

The effect of wheat straw substitution by different levels of date palm leaves on performance and health of Baluchi ewe lamb

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Introduction A major constraint of animal production in south of Iran is the lack of cheap source of roughages. Date palm leaves (DPL) is one of the most abundant agricultural by-products in south of Iran. Almost all pruned leaves are discarded in the fields, mainly for nutrients recycling and soil conservation (M. Wan Zahari, et al 1999). The yearly maintenance of date palm tree produces a (around 20 kg per each tree) considerable quantities of green leaves (Bahman et al (1997); Pascual et al (2000)). Ruminant can utilize crop residues, with poor nutritional value. These residues are traditionally fed to animal as the main part of diet in many developing countries. However; dry matter intake of these by-products are not adequate to fulfill the nutrient requirements of livestock even at maintenance level (Dixon and Egan, 2002). DPL has a great potential for use as a roughage or bulk source in total mixed ration (TMR) for ruminants in dry areas. Detailed studies on fermentation characteristics and palatability of DPL silage, as well as on animal performance, have been reported by many workers (e.g. Abu Hassan and Ishida, 1991; Ishida and Abu Hassan, 1997; Oshio et al., 1999). Some researchers such as El-din and Tag-El-Din, 1996; and Bahman et al., 1997 have reported that DPL cannot be fed to animals because of low crude protein (6-7%) and high level of fibrous cell wall content low palatability and digestibility. Therefore we design one experiment that investigates possibility of using DPL without any enrichment. The objective of this trial was to study the effect of replacement DPL with wheat straw and voluntary intake, average body gain and health of Baluchi ewe lambs.

Materials and Methods Twenty-four Iranian Baluchi female lambs with initial body weight (BW) of 20.48 ± 0.5 kg and age of 130 ± 10 days were assigned to 4 dietary treatments in a completely randomized design. Groups were balanced for weight and experimental trail lasted for 76 days. All lambs were given a TMR composed of 39% forage (alfalfa and wheat straw or DPL) and 61% concentrate. The concentrate portion (61%) was the same for all treatments, therefore the dietary treatments differed only in forage part of diet (39%) and they were 1) wheat straw (24%), 2) wheat straw (16%), DPL (8%), 3) wheat straw (8%), DPL (16%), 4) DPL (24%). The diets were fed in form of ad libitum and total mixed ration (TMR). DPL were collected in fall season (time of pruning), dried under the sun light and stored in a dry clean shed up to starting the feeding trial. DPL were chopped in particle size of 3-5cm before mixing. Approximately all diets were isocaloric and isonitrogenous. Diets were offered to the lambs twice daily in almost equal meals at 8 am and 4 pm to meet their feed requirement and fresh water was also available for sheep at all times during the trail.

Results and Discussion Feeding of DPL to Baluchi sheep did not affect their health. Such results have been reported elsewhere by other workers (Osman Mahgoub, et al., 2005). Lambs fed by diet 4 had higher fibrinogen content in their blood samples than other animals in other treatments. The highest feed intake (1033 g/day) was observed in diet 4 containing 24% of DPL. In contrast the animal fed by diet 1 (24% wheat straw) had the lowest feed intake (856 g/day) among all treatments. Average body weight gain of lambs fed by diet 1 was significantly ($P < 0.05$) lower than other groups. The best feed conversion ratio was recorded for the lambs on diet containing 24% DPL (6.38 kg feed intake for each kg of weight gain). Lower BW gain of animals fed by non-conventional feed is well documented mainly because of higher lignifications. Kafilzadeh et al (2009) reported that low digestibility of DPL than wheat stubble could be due to lesser accessibility of ruminal microbes to the fiber cell wall of DPL.

Conclusion This study indicates that Baluchi lambs can be fed with a diet based on date palm leaves and improve performance in tropical areas where lack of feedstuff source is the first limitation for livestock industry. Feeding of DPL requires further investigation and research about anti nutritional factor but in this study observed that feeding of DPL to lambs at 24% is possible without any negative effect and it is a good result for this agricultural by-product.

Key word: Baluchi lambs, Date palm leaves, Growth performance, Wheat Straw.

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Effects of Bacterial Inoculants and Absorbents on Fermentation Properties and Chemical Composition of Fresh Sugar Beet Pulp Silage Using Laboratory silos

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Introduction Ensiling is one of the common preserving methods for forage or other organic materials. In this method, organic matters were preserved by proving an aerobic condition and then by reducing the pH with increasing acids production (mainly lactic acid). Some circumstances like enough soluble carbohydrates, low buffering capacity and appropriate dry matter concentration are needed in ensilages for an ideal silage production. Seepage production during ensiling is one of the most problems especially when high moisture materials (like fresh beet sugar pulp) are ensiled. Silage seepage can pollute the environment and make loses in nutrients like soluble carbohydrates, protein, organic acids and etc. Moreover, lactic acid bacteria inoculants (Mainly consist of *Lactobacillus plantarum*) have been widely used for improving fermentation pattern in ensilages. These external provided bacteria usually enhance lactic acid production in silage and then accelerate the falling of pH values in silages. Rapid decrease in pH can inhibit non-beneficial bacteria from activity which finally preserves nutrients from un-necessary fermentation or oxidation. The aim of this study was to investigate the interactive effects of lactic acid bacteria inoculants and some absorbents (straw and pith) on chemical properties and fermentation profile of wet sugar beet pulp silage.

Materials and Methods In the first experiment, fresh wet sugar beet pulp was treated with 5% straw or 5% pith in order to investigate the effects of these absorbents on chemical composition, fermentation characteristics and effluent production during ensiling period. In the second experiment, fresh wet sugar beet pulp was treated with a commercial lactic acid bacteria inoculants (Ecosyle) and/or 5% pith in order to investing the main and interaction effects of the bacterial bacteria inoculants and the best absorbents from the experiment 1. In both experiments, triplicate samples were prepared for each treatment after mixing the fresh sugar beet pulp with absorbents or inoculants. Ensilages were filled in laboratory silos and packed and then were kept for 90 d in room temperature at dark. After opening the concentration of volatile and non-volatile fatty acids, crude protein, fibers, total and ammonia-N and the values of pH were measured in final produced silages.

Results and Discussion In the experiment 1, concentration of dry matter (DM), neutral detergent insoluble fibers (NDF) and acid detergent insoluble fibers were higher in absorbents treated silage ($P>0.01$) when compared with untreated one. Application of absorbents resulted in silages with lower seepages ($P>0.01$) production compared to the control. However, application of the absorbents to the beet pulp produced silages with lower in vitro DM digestibility ($P>0.05$). Straw treated silage had the highest NDF concentration and the lowest apparent and true in vitro DM digestibility. Application of absorbents produced silages with lower lactic acid ($P>0.01$) and higher pH ($P>0.05$) and ammonia-N ($P>0.01$) concentration. Adding straw to sugar beet pulp produced silages with higher acetate concentration, total volatile fatty acids concentrations (VFA) ($P>0.01$), the ratio of ammonia-N from total N ($P>0.01$) but lower ratios of lactate to acetate ($P>0.01$), lactate to acetate + propionate ($P>0.01$) and lactate to VFA ($P>0.05$) when compared with control group. In contrast, adding pith to sugar beet pulp produced silages with lower acetate concentration ($P>0.01$), propionate concentration ($P>0.01$), total VFA ($P>0.01$) but higher ratios of lactate to acetate ($P>0.01$), lactate to acetate + propionate ($P>0.01$) and lactate to VFA ($P>0.05$) when compared with control group. The Fleig point was not affected by the different treatments. Application of bacterial inoculant resulted in silages with higher DM concentration ($P>0.01$), water soluble carbohydrates concentration ($P>0.01$) and in vitro DM digestibility ($P>0.05$) but lower crude protein

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concentration ($P>0.05$). Moreover, Application of bacterial inoculant resulted in silages with higher butyric acid concentration ($P>0.01$), VFA concentration ($P>0.01$), ammonia-N concentration ($P>0.01$) and Fleig point ($P>0.01$) but lower pH values ($P>0.01$). Simultaneous application of bacterial inoculant and straw to sugar beet pulp silage resulted in silages with lower concentrations of lactic acid, acetic acid, propionic acid, total VFA but higher Fleig point.

Conclusion Finally, according to homolactic fermentation, lower ammonia-N and less negative effect on digestibility in pith in comparison with straw and bacterial inoculant, it is suggest that to use 5% pith for treating sugar beet pulp prior to ensiling.

Keywords: *Lactobacillus plantarum*, Pith, Silage additives, Straw, Water absorbents.



Survey the frequency and type of Fungal Contaminants in Animal Feed of Yazd Dairy Cattles

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Introduction About 500,000 species of fungi have been realized up to now. There are abundant fungi in air, soil and our environment. So the growth of them increases in the presence of air moisture and appropriate temperature. However saprophytic fungi have a wide distribution in nature, they are responsible for decomposition of organic materials and playing an important role in the biogeochemical cycles of major nutrients. Some saprophytes are toxic that contaminate human foods and animal feeds by production of mycotoxins. Aflatoxins are the most common and dangerous mycotoxins produced by few species of *Aspergillus* and *penicillium*. This group of mycotoxin has disorder and risks, including the induction of liver cancer. They are mutagenic and teratogenic. Aflatoxin B1, B2, G1 and G2, which are naturally produced by several toxic fungi, may contaminate a wide range of dairy animal feeds resulted severe economic loss of cattle meat. Since Aflatoxin B1 and B2 can be transmitted via mammalian's milk and cheese in form of synthetic Aflatoxin M1 and M2 to human consumers, cause significant health problems. Therefore contamination of animal feed with common toxic airborne saprophytic fungi is a major concern of health officials. Wheat, barley, corn, soybean and other animal feeds may be contaminated with toxic fungi during implantation, harvesting and storage. There are many dairy and livestock centers in Yazd that prepare milk and dairy products for Yazd and neighboring provinces. The aim of current study was to evaluate the amount and type of fungal contaminates of dairy feeds in Yazd dairies.

Materials and methods This cross-sectional descriptive study was conducted in the summer of 2012 on 23 dairies in Yazd. Samples of different animal feeds including concentrates, wheat straw, hay, corn, silage corn, soybean and canola as well as waste of bread, were randomly selected from their bulks. The temperature and humidity of feed storage were recorded by using thermometer and portable hygrometer at the time of sampling. Samples were transferred to medical mycology laboratory in paramedical school by sterile containers. Samples were cultured on Sabouraud dextrose agar plates based on standard method. Isolated fungal colonies were firstly enumerated and identified using macroscopic and microscopic characteristics for determination of their genus and species of saprophytic and toxic fungi. The suspected fungi with definitive diagnosis by the use of the mentioned methods were then identified by performing slide culture by Riddle method. For the detection of aflatoxin producer species UV radiation was used. Results were analyzed statistically by Chi-square and Mann-Whitney test using SPSS 16 software.

Results and Discussion Saprophytic hyphomycets including *Cladosporium*, *Alternation* *Penicillium*,

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Verticillium, Aspergillus, Penicillium species and yeast were the most prevalent isolated fungi from cattle feeds in current study. Bread waste showed maximum contamination with opportunistic fungi such as Mucor and Rhizopus species, other saprophyte moulds and yeasts. Wheat straw had the highest contamination of aflatoxin-producing toxic fungi particularly Aspergillus flavus. Silage and concentrate were ranked as highest average contamination with 42600 and 40600 CFU/g, respectively in present study. There was seen a significant relationship between the average humidity of the environment of open and covered storage of feed with frequency of isolated fungal species ($P < 0.001$). In indoor environments, the humidity was higher than outdoors, and this is one of the factors effecting the increase aggregation in animal feeds stored in indoor storages. Results of present study were supported by similar previous studies that can be beneficial for the management of cattle feeds and public health surveillance particularly in disorders of aflatoxin.

Conclusion According to results of present study, there are a high fungal contamination in cattle food with saprophytic and toxigenic fungal spores. Monitoring of animal feed and control of humidity can control the proliferation of micro-organisms in food, eliminate contamination of microorganisms and prevent cross-contaminations. High levels of fungal contamination in intake dairy feed shows that the major source of contamination seems to be in raw materials used for animal feed. With respect to the effect of cattle food contamination especially contamination with Aspergillus flavus on health of cattle and dairy products and the secondary effects on human health, Control of fungal contamination in livestock foods is the best way to prevent aflatoxin contamination in milk and other dairy products, which help to improve the public health.

Key words: Aflatoxin, Animal feed, Dairy cattle, Fungal contamination, Yazd.



Digestibility, chemical compound and protein quality of *amaranthus* forage at two harvested cut

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Introduction Amaranth, genus *Amaranthus*, belongs to the *Amaranthaceae* family and includes more than 60 species (28). Amaranth forage, is distinguished by high yield performance of up to 70 t/ha (30). Plant maturity was found to affect neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, CP and nitrate content of amaranth forage (27). Several studies (21, 27, 29) have shown that the nutritive value of amaranth as a ruminant feed is equal to, or better than, commonly used forages such as alfalfa. Its favorable chemical analysis (e.g., high crude protein (CP) and low lignin ranges from 80 to 285 and from 17 to 73 g/kg DM, respectively) and its high dry matter (DM) digestibility (590–790 g/kg DM) low nitrate and oxalic acid concentrations (below toxic levels) suggest that it has potential value as a ruminant feedstuff (21, 23, 27).

Materials and Methods The forage was planted in spring at one field (10 ha) near Karaj city (Iran). The area is at an altitude of 1215 m above sea level, with a mean annual rainfall and temperature is 305.8 mm and 15°C, respectively. In the early autumn, samples were harvested by hand from at least 10 locations within the field and pooled to five samples. The dry samples were analyzed for DM (method 930.15), ash (method 924.05) and CP (method 9 84.13) of AOAC (6). Neutral detergent fibre (NDF) and acid detergent fibre (ADF) according to the method of Robertson and Van Soest (35) with NDF and ADF assayed sequentially without a heat stable amylase, and expressed inclusive of residual ash (25). Insoluble CP, soluble CP (SP), true protein (TP), AD insoluble CP (ADICP) and ND insoluble CP (NDICP) were determined according to Licitra method (15). Nitrate was determined by a colorimetric method (8), and oxalic acid was determined according to Abaza method (2). Ca and Mg were determined by atomic absorption, P was analyzed by spectrophotometer and K by flame emission spectrometer (6). DMD, OMD and DOMD were determined by a two stage technique for the *in vitro* digestion of forage crops (32). In a completely randomized experiment with factorial arrangement (2×2) the digestibility, chemical compound and protein quality of two varieties of amaranthus including: Kharkof (K), A. Spp.(S). which were planted as forage crops and harvested at two cuts, were studied. Data were subjected to analysis using the GLM procedure of SAS (26), using the statistical model:

$$Y_{ijk} = \mu + V_i + D_j + V_i D_j + e_{ijk}$$

Results and Discussion Results of *in vitro* two stage digestibility (IVD) for DM, OM and DOMD were 60.19, 58.16 and 49.09, respectively which were significantly ($p > 0.05$) affected by cultivars and interaction between cultivars and cuts. Results indicated that the means of CP, ASH, NDF, ADF, ADL, NDIN, ADIN, Ca, P, Mg, and K were 14.72, 15.44, 40.85, 27.90, 5.07, 21.76, 6.60, 1.55, 0.25, 0.28, 1.48, percent; respectively. Plant maturity was found to affect neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, CP and nitrate content of amaranth forage. Amaranth contains a high ash content due to its C4 metabolism and a very high carbon uptake per unit area (29). This high ash is in agreement with other researchers (1, 21). Our amaranth accession had higher NDF (27) and lower ADF (22, 23) in comparison to *A. Hypochondriacus* reported by other researchers. Lignin content in amaranth is less than that reported by other researchers (27).

With increased plant maturity, fraction B1 decreased, probably due to an increase in cell wall bound N (16). Researchers observed that Similar results in CP fraction of amaranthus hypochonderyacus (21). We observed that a negative relationship between NDF and fraction B1. There was a tendency for fraction B2 to increase with advancing maturity, similar to other researchers (21). At the second harvest, fraction B3 was higher than that at first harvest, similar to reserchers with amaranthus hypochondriacus (1). Different changes in NDF concentration of plant parts with maturity may explain the differences in proportions of fraction B3 (1). At the

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second harvest, fraction C increased due to increasing lignification (1). However, the content of fraction C was lower than that reported by abbasi et al. (1) and this fraction is unavailable to the animal (1).

Similar to our results, Sleugh et al. (27). Observed that as forages advance in maturity the CP content decreases (27). The total means of metabolizable energy obtained from IVD method was 2.56Mcal/kg DM. Our amaranth accession had lower metabolizable energy comparison to A. Hypochondriacus accession reported by other researchers (21). The different parts of protein including A, B1, B2, B3 and C, according to CNCPS system orderly were: 40.48, 2.24, 34.19, 16.09 and 7.01 percent of CP and NPN were 94.55 percent of soluble protein, the quantities of nitrate and oxalate were 0.34 and 5.37 percent (DM basis) respectively. Nitrate levels in our amaranthus forage were lower than the toxic level (i.e., >10–30 g/kg DM) reported by some researchers (3, 35). Sleugh et al. (27) reported higher nitrate levels (i.e., 18g/kg DM) for A. Hypochondriacus (27). The oxalic acid content of our amaranth is lower than that observed by Several studies (i.e., 2–114 g/kg DM) (5, 11, 31). Several studies have shown that the amount of amaranthus CP is between 12 to 27 Percent in whole crop (17, 29) and the other researchers show that the amount of amaranthus cp significantly decries by plant maturity (29). Some researchers reported that average of calcium, phosphorus and magnesium in the amaranths forage, is: 1.9, 0.26, and 0.6 respectively (1, 24).

Conclusion In general, results indicated that the Two varieties of Amaranthus tested in this study, potentially have considerable nutritive value as forage crops, meanwhile some of them have higher nutritive value.

Key words: *Amaranthus* forage, CNCPS, digestibility, harvested cuts.



Effect of Digestible Protein and Sulfur Amino Acids in Starter Diet on Performance and Small Intestinal (Jejunum) Morphology of Broilers

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Introduction Protein is an essential constituent of all tissues of animal body and has major effect on growth performance of the bird. A better understanding of the nutritional requirements of amino acids allows a more precise nutrition, offering the possibility for the formulator to optimize the requirement of at least minimum levels of crude protein by essential amino acids requirements, generating better result and lower costs for the producer. Methionine + Cystine (total sulfur amino acid = TSSA) perform a number of functions in enzyme reactions and protein synthesis.

Methionine is an essential amino acid for poultry and has an important role as a precursor of Cystine. Methionine is usually the first limiting amino acid in most of the practical diets for broiler chicken. The efficiency of utilization of dietary nutrients partly depends on the development of the gastro intestinal tract.

Material and methods A 2×3 factorial arrangement in a CRD experiment was conducted to study the effect of digestible protein (DP) and sulfur amino acids (DSAA) during the starter period on performance and small intestinal (jejunum) villous morphology. A total number of 300 day-old Ross 308 male broiler chicks were randomly distributed to 30 groups with 10 chicks each. Treatments consisted of two dietary levels of DP (19.5 and 21.5%) and three dietary levels of DSAA (0.94, 1.02 and 1.1%) that were fed for 10 days. For Each group and treatment, Feed Intake (FI), Weight Gain (WG) and Feed Conversion Ratio (FCR) were calculated and all the data were statistically analyzed by the SAS software.

Results and Discussions The effects of different levels of protein and digestible sulfur amino acids on the mean feed intake, feed conversion ratio and daily weight gain are shown in the Table 3.

Increase in the percentage of digestible sulfur amino acids, increased the levels of feed intake and feed conversion ratio in the starter period but, had no effect on the WG. Adding the DSAA amount, higher than the recommended levels by Ross in the starter period, had no significant increase in the WG, FI and FCR. Increasing DSAA in the starter diet, although did not improve the FCR, but significantly increased daily weight gain and feed intake at the end of the period. With a decrease of 10 percent in digestible protein of the starter diet and its counter-action with digestible sulfur amino acids had no effect on WG, FI and FCR in any experimental periods. By taking the use of the diets of 20% Crude Protein (CP) and 0.641% and 0.926% of Total Sulfur Amino Acids (TSAA) or the 22% of CP and 0.705% and 0.926% of TSAA, no weight gain difference was observed from 1 to 7 days of age. Also, a 2% reduction of CP in diet of the starter period was not effective on the WG. The results of the present experiment showed that in the starter diets containing 3025 kCal/kg of metabolism energy, the levels of the digestible protein could be increased 20 gr per every kg of diet, without observing any negative effect on the performance of the broilers and in this situation extra amino acids are added to the diet.

Results of the effects of different levels of the digestible protein and digestible sulfur amino acids on the carcass parts and the gastro intestinal tract are presented in the table 4.

Increasing the DSAA, had no significant effect on carcass parts and relative weight of the gastro intestinal tract in the day 42, but the abdominal fat decreased ($P < 0.05$). Similar levels for the methionine are needed to improve the output of the diet and the breast yield among the range of 3 to 6 weeks age.

The morphology of the jejunum of the broilers of 10 days age, are shown in the table 5. With an increase of 8.5% in DSAA of the diet, length and width of the villous of the jejunum were increased in the day 10. By decreasing 10% of the digestible protein of the starter diet, muscle thickness of the jejunum was lessen in the age of 10 days. In the starter period, the levels of the digestible lysine had no effect on the length of the duodenum and jejunum villous. Relative lengths of the gastro intestinal tracts are accessible in table 6.

Conclusion Increasing the percentage of the digestible sulfur amino acids in the starter diet, improved the FI

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and FCR but had no effect on the WG. Increasing the DSAA in the starter period, daily weight gain and feed intake were significantly increased in the final total periods but not for the FCR. 10% decrease in the DP and its interaction with digestible sulfur amino acids of the starter diet, had no effect on any of the parameters in any periods. With an 8.5% increase in DSAA of the starter diet, the abdominal fat in the 42 days age was decreased. 10% decrease in digestible protein of the starter diet, decreased the muscle thickness in the age of 10 days.

Keyword: Broiler, Digestible protein, Digestible sulfur amino acids, Jejunum morphology.

Effect of different levels of marigold (*Calendula officinalis*) oil extract on performance, blood parameters and immune response of broiler chickens challenged with CCl₄

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Introduction Although use of antibiotic as growth promoter in poultry and animal nutrition have led to positive effects, researches indicated that antibiotic residues in animal and poultry products caused resistance of bacteria and fungi strains and a resistance to antibiotics as a treatment tool for human diseases. Herbal extracts, probiotics and enzymes are suggested as replacers for antibiotics in animal and poultry nutrition. Plants and their active substances with their variety of functions are used as medicinal plants for years to prevent and treat many diseases in human, animal and poultry. Oil extracts of marigold has many active substances such as saponins, flavonoids and antioxidants and serve as a strong antioxidant to control free radicals. Therefore, the extract of marigold was used to test its curing effects on challenged birds with tetra hydrochloride (CCl₄), an inducer for liver damage.

Material and methods This experiment was conducted to evaluate the effects of marigold oil extracts (MOE) on performance, blood parameters and immune response of broiler chickens in a 42-day period. A total of 200 Ross 308 male broiler chickens were allocated to five dietary treatments with four replicates of 10 birds each. Treatments consisted of 1) control (without marigold extract and CCl₄), 2) CCl₄, 3-5) 150, 300, and 450 mg/kg marigold oil extract as supplement + CCl₄ (1 mg/kg body weight). CCl₄ was injected intraperitoneally from 21 to 30 days of age in a 2- day intervals. During this period sodium chloride (0.9%) was added to control group. At day 33, one chick from each replicate of treatments was selected, and their blood and internal organs were used for different bio assays.

Results and Discussion No significant differences detected among treatments for performance. However, the highest and the lowest feed intake at starter and grower periods obtained from the treatments used MOE and control groups, respectively (table 2). The highest and the lowest weight gain were also observed for the birds received 300 mg /kg MOE + CCl₄ and CCl₄ groups, respectively. Many studies have revealed that phytobiotics had no effect on performance of the birds when marigold, cinnamon and Garlic powder were applied as supplements into the diets. The results are in agreement with those of this study. Birds fed different levels of MOE + CCl₄ had higher liver weights than those of birds received only CCl₄. However, this effect was not significant. In control group, liver weight was lower ($P > 0.05$) than that of the birds received CCl₄ (1.581 vs 1.451) and MOE alleviated the negative effect of CCl₄ on this trait (table 5). The relative weight of the thymus at 33 days of age (table 4) and the IgG level (table 3) at 28 and 35 days of age in the group received 300 mg/Kg of MOE + CCl₄ were significantly higher ($P > 0.05$). Levels of blood metabolites (SGOT and SGPT) were increased when CCl₄ was added to the diets (table 5) and MOE was able to reduce the negative effects of CCl₄. Levels of cholesterol and triglycerides were significantly decreased in the birds received MOE when compared with those of the birds in positive control group ($P < 0.05$). MOE again was able to decrease the negative effect of CCl₄ on blood metabolites of bilirubin, cholesterol, and triglyceride. MOE possess hepatoprotective property that might be attributed to its active substances (flavonoids) present in the flower of marigold. Plants and plant extracts having flavonoids and phenolic compounds are able to have the most antioxidant properties. The extracts of marigold contain abundant amounts of active biological metabolites of flavonoids and terpenoids and the alleviating effect of MOE on organ weights, blood metabolites and immunological indices might be due to these substances.

Conclusion Under the conditions of this study, it was concluded that marigold oil extract at moderate levels is able to decrease the negative effects of CCl₄ induced hepatic cell injuries and can be used as a suitable natural antioxidant in poultry nutrition.

Keywords: Broiler chickens, carbon tetrachloride, liver, marigold oil extract.

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Effect of Barley and Enzyme on Performance, Carcass, Enzyme Activity and Digestion Parameters of Broilers

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Introduction Corn has been recently used for producing ethanol fuel in the major corn-producing countries such as the US and Brazil. Recent diversion of corn for biofuel production along with the increased world's demand for this feedstuff has resulted in unprecedented rise in feed cost for poultry worldwide. Alternative grains such as wheat and barley can be successfully replaced for corn in poultry diets. These cereal grains can locally grow in many parts of the world as they have remarkably lower water requirement than corn. Wheat and barley are generally used as major sources of energy in poultry diets. Though the major components of these grains are starch and proteins, they have considerable content of non-starch polysaccharides (NSPs), derived from the cell walls (Olukosi et al. 2007; Mirzaie et al. 2012). NSPs are generally considered as anti-nutritional factors (Jamroz et al. 2002). The content and structure of NSP polymers vary between different grains, which consequently affect their nutritive value (Olukosi et al. 2007). Wheat and barley are generally used as major sources of energy in poultry diets. The major components of these grains are starch and proteins, they have considerable content of non-starch polysaccharides (NSPs), derived from the cell walls. NSPs are generally considered as anti-nutritional factors. The content and structure of NSP polymers vary between different grains, which consequently affect their nutritive value. The major part of NSPs in barley comprises polymers of (1→3) (1→4)-β- glucans which could impede growth factors and consequently carcass quality through lowering the rate and amount of available nutrients in the mucosal surface of the intestinal.

Materials and Methods This experiment was conducted to evaluate the effect of corn and barley based diets supplemented with multi-enzyme on growth, carcass, pancreas enzyme activity and physiological characteristics of broilers. A total number of 375 one day old Ross-308 broiler chickens were allocated randomly to 3 treatments with 5 replicates using a CRD statistical design. Treatments were included control, barley and barley+ enzyme. The experimental diets were formulated to have similar contents of crude protein, metabolizable energy, total non-starch polysaccharides (NSP).

Results and Discussion According to the results, effect of barley with or without enzyme on growth performance at starter, grower and the entire period and also on carcass characteristics, pancreas enzyme activity and measures of ileal acidity and viscosity at the age of 42 were significant ($P < 0.01$). Birds fed barley diets gained significantly lower weight and carcass quality than those fed corn or supplemented with enzyme. The best growth performance and carcass characteristics belonged to control and the worst belonged to barley treatment, but enzyme supplementation of barley resulted in significant improvement of the traits ($P < 0.01$). Results indicated that birds on barley diet supplemented with multi-glycanase consumed significantly ($P < 0.01$) higher feed than those fed corn. Birds fed barley diets gained significantly lower weight and higher feed conversion ratio (FCR). Also results indicated that birds on barley diet supplemented with multi-glycanase gained significant ($P < 0.01$) higher dressing percentage as well as higher breast and leg percentage than those fed on barley. Among all treatments the highest pancreas weight and the longest gut length belonged to barley treatment. The growth performance data are consistent with the viscosity of ileal digesta. The viscosity of intestinal digesta in chicks fed barley showed the highest value, which was significantly ($P < 0.01$) greater than the control group fed on corn. The highest acidity and lowest viscosity of ileal content belonged to control and barley+ enzyme respectively, but reversely lowest acidity and highest viscosity related to barley treatment ($P < 0.01$). In fact, NSP of barley increased digesta viscosity in the intestine. Increased viscosity per se creates ideal environment for maximal proliferation of anaerobic and Gram negative bacteria as resulted high nutrient consumption rate by the bacterial population and low absorption of digested nutrients in the gut by host. Supplementation of barley diet with multi-glycanase significantly reduced the negative effects of NSP on

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proliferation of desirable bacteria (especially on probiotic-type bacteria) in the intestine through breakdown of NSP polymers. Barley treatment had the highest activity of amylase and lipase in the pancreas, but enzyme supplementation significantly reduced the enzyme activity ($P < 0.01$). This finding reflects the fact that water soluble NSP of barley impede pancreatic α -amylase and lipase activities in the intestinal lumen, which necessitate greater secretion of these enzymes from pancreas. Research has shown that intestinal enzyme activity depends on dietary nutrient source and presence of anti-nutrients in the gut.

Conclusion In conclusion, NSP components of barley have adverse effects on digesta physicochemical properties of the gut and subsequent transmission of hydrolyzed products to the enterocyte cells and nutrient absorption. Besides, NSP polymers of barley decrease the growth rate and carcass traits but increase the pancreatic α -amylase and lipase activities. Such changes are remarkably restored by supplementing NSP-degrading enzymes to broiler diets.

Key words: Broiler, Barley, Enzyme Activity, Performance.

The effect of lemon grape and apple pulps on performance, carcass traits, digestive tract characteristic, intestinal morphology and immune status of broilers

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Introduction Iran has good condition in fruit production among all countries in the world. Lemon, grape and apple are fruits that are produced in large scale in our country. For economical use of these fruits, there are different factories which produce fruit juices. Pulpes are the main waste that remain after juicing fruits. Pulpes contain some nutrients such as vitamins and minerals and secondary substances, also in contrast to common diets ingredients, there are very cheap and using them in diets may reduce the production price. As pulpes contain high level of fiber, can be easily used without any adverse effects in ruminant, but in poultry, especially in broilers using higher level of pulpes may be not possible. However, using low level of pulpes do not have any adverse effects on performance and blood parameters and can improve the meat quality and blood biochemical and immune parameters. Moreover discharge of these wastes to environment can cause serious environmental problems. This experiment was conducted to evaluate the effects of lemon, grape and apple pulpes on performance, carcass traits, digestive tract characteristic, intestinal morphology and immune status of broilers.

Materials and Methods In this experiment 240 Ross-308 broilers were used in 5 treatments, 4 replicates and 12 chicks in each replicate in a completely randomized design. Experimental groups included: 1) control group (without using pulp), 2) group with 150 mg/kg vitamin E (as positive control group), 3) group with 3% lemon pulp, 4) group with 3% grape pulp, 5) group with 3% apple pulp. Grower diets from 11 to 24 days and finisher diets from 25 to 42 days were used by broilers. In the experimental periods all chickens in experimental groups had free access to feed and water. The lighting program included: 23 h light and 1 h darkness in all the experimental period. Feed intake and weight gain were measured at the end of growing and finishing and whole periods. Feed conversion ratio was measured by dividing the amount of feed intake to the amount of weight gain. Carcass quality traits, digestive tract characteristic, intestinal morphology and immune condition of broilers were measured at the end of experimental period.

Results and Discussion Grape and apple pulpes significantly improved the feed conversion ratio ($P < 0.05$), whereas lemon pulp and vitamin E in contrast to control group had no significant effects on the performance of broilers ($P > 0.05$). The main reasons for these results may be related to the amount of vitamin E in the diet, the mixing condition of feed, diet preserving time and thermal condition during preserving and using diet, and the amount of fat in diet. Using lemon, grape and apple pulpes in contrast to vitamin E and control groups, caused the percentages of carcass and breast meat increase and the percentages of gizzard and liver decrease ($P < 0.05$). Experimental diets had not effects on digestive tract characteristic, intestinal morphology and immune condition of broilers ($P < 0.05$). Pulpes contain some nutrients such as vitamins and minerals and secondary substances. Most of these compounds have beneficial effects on the intestinal microorganisms population, level of intestinal pH and finally body health. So, using these pulpes in diets caused broilers effective use of their essential contents and improve their body and health condition, so feed conversion ratio and carcass traits significantly improved in groups used pulpes in contrast to control and group contain vitamin E, but other measured parameters did not significantly change at the end of experiment period. It may be related to pulp levels, experimental period length, pulp composition, other diets ingredients and farm management.

Conclusion The overall results of current study showed that in broiler chickens, using 3% of lemon, grape and apple pulpes without having any effects on the digestive tract characteristic, intestinal morphology and immune condition can improve the performance and carcass traits. For more information, using other levels, other sources, upgrading of pulpes by using different chemical agents and physical methods and mixing different pulpes to each other with consideration of economically points is recommended. As laying hens have developed digestive tract in contrast to broilers, they can tolerate higher levels of different kinds of pulpes in their diets.

Keywords: Broilers, Digestive tracts, Intestinal morphology, Performance, Pulp.

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Factors affecting on longevity in Northern Khorasan Kordish sheep

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Introduction The longevity is normally defined as the length of its productive life in the flock, which is the amount of time an animal spends producing (1). Longevity reflects the ability of ewe to avoid being culled for low production, low fertility, illness and influences the number and cost of replacements required to maintain the flock size. The benefit of increasing longevity are increased average age of the flock, having more ewes available for sale at the end of their four parity, having more ewe lambs to sell, and higher productivity from a slightly older flock age profile (2).

Sheep population of Khorasan province (10). In sheep production, longevity has an important influence on the economic returns. Reliable estimates of non genetic effects of longevity are needed to aid establishing an efficient strategy for improving ewe productivity.

Thus, the objective of this study was the factors affecting on longevity in Northern Khorasan Kordish sheep. We want to determine effect of environmental and non-genetic factors on longevity.

Material and Methods In order to investigate on factors affecting longevity trait in Northern Khorasan Kordish sheep, records of 7469 sheep (187 sire and 2258 dam) between 1990 to 2012 that were collected by breeding station of Hossein Abad in Shirvan (This city is located in the north of Mashhad) were used. Flock has been kept under village system. Breeding ewes were identified in the data set as those that lambed at 2 year of age and culled at 6 to 7 year of age duo to oldness. Ram kept until a male offspring was available for replacement. In this study longevity was defined as the age of a ewe (in day) when it leaves the flock. Ewes were generally removed from the flock due to poor production, low fertility, death and illness. Ewes were identified as being removed from the flock if a lambing record was present at n year of age but not at n+1 years of age. All females were culled before reaching 7 years of age. Conditional stayability measures were recorded as binary indicators of a lambing record at a given age (failure=1, success=0). Overall stayability traits (STAY n/2) were defined as the probability that a ewe that lambed at 2 year of age also lambed at n year of age for n equal to 3, 4, 5, 6, and 7 year of age. Data for overall stayability only included individuals that had opportunity to have a lambing record at n year of age. Marginal stayability traits (STAY n/n-1) were defined as the probability that a ewe that lambed at n-1 year of age also lambed at n year of age for n equal to 3, 4, 5, 6, and 7 years of age. Marginal stayability was missing for ewes without lambing records at n-1 years of age. Also, longevity of ewes was calculated according to lambing date and culling date in flock. So, culling reasons are divided in to two groups: Involuntary and voluntary culling. Involuntary culling includes death, barren ewes and deficiency, and voluntary culling includes extension sale, excess ewes and ewe with inappropriate phenotype.

The Fixed effect included year of production, sex and birth type. The Fixed effects to be considered was made after were when significant. Tables of productive life were also determined table procedure of SAS 9.1.

Result and Discussion According to the results, 99% of productive ewes were culled due to the culling reasons but not due to their ages. The highest culling rate in ewes was due to disease (33.1%) and the lowest culling rate in rams was Poisoning (0.11%). The disease had the highest culling rate in twins (33.5%). The highest percentage of involuntary culling was observed in barren ewes (1.65%). In voluntary culling, the highest percentage of culling was due to sales (55.75%). The effect of year and season of birth was significant on productive life. Mean comparison test showed that ewes lambing in winter and spring had the highest and lowest of productive life, respectively ($p < 0.05$).

Also the number of removed ewes from the flock decreased when increased age of ewes, but culling rate in ewes that have more than four ages. Survival of ewes with having single type were more than ewes with twin type. Ewes with one lamb had more survival) 1%) compared to ewes with 2 and 3 lambs (0.7-0.9%). Results show that ewes with single type in all of lambing had the most percentage of longevity, while ewes with 2 and 3 lambs showed the less survival. However the general policy trend to increased twin type, but according to the

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results, emphasis on twin type removed ewes from the flock in low age and could be decrease longevity in order to improving longevity, the management and environmental conditions should be improved.

Conclusion Illness is the most important reason that cause to ewe left the flock to improve longevity. Thus, in order to decrease the illness, it should be improve the environmental factors (health, feeding, management, Poisoning) and genetic resistance to disease.

Key Words: Culling, Disease, Ewe productive life, Kordish sheep, Lamb survival.

Development of Quantitative Competitive PCR and Absolute Based Real-Time PCR Assays for Quantification of The Butyrate Producing Bacterium: *Butyrivibrio fibrisolvens*

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Introduction *Butyrivibrio fibrisolvens* strains are presently recognized as the major butyrate-producing bacteria found in the rumen and digestive track of many animals and also in the human gut. In this study we reported the development of two DNA based techniques, quantitative competitive (QC) PCR and absolute based Real-Time PCR, for enumerating *Butyrivibrio fibrisolvens* strains. Despite the recent introduction of real-time PCR method for the rapid quantification of the target DNA sequences, use of quantitative competitive PCR (QC-PCR) technique continues to play an important role in nucleic acid quantification since it is more cost effective. The procedure relies on the co-amplification of the sequence of interest with a serially diluted synthetic DNA fragment of the known concentration (competitor), using the single set primers. A real-time polymerase chain reaction is a laboratory technique of molecular biology based on the polymerase chain reaction (PCR). It monitors the amplification of a targeted DNA molecule during the PCR.

Materials and Methods At first reported species-specific primers targeting the 16S rDNA region of the bacterium *Butyrivibrio fibrisolvens* were used for amplifying a 213 bp fragment. A DNA competitor differing by 50 bp in length from the 213 bp fragment was constructed and cloned into pTZ57R/T vector. The competitor was quantified by NanoDrop spectrophotometer and serially diluted and co-amplified by PCR with total extracted DNA from rumen fluid samples. PCR products were quantified by photographing agarose gels and analyzed with Image J software and the amount of amplified target DNA was log plotted against the amount of amplified competitor. Coefficient of determination (R^2) was used as a criterion of methodology precision. For developing the Real-time PCR technique, the 213 bp fragment was amplified and cloned into pTZ57R/T was used to draw a standard curve.

Results and Discussion The specific primers of *Butyrivibrio fibrisolvens* were successfully used for amplifying the specific fragments from this bacteria. The main and important factors for increasing the accuracy of Q-C PCR is the degree of similarity between competitor and target fragment. In this study the competitor fragment was highest homology to target sequences. In this regards it seems obtained results have considerable accuracy. The intensity of bands was evaluated and analyzed using Image J software. The results of band intensity analysis showed linear trend between competitor and target in different serial dilution of competitors. The specific fragment from 16S rDNA region of *Butyrivibrio fibrisolvens* Bacteria was amplified using specific primers and cloned in pTZ57R/T plasmid. After amplifying the competitor fragment and target sequence simultaneously, the two bands were detectable in gel electrophoresis. The range of 10^{-1} to 10^{-6} serial dilution from competitor was selected for QC-PCR reaction. The results of this section showed that the considerable linear correlation was exist between competitor and target fragment in QC-PCR reaction ($R^2=0.985$). In this study, Real-time PCR was also used for quantification of *Butyrivibrio fibrisolvens* Bacteria strain. Melting analysis was showed that the reaction in Real-time PCR had appropriate condition for amplifying the target sequence. We used a standard in this study. This standard was designed as a vector contain of competitor fragment. This kind of standard was successfully used for QC-PCR and Real-time PCR analyses. Standard competitor could be used for absolute quantification for this bacterial strain. Overall, our results showed that the designed standard and optimized QC-PCR have considerable potential for *Butyrivibrio fibrisolvens* Bacteria strain.

Conclusion In this study, the two methods of quantification of nucleotide acids in biological samples were optimized. These two methods were performed in order to optimize *Butyrivibrio fibrisolvens* Bacteria strain quantification. Our results showed that both techniques have the capabilities to use as valuable research methodologies for enumerating the *Butyrivibrio fibrisolvens* strains.

Keywords: *Butyrivibrio fibrisolvens*, Nucleic acid quantification, QCPCR, Real-time PCR.

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Genetic analysis of production traits of Holstein cows in the Mediterranean climate of Iran using random regression and animal model

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Introduction Productive traits such as milk production and fat and protein percentage have economic importance in the livestock industry. Accurate prediction of breeding value of animals is one of the best tools available for maximizing response to selection program. It is a fact that the main objective of the breeding program, is to achieve the maximum economic benefit. For breeders of dairy cattle, milk, fat, and protein are the main sources of income that are the most important traits in the firm goals. For evaluating the dairy cattle based on these traits (milk production, fat, and protein percentage), prediction of breeding values is essential. The present study was performed in order to estimate the genetic and phenotypic parameters and genetic and phenotypic trends of production traits in the Mediterranean climate of Iran (including; Ardebil, Hamadan, East and West Azerbaijan and Zanjan provinces) using 105118 records for Test Day and 30985 records for 305-day lactation records Related 8808 Herd of first lactation Holstein Cattle calving between 2003 to 2013. All records collected by Animal Breeding Center of Iran.

Materials and Methods Records were edited using Fox pro 8.0 and ACCESS 2010 software and the wrong and unusual records were removed from the dataset. All analyses were performed using the RR (random regression) routine of the WOMBAT software package using AIREML algorithm on Linux operation system. Test day records were analyzed with the following random regression model (RRM):

$$Y_{klmptv} = Ostan_k + YS_l + HTD_m + \sum_{a=0}^{i-2} cf(age_n)^a + \sum_{f=0}^k \beta_f \phi_f(dim_t) + \sum_{p=0}^{k_p-1} \alpha_{pr} \phi_r(dim_t) + \sum_{r=0}^{k_r-1} \gamma_{pr} \phi_r(dim_t) + e_{klmptv}$$

Where; P_k ; k th fixed effect of province, YS_l ; l th fixed effect of year-season of calving, Y_{klmptv} ; test day record i obtained at dim_t of cow p calved at the n th age group in herd-test day m , HTD_m ; fixed effect of m th herd-test date, C_f ; The f th fixed regression coefficient for calving age, age_n ; The n th calving age, k ; The order of fit for fixed regression coefficients ($k=4$), β_f ; The r th fixed regression coefficient, ka ; The order of fit for additive genetic random regression coefficients, k_p ; The order of fit for permanent environmental random regression coefficients, α_{pr} ; The r th random regression coefficient of additive genetic value for p th cow, γ_{pr} ; The r th random regression coefficient of permanent environmental effect for p th cow, $\phi_r(dim_t)$; The r th coefficient of Legendre polynomials evaluated at days in milk t , e_{klmptv} ; is The residual effect.

Results and Discussion The heritability of milk yield, fat percentage, and protein percentage during days 5 to 305 of lactation were 0.07 to 0.2, 0.019 to 0.041, and 0.019 to 0.217, respectively. The repeatability of milk yield, fat percentage, and protein percentage during this period of lactation were 0.65, 0.09, and 0.16, respectively. The study of production traits suggested that during the last 10 years in the Mediterranean climate of Iran, Genetic trend of Milk production was positive, but the genetic trend of fat and protein percentage, negative or zero. Figures 2 and 3 clearly indicate that using both types of records; test day records and 305-corrected records, genetic trend for milk production compared with fat and protein percentage was positive and increasing. Heritability of production traits in early lactation was low. The great influence of environment on animals and the negative energy balance are the reasons for the low heritability in this period (2). This amount was being increased and reaches its maximum in the second half of lactation. Increased heritability in the second half of lactation is a function of increasing additive genetic variance and sharp decline in the variance of permanent environment. Similar trends for the results of other studies were reported in the country (1, 14).

Conclusion Recent studies showed that the accuracy of test day records using random regression method was higher than 305 days lactation records. The results of random regression method and Test Day records

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showed that the heritability of milk production is the highest and heritability of fat percentage is. Result of this study showed that, during the last 10 years (from 2003 to 2013) in the Mediterranean climate of Iran, Genetic trend was positive in the amount of milk production, but genetic trend of fat and protein percentage, negative or near zero.

Key words: Holstein cows, Mediterranean climate of Iran, Production traits, Random regression model, Univariate animal model.

Study of BMP 15 Gene in Afshari and Afshari × Booroola Merino Cross Sheep

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Introduction One of the most important sources of the red meat in Iran is the meat produced from sheep. Increasing lamb per ewe considered as a strategy for improving the efficiency for sheep production, although reproduction traits have low heritability. Several genes associated with reproduction were investigated in the recent years. The BMP 15 gene and its paralog GDF 9 and receptor, BMPR-IB, are related to fecundity in sheep and attracted the interest of breeders recently. All these genes that are members of TGF β super family are functionally closely related together and they affect expression and secretion of hormones affecting follicle growth and ovulation rate in mammals. BMP15 plays a key role in regulating many processes in granulosa cells and ovulation rate. Mutations in some candidate genes such as BMP 15 proved to affect the lambing rate. Since 2008, introgression of the BMP Receptor IB mutant (FecB) from Booroola Merino (from New Zealand) into Afshari sheep was initiated. Thereafter, several genes that proved to have an effect on reproductive traits were studied in this breed. This study was conducted to identify possible polymorphism(s) in BMP 15 and to compare its expression in ovaries of pregnant and non-pregnant ewes.

Materials and Methods To study these, blood samples were collected from 35 and 45 Afshari and Afshari × Booroola Merino ewes, respectively. DNA was extracted from all samples using phenol-chloroform procedure and Total RNA was extracted using the RNA extraction kit, CinnaPure RNA Kit (Cinnagen Inc®, Iran), extraction was performed according to the manufacturer's instruction. To remove any possible residual DNA contamination, RNA samples were treated with 1 unit of DNase (Vivantis Inc®, Malaysia). The specific primers were designed for three areas of BMP 15, namely promotor (581 bp), exon one (325 bp) and exon two (857 bp) and the targets were amplified using PCR. The PCR products were sequenced using forward and reverse primer for all of the samples.

Result and discussion There was no difference among sequences of the promoter and the first exon among samples. But, a nucleotide in position 134 of the second exon, C was replaced by A, was observed in two samples with heterozygote genotypes AC instead of CC. Nonetheless, the codon of amino acid encoding proline is remained unchanged. This mutation occurred in two Afshari × Booroola Merino ewes. This mutation, to our knowledge, was not reported to date. Parents of these ewes were not available and also due to the low frequency of the mutation, detection and identification breed of the origin for the mutation was not possible. To date, such a mutation neither was reported in Afshari nor in Booroola Merino breeds. Obviously, the promoter of the gene is conserved and it shows high similarity amongst related species. Nevertheless, in our study sample size was limited to conclude this well. Considering conservation of the promoter of the gene within the species and closely related species, it appears that the regulatory regions were very protected and required for its sustained action. Given that a number of animals used in the study were twine bearing Afshari × Booroola Merino crosses, but there was no difference between them and Afshari pure breed in terms of BMP15 gene expression and gene sequences. Thereupon, the results of this study indicate that this gene plays no role in litter size of this new genetic component. In order to assess the expression of this gene in ovaries of the ewes, after slaughtering, ovary samples of 22 pregnant ewes and those of 8 none pregnant were collected. Total RNA was extracted from the samples and mRNA converted to cDNA using oligo d (T) primer and reverse transcriptase. Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used as the endogenous control for normalization. Results of real time PCR using designed specific primers showed no difference in BMP 15 gene expression between pregnant and non-pregnant ewes.

Conclusion It is possible that this gene plays its role in relation to other genes include their receptors and its expression is needed in different steps of reproduction. This result and reports of other studies suggest that more data on BMP15 gene with a simultaneous expression of other genes in the ovary is needed to clarify the integral role of BMP 15 in reproduction.

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Key words: Afshari sheep, BMP 15 gene, multiple lambing, polymorphism.

The Effects of Continuous and Intermittent Feeding of β -agonist Zilpaterol Hydrochloride on Muscle β -adrenergic Receptors Gene Expression in Feedlot Male Lambs

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Introduction The compounds known as β -adrenergic agonists (β -AA) are organic molecules that have the ability to bind to β -adrenergic receptors (β -AR) and start biochemical reactions that will result in increase of accretion of skeletal muscle and reduction in accretion of fat. The anabolic responses to β -AA are temporary, with a peak time occurring during the first 14 days, after which there is a linear decline in growth response, due to either down-regulation or desensitization of the β -AR. Based on down-regulation of β -AR research, the hypothesis that intermittent feeding of β -AA could enhance response on growth performance was created. The objective of this study was therefore to ascertain the effects of continuous and intermittent use of ZH for a period of 42 d on β -AR gene expression in Lori-Bakhtiari feedlot lambs.

Materials and Methods The continuous feeding of ZH (daily regimen), intermittent 1 d feeding ZH followed by 1 d of withdrawal (1 on 1 off regimen), and intermittent 2 d feeding ZH followed by 2 d of withdrawal (2 on 2 off regimen) were employed as the different feeding methods. Thirty two Lori-Bakhtiari male lambs (initial BW=44±4.7 kg) were assigned to one of four treatments (8 lambs/treatment) based on initial BW and were fed with a diet content of 14% protein and supplemented with 0.2 mg/kg of live weight d⁻¹ZH. The basal diet without ZH was the added control group. For evaluating gene expression, biopsy samples of the semimembranosus muscle were collected from 3 lambs per treatment before ZH supplementation on d 0, and subsequently at d 21, and d 42. Samples were rapidly frozen in liquid N₂. In laboratory after total RNA isolation from muscle, the RNA was then reverse-transcribed into complementary DNA (cDNA). Real time PCR for cDNA samples was performed using an iQ5 BioRad instrument.

Results and Discussion The results of this study showed that the main effects and period × regimen interactions effect was not significant on β 1 and β 2-adrenergic receptors gene expression. Also there were no significant differences between treatments and control group.

Most of the physiological effects of catecholamines are due to specific interactions with β -adrenergic receptors (β -AR). The growth response to β -AA administration commonly is often reduced over time which is possibly a result of desensitization of the β -AR after chronic continuous exposure to the β -AA. The modification in the dosage of β -AA during the feeding period was previously proven to be a useful approach for compensating the reduced response in rats and lambs. Likewise, Hossner (2005) suggested that the intermittent treatment, e.g. treating for 2 days and resting for 2 days is one of the solutions for avoiding the desensitization and down-regulation of receptors. In studies evaluating mRNA responses to β -AA, no change occurred in β 1-AR mRNA abundance due to β -AA treatment. Baxa et al. (2010) explained that no detectable changes of β 1-AR could be attributed to generally low abundance, which results in differences hidden by sample-to-sample variation. We were unable to detect differences in the mRNA expression of β 2-AR mRNA in muscle samples. The decrease in β 2-AR protein expression due to ZH treatment may be in response to a posttranscriptional event.

Conclusion According to our results, it can be concluded that we could use ZH intermittently (1 on 1 off-42 d) in feedlot lambs to decrease fattening costs versus of daily administration of ZH. Further investigation is suggested to detect the alteration of β -adrenergic receptors in continuous and intermittent feeding regimens for clarifying possibly down-regulation or desensitization of them.

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Keywords: β -adrenergic Receptors, Feedlot Lambs, RT-PCR, Zilpaterol Hydrochloride.