass slides. Sections were deparaffinized in xylene, rehydrated in a graded alcohol series, stained with hematoxylin and eosin, and examined by light microscopy. Data were analyzed using the GLM procedure of SAS (2001). Differences among treatments were compared using a Duncan's multiple range tests.

Results The effects of addition of avilamycin, butyric acid and MOS on broiler performance, feed intake, body weight, villi height and crypt depth in the jejunum are shown in table 1. Birds fed MOS and butyric acid were significantly heavier than negative controls ($P < 0.05$). There were no significant differences among dietary additive groups with respect to body weight gain. Dietary supplementation with avilamycin, butyric acid and MOS significantly improved FCR ($P < 0.01$) although this improvement was greater in butyric acid and MOS compared to avilamycin groups. Carcass yield in butyric acid groups was greater than negative controls ($P = 0.08$). Birds fed avilamycin had lowest small intestine weight ($P < 0.01$). There were no significant effects of feed additives on villi height and crypt depth.

Table 1 Effects of avilamycin, butyric acid and MOS on growth performance, villi height, crypt depth, small intestine weight and carcass yield of broiler chickens

Treatment	BW gain 1-42days (g)	FCR 1-42days	Villi Height (µm)	Crypt depth (µm)	Small Intestine (% of BW)	Carcass yield (% of BW)	Breast weight (% of BW)
Negative control	2212 ^b	1.99 ^a	862	201	4.56 ^a	71.9	27.1 ^b
Avilamycin	2344 ^{ab}	1.83 ^b	871	189	3.45 ^c	74.5	29.8 ^a
Butyric acid	2370 ^a	1.72 ^c	925	194	3.85 ^{cb}	75.3	30.3 ^a
MOS	2376 ^a	1.75 ^c	869	209	4.14 ^{ab}	72.8	29.0 ^a
SEM	48.8	0.03	46.3	9.6	0.152	0.95	0.51
Significance	*	**	NS	NS	**	NS	**

^{a-c} Means in a column without a common superscript are significantly different. ^{NS} non-significant * $P < 0.05$ ** $P < 0.01$

Conclusions It was concluded that addition of butyric acid and MOS can improve broiler performance, without significant effects on intestinal morphology, including villus height and crypt depth. No difference in performance was observed between the two additives. The inclusion of avilamycin as a growth promoter was also associated with positive effects on performance of broilers. The results of this study indicated that addition of butyric acid and MOS to the diet could be an alternative to the use of antibiotics as growth promoters in broiler production.

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Growth curve models for commercial pullets under severe heat stress condition

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Introduction The objectives of our study were to propose a growth curve and to develop a mathematical model to describe the body weight of pullet experiencing severe heat stress (42°C). Poultry producers who raise their own replacement pullets can control their pullet's growth, condition and development. Many of the problems which occur during the early part of lay can be traced back to insufficient or improper type of body weight attained during the various stages of the growing period. In order to avoid these problems, the body weight of pullets must be controlled. Rearing conditions for pullets vary depending on environmental pressures and can affect growth rate. Without the basic knowledge of the flock grow-out, it is virtually impossible to understand and possibly solve problems which may later occur during the laying period. It must be kept in mind that once egg production begins, it is too late to solve problems resulting from growing period. The two most important criteria of pullet quality are uniformity within the flock and proper body weight at a specific age. Almost anything that adversely affects a pullet will usually be reflected in lower body weights and poorer flock uniformity. High ambient temperatures can be devastating to commercial pullet growth rate; coupled with high humidity they can have an even more harmful effect on proper and recommended body weight. Heat stress interferes with the poultry comfort and suppresses performance efficiency. In order to verify the effect of heat stress on pullet growth rate, many curve modelled, fitted and verified to proposed best one.

Materials and methods 2250 leghorns (HyLine-W36) pullets were used for the study which was conducted during the hot months of year in Mollasani educational and research centre (Ramin agricultural university, north west of Iran). All the bird fed isoenergetic and isonitrogenous diets. Body weight, age, mortality and DMI were recorded every week. Temperature and humidity were recorded and the Thermal-Humidity Index (THI) was calculated every day. The collected data were analysed by the Statistical Analysis Systems (SAS) NLIN procedure (SAS, 2005), and CurveExpert 1.3 software to fit and compare the Gompertz, Richards, Weibull, MMF, Logistic, Von bertalanffy and some other curves.

Results The most fitted one was Gompertz model with $r=0.998$ and $MSE= 24.11$ and with parameters $a= 1441.431$; $b=1.205$; and $c=0.023$. Consequently, under heat stress condition the growth model proposed by company is changed, and this must be considered by producers to achieve the best profit in production period.

Table 1 comparing some models fitted for pullet reared in hot environment conditions.

Model Name	Coefficient of determination	Standard Error	Parameter of model			
			A	b	c	d
Gompertz	0.998423	24.107	1441.4318	1.2053797	0.022682751	
4th Degree Polynomial	0.998499	25.041	50.305531	1.9294874	0.23985574	-0.002306
Richards	0.998352	25.397	1405.1059	-0.7664503	0.024911281	0.11889103
Weibull	0.998344	25.462	1451.4692	1400.4615	0.000732897	1.6011592
MMF	0.998323	25.620	58.263176	3332.0639	1869.7764	1.7846243
Logistic	0.996937	33.583	1265.264	13.149497	0.041046892	

The form of equations mentioned above was as $y=a*\exp(-\exp(b-cx))$ for Gompertz model, $y=a+bx+cx^2+dx^3...$ for 4th Degree Polynomial model, $y=a/(1+\exp(b-cx)^{(1/d)})$ for Richards model, $y=a-b*\exp(-c*x^d)$ for Weibull model, $y=(a*b+c*x^d)/(b+x^d)$ for MMF model and $y=a/(1+b*\exp(-cx))$ for Logistic model. The Coefficient of determination or R^2 was high for all models but the lowest standard error was seen in Gompertz model. So the Gompertz model proposed for its lower standard error and for using 3 parameters.

Conclusion Under heat stress condition the growth model proposed by the company is changed, and this must be considered by producers to achieve the best profit in production period.

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The effect of increasing levels of fish oil on immune responses of broiler chickens

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Introduction There has been interest in the enrichment of poultry meat with long chain n-3 polyunsaturated fatty acids (PUFA) as a means of increasing the low consumption of these acids by humans consuming western diets. There is some concern, however, that at high levels of consumption, n-3 PUFA may have detrimental effects on immune function. However, research to date shows that strong controversy surrounds this immunomodulation. The aim of this experiment was to determine the effects of dietary long chain PUFA on aspects of immune function in broiler chickens.

Materials and methods A total of 18 one-day-old male Ross 308 broiler chicks were reared as a single group for 21 d, and fed a common starter diet. At 21 d, birds were randomly allocated to one of six pens (106x106x108cm), three chicks per pen. Water and feed were provided ad libitum. The broilers were fed for 33 d one of three wheat-soyabean meal based diets. All diets contained 60g/kg added oil, which was either 0,60; 30,30; or 60,0 g fish oil (FO) and soya oil respectively. Chickens were sacrificed between 54 and 60 d of age and samples of blood, bursa of Fabricus, spleen and thymus were collected. The bursa of Fabricus, spleen and thymus were weighed. Heparinised blood was layered on equal volumes of histopaque for preparation of peripheral blood leukocytes. Lymphocyte subsets from the freshly harvested spleen and thymus were prepared and stained with four monoclonal antibodies (mAb): anti-CD3, anti-CD4, anti-CD8 and BU-1A (B cell marker). Immune cells were then enumerated using the FACSCalibur™ flow cytometer. Quantitative analysis of the phagocytic activity of peripheral mononuclear phagocytes in whole blood was performed using phagotest commercial kits, (ORPEGEN-Pharma). Results were expressed as percentage of fluorescent cells and data were analysed by CellQuest™ software. The overall differences of the effects between the three dietary treatments were analysed using one-way analysis of variance (ANOVA) and the general linear model procedure of Minitab was applied in all the tests. Differences between the treatment groups were considered statistically different at $P \leq 0.05$.

Results Increasing levels of fish oil did not affect the weights of the spleen. However, chickens fed diet with 60 g/kg FO had significantly lower bursa weights ($p < 0.01$) than those fed diets with 0 g/kg and 30 g/kg FO (Table 1). Chickens fed diets containing 30 g/kg FO had significantly higher thymus weights compared to chickens fed 0 and 60 g/kg ($p < 0.05$). There was no significant effect of increasing fish oil level on the percentage positive and mean fluorescence intensity (MFI) of the leukocyte subsets in peripheral blood, spleen and thymus. However, the proportion of B-cells in peripheral blood and the MFI of CD8 subsets in the spleen approached significance, $P = 0.058$ and 0.054 , respectively (data not shown). Results of phagocytic activity show that the different levels of FO in the experimental groups neither affected the positive percentage of cells nor MFI. However, there was a trend towards a lower proportion of monocytes being engaged in phagocytosis when broilers were fed diets containing 60 g/kg FO ($p = 0.055$) (Table 2).

Table 1 % Body weight of different tissues

Diet (g/kg FO)	Tissue ¹		
	Spleen	Thymus	Bursa
0	0.12	0.15 ^a	0.30 ^a
30	0.15	0.17 ^b	0.28 ^a
60	0.12	0.12 ^a	0.14 ^b
SE Mean	0.01	0.01	0.02
<i>P</i> value	0.164	0.022	0.001

Table 2 Effect of diet on % of phagocytic leukocytes and their MFI

Diet (g/kg FO)	Monocytes		Granulocytes	
	% of +ve cells	MFI	% of +ve cells	MFI
0	46.78	2148	69.77	1737
30	48.76	2306	82.34	3448
60	35.16	2044	72.84	2204
SE Mean	3.788	630	3.76	992
<i>P</i> value	0.055	0.962	0.102	0.267

¹For each tissue weight %, values with different superscripts are significantly different

Conclusions These results show no evidence of detrimental effect of enrichment with fish oil under the circumstances of this study. However, further studies should be conducted on different immune functions under different situations (e.g. challenge with pathogen), especially as there is an evidence of a potential threshold effect in some cases. Such studies would help the poultry industry to improve or maintain the health status of poultry at an optimum level in circumstances when poultry meat is being enriched with long chain n-3 polyunsaturated fatty acids.

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Effect of dietary Phytase and NSP-degrading enzymes in diets containing rape seed meal on broiler performance

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Introduction In comparison to 44% crude protein of soybean meal (SBM), the protein content of rapeseed meal (RSM) is about 35- 40% and has a physiologically suitable amino acid combination in animal nutrition, but RSM contains nutritionally unfavourable substances such as glucosinolates, sinapin, tannin, phytate and non starch polysaccharides (NSP) (Kocher *et al.*, 2000). Enzymes have the potential to be used in diets contain antinutritional factors that hinder nutrient availability. NSPs include cellulose, B-glucans, arabinoxylans, and pectins that may increase viscosity of digesta and cause a decrease in nutrient digestibility and performance of broiler chickens. Phytase activity from digestive secretions, some feed ingredients, resident bacteria, exogenous microorganisms, or both resident bacteria and exogenous microorganisms is present in the digestive tract of broiler chickens (Kornegay, 2001), but its efficiency at a practical level is very low. It is accepted that broilers lack sufficient levels of phytase activity to effectively hydrolyse the phytate molecule. Phytate-bound P is not well digested, so inorganic P is added to broiler diets that increased feed costs (Lescoat *et al.*, 2005). The purpose of this study was to investigate the replacement value of SBM with locally grown RSM and two types of enzymes (NSP-degrading and phytase) on performance of broiler chickens.

Materials and methods Three levels of (0.0, 25.0 and 50%) SBM protein was replaced with RSM protein and two levels of Phytase enzyme (0 and 500 FTU Phyzyme/kg, Phyzyme XP is a bacterial phytase from *Schizosacchomyces*), two levels of a dietary NSP-degrading enzyme (0, 0.17%, Grindazyme, minimum activity of 36000 U/g xylanase and 15000 U/g Beta-glucanase) were added to the diets during starter (7-21 days of age) and grower (21-42 days of age) periods. All diets had 0.5% available P and 0.95% Ca, and were isocaloric and isonitrogenous (2969 kcal/kg ME and 21.70% CP in starter; 3118.43 kcal/kg ME and 19.65% CP in grower period). 360 Ross strain chickens were used in a 2×2×3 factorial arrangement in a completely randomized design with twelve treatments, three replicates and ten birds per replicate. Feed consumption and body weight gain of chicks were recorded 4 h after the removal of feed and feed conversion ratio (FCR) calculated at end of every week. All Data were analysed by SAS (2000). Means compared by Duncan's test.

Results The results of this experiment indicated that FI, BWG and feed efficiency of broiler were significantly ($P<0.05$) decreased by increasing RSM in all period of experiment (table 1). These factors were highest in diet without RSM. The BWG and FI of broiler were significantly ($P<0.05$) increased by addition of Grindazyme, but were not affected by supplementation of Phytase in the diet. However Broiler fed diets contains phytase have numerically more FI and BWG than control diet.

Table 1 The main effects of diets contain Phytase and NSP-degrading enzyme and rapeseed meal on broiler performance

	Weigh gain (kg)			Feed intake (kg)			FCR		
	7-21	22-42	7-42	7-21	22-42	7-42	7-21	22-42	7-42
Phytase	0.50	1.31	1.81 ^a	0.79	2.60	3.39 ^a	1.56	1.99	1.87 ^a
No Phytase	0.52	1.33	1.84 ^a	0.78	2.63	3.41 ^a	1.51	1.99	1.85 ^a
SEM	0.01	0.01	0.02	0.007	0.02	0.01	0.03	0.01	0.01
Grindazyme	0.51	1.28 ^b	1.80 ^b	0.78	2.60 ^b	3.37 ^b	1.52	2.02 ^a	1.87 ^a
No Grindazyme	0.51	1.35 ^a	1.86 ^a	0.79	2.64 ^a	3.43 ^a	1.55	1.95 ^a	1.85 ^a
SEM	0.01	0.05	0.04	0.007	0.03	0.04	0.02	0.05	0.01
0% RSM	0.54 ^a	1.36 ^a	1.90 ^a	0.79 ^a	2.65 ^a	3.44 ^a	1.47 ^b	1.95 ^a	1.81 ^b
25% RSM	0.49 ^b	1.31 ^{ab}	1.80 ^b	0.78 ^b	2.62 ^{ab}	3.39 ^{ab}	1.58 ^a	2.00 ^a	1.89 ^a
50% RSM	0.51 ^b	1.28 ^b	1.78 ^b	0.79 ^{ab}	2.57 ^b	3.36 ^b	1.55 ^a	2.02 ^a	1.89 ^a
SEM	0.02	0.04	0.06	0.006	0.04	0.04	0.07	0.04	0.07

SEM: standard error of means; FCR: Feed conversion ratio; RSM: Rape seed meal

Conclusion Levels of RSM showed more adverse effects on performance. It seems addition of enzyme was an effective method to overcome the anti nutrition factor of RSM. Therefore, it may be concluded that NSP-degrading and Phytase enzymes incorporated in rapeseed meal based broiler diet could be beneficial.

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