



Chemical composition, rumen degradability and fermentation characteristics of fresh *pragmates australis* ensiled with different additives

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Introduction: *Pragmates australis* (Pa) (common reed) is a riverside perennial grass found in wetlands throughout temperate and tropical regions of the world. Pa grows in many wetlands around rivers in Iran. Animal feed restriction is the main problem of Iranian animal production systems and this feed resource can be fed to native livestock especially in rural areas. Ensiling Pa could improve its feeding value. The aim of this study, therefore, was to measure the chemical composition, gas production and rumen degradability characteristics of the fresh and ensiled Pa with different additives.

Materials and Methods: Plant samples were harvested during growth season from the city of Bojnourd, in Iran. The Pa samples were chopped and ensiled into airtight plastic bags as follows: 1) the fresh whole plant of Pa as control (Pa), 2) pa + 4% NaOH, 3) Pa+4% urea, 4) Pa+10% molasses, 5) Pa+4% urea +10% molasses and 6) pa+4% urea + 10% molasses +4% NaOH (on DM basis). Duration of the ensiling process lasted 60 days. Chemical composition of the samples was measured through the ordinary lab methods. The *in vitro* gas production was determined at 2, 4, 6, 8, 12, 24, 36, 48, 72 and 96 hrs intervals after incubation. The *in situ* rumen degradability was also determined at 0, 2, 4, 8, 12, 24, 36, 48, 72, 96 hrs after incubation. The experiment data were analyzed in a completely randomized design.

Results and Discussion: NDF and ADF contents of the ensiled samples with urea were the highest whereas they were the lowest in the NaOH treated samples. CP content of the urea treated Pa was higher than other samples. Ash content of the NaOH treated forage was significantly ($P<0.05$) higher than other treatments. Ammonia nitrogen level of urea-treated silage was highest whereas it was lowest for the Pa treated with the molasses. The urea-treated Pa samples had lowest rate of gas production. Potential of degradability (PD) level of molasses and NaOH treated silages were higher ($P<0.05$) than the control and urea-treated silages. The results of rumen dry matter degradability demonstrated that the quickly degradable fraction of Pa treated with urea and molasses was significantly ($p<0.5$) highest among the silages whereas the value b fraction (slowly degradable) for urea-treated silage and urea and molasses treated silage were higher than other treatments. Quickly degradable fraction of molasses treated samples was lowest among all treatment samples probably because of higher fermentation process in the silages. The c fraction (rate of degradation) of NaOH treated forage was significantly ($p<0.5$) higher than that for other treatments whereas lowest level of c was observed in urea and molasses treated samples. According to the results urea-treatment led to higher crude protein content and NaOH-treating was perfect for destruction of crude fiber of the Pa samples. Effective degradability (ED) of NDF was significantly ($P<0.05$) highest in NaOH treated silage while PD of NDF was highest in Pa treated with urea and molasses. Gas production rate in all silages with different additives was higher ($P<0.05$) than the control samples.

Conclusion: It was concluded that the fresh Pa can be harvested and treated with the appropriate supplements (e.g. molasses and NaOH) and ensiled for subsequent better utilization in ruminant feeding specially in the areas with low rainfall and feed scarcity problem. Quality of Pa silage is directly affected by harvesting time and its lignification. Therefore by harvesting at the appropriate time the Pa could be a perfect forage or silage for ruminant animals.

Key words: Chemical composition, Degradability, Gas production, *Pragmates australis*, Silage.

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The effects of different levels of water restriction on growth performance and blood metabolites of male Baluchi lambs

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Introduction: Sheep has played an important role in meat production and income generation in Iranian farming systems. Their profitability, however, is highly depended on nutrient supply. Drinking water is major nutrient that is responsible for different utilization of nutrients and thereby productivity and gain of animals in most parts of the world. Inadequate water and its quality such as salinity, acidity, and toxic elements depress the biological, physiological process and performance of sheep. Iran has a different type of Agro-ecological climate but most part of this country is under arid or semi arid condition and thus face to shortage of water. Baluchi sheep is the most important breed which is well adapted in harsh marginal arid in the east region of Iran, however, little information is available regarding of the mechanism of adaptation in this breed for biological process to allowing them to cope during water restriction in this zone. Therefore this experiment was conducted to investigate some electrolytes and blood metabolites and hormones under the influence of different levels of water availability and the ability to tolerate water restriction in Baluchi lambs.

Materials and Methods: This study was conducted during May-June 2013 at the Baluchi sheep research center (Abbas Abad) in Northeast of Iran. Twenty-one single lambs, (90 ± 4 days old and 26.7 ± 2.2 kg BW) were used in this study. Lambs were randomly subjected to three levels of water restriction (seven lambs per group). Lambs in control group, allowed to drinking water freely. For the second treatment, the water supply to the lambs were restricted to 72% of their average daily intake and for the third treatment, water intake was restricted to 44% of their average daily intake. The whole experiment period was 49 days and was divided in 3 periods with 14 days adaptation.. The three time periods were including, 7 days for the stepwise water reduction period, and 14 days fixed limit to the amount of 72% and 44% of their average intake for treatment 2 and 3 and during last period (14 days) all groups had free access to water. The sheep were allocated to individual feeding pens and feed were provided ad libitum. Weight gain, blood metabolites and electrolytes (glucose, triglycerides, creatinine, blood urea, cholesterol, total proteins, cortisol, hematocrit, blood hemoglobin, Na^+ , Ca^{2+} , K^+ were determined on 1, 7, 21 and 35 days of experiment. The data from experiments were analyzed using Minitab and GLM procedure and for comparison of means Duncan test was applied.

Results and Discussion: The obtained results indicated that feed intake, daily weight gain and the ratio of water consumption to feed intake in treatments 2 and 3 in compare with control group was statistically different. The results showed that there was a direct relationship between the reduction in water consumption and feed intake. It seems that this weight loss in treatments 2 and 3 can be due to a combination of reduced feed intake and loss of body water. Additional dehydration led to increase blood concentration creatinine, blood urea in compare with the control group. High blood urea and creatinine may be due to an imbalance in the production and disposal of these substances by the kidneys and fecal .Water restriction led to increased concentration of plasma sodium than the control group, however this increase was not significant. Treatment 3, on day 21, compared with other treatments, was an exception and one of the possible reasons for this can be due the influence of the hormone aldosterone and ADH on the kidneys. Hematocrit and hemoglobin in the blood tend to rise by water restriction but this will not led to significant differences between treatments. Except the hemoglobin on day 21 between treatments 3 with other treatments, significant differences were found which can be due to reduced blood plasma volume because of dehydration lambs. Significant differences in blood cholesterol levels were observed between treatments 3 and others on day 21 which probably this increase may be related to mobilization of fat tissue in lambs that are encountered with reducing water consumption.

Conclusion: The overall results showed that water restrictions could lead to a significant reduction in dry matter intake and daily weight gain in Baluchi lambs. In addition, results showed that Baluchi lamb has potential to withstand under water restriction up to 44% of their average daily water intake without significant changes in

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blood electrolytes. This breed can survive and adopt under arid conditions which is common in some desert area in Iran.

Key Words: Blood metabolites, Male Baluchi lambs, Performance, Water restriction.



Effects of different levels of digestible arginine and protein in starter diets containing ideal amino acids ratio on performance, carcass traits and serum parameters of broiler chickens

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Introduction: Nutrition and health during the early days of life have critical effect on broiler chickens performance. It is well known that the diet formulation based on digestible nutrients is superior to formulation based on total nutrients. The suitable supply of essential amino acids in broilers' diets requires proper knowledge of their metabolic effects. The excessive or unbalanced intake of essential and non-essential amino acids can be harmful to broilers' metabolism, due to amino acid antagonisms. Arginine is an essential amino acid for broilers since the urea cycle is not functional in birds. Arginine involves in the synthesis of ornithine, a precursor of polyamines that have a key role in cell division, DNA synthesis, nitric oxide (NO) synthesis, and cell cycle regulation. Arginine also increases the release of insulin, growth hormone, and IGF-A and luteinizing hormone (LH) in the blood stream. On the other hand, in corn-soybean meal based diets, arginine is the fifth limiting amino acid after methionine, lysine, threonine, and valine. Thus, this study was carried out to investigate the effects of different levels of digestible arginine (DA) and digestible protein (DP) of starter diets (1-10 d) based on ideal amino acids ratio on performance, carcass traits and serum parameters in broiler chickens.

Materials and Methods: Four hundred day-old male broiler chickens (Ross 308) were distributed in 10 treatments of 4 replicates (floor pens). The experiment was designed as a 2×5 factorial arrangement in a completely randomized design. Experimental diets were formulated using five levels of digestible arginine (1.05, 1.18, 1.31, 1.44 and 1.57%) and 2 levels of digestible protein (18 and 20%). Chickens were fed experimental diets during 1 to 10 days of age, and then received similar diets formulated according to Ross 308 (2009) recommendations. All birds were provided feed and water ad libitum during the whole rearing period. Temperature was initially set at 32 °C on first day and decreased linearly by 0.5 °C per day up to 42 days and kept constant temperature. During the experiment, the birds received a lighting program (23-h light and 1-h dark cycle) from day 1 to 42. Weight gain, feed intake and feed conversion ratio were measured weekly. Blood samples were collected from wing veins of birds at 10 days age. After 15 minutes, the blood samples were centrifuged at 3000xg for 15 minutes and serum samples were separated into tubes. Then, serums were stored at -20°C until analyses were carried out. At 10 and 42 days age, one bird from each pen with body weight close to the mean of each pen was selected for carcass analyses. After feed withdrawal, the selected birds were transported to the Ferdowsi university pilot for processing. The chickens were slaughtered by cervical dislocation to determine the carcass characteristics. Data were analyzed by analysis of variance using GLM procedures (SAS, 2001). Means were compared using Duncan's new multiple ranges test (Duncan, 1955). The level of significance was reported at $P < 0.05$.

Results and Discussion

The results showed that feed intake, daily weight gain and feed conversion ratio (FCR), as well as their interactions were significantly affected by digestible arginine (DA) and digestible protein (DP) levels in starter diet. The best feed conversion ratio during starter period was related to 20 % DA and 1.31 % DA. DP levels in starter diet had a significant effect on relative weights of liver and abdominal fat (AF) on day 10 and relative weights of gizzard and ileum and also relative lengths of duodenum and ileum on day 42. DA levels significantly affected liver and AF relative weights on day 10. DA and DP interactions had significant effects on relative weight of liver and relative lengths of duodenum, jejunum and ileum on 10th day of age. Effects of DP levels on serum total protein, albumin and phosphorus concentrations were significant. DA levels had significant effects on serum concentrations of uric acid and calcium.

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Conclusion: The results of current study showed that dietary recommendations of digestible arginine and digestible protein by Ross 308 Company improve male chicken's growth performance.

Keywords: Blood parameters, Broiler, Digestible arginine, Digestible protein, Performance.



Effects of different levels of mustard seed treated with FeSO₄-7H₂O on performance and blood metabolites of Japanese quails

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Introduction: In comparison with rapeseed (canola), mustard has agronomic advantages such as drought tolerance and disease resistance, which empower this crop with a considerable oil production potential. Mustard contains toxic substances e.g. glucosinolate, erucic acid, sinapine and tannin. The substances induce unpalatability, growth retardation, low feed efficiency, thyroid gland enlargement and reproductive problems, particularly when the seed is incorporated in the diet at high levels (19, 26). The FeSO₄ treatment of mustard meal is an effective method of detoxification of the meal for using in poultry diets. It can reduce the oxazolidinethione content of the meal by about 88% and the isothiocyanate content by 74% (8).

As mustard seeds are available at a lower price than canola seed, in some regions of Iran, this study was conducted to investigate the effects of different dietary levels of wild black mustard seeds treated with FeSO₄ on growth performance, blood metabolites, carcass characteristics and meat quality of Japanese quails.

Materials and methods: Two-hundred Japanese quails of age seven-day old were randomly assigned to 4 treatment groups with 15 birds in each one of them. The experiment consists of 4 replicates in a completely randomized design. Mustard seeds were treated with FeSO₄-7H₂O according to Dagher and Nawazish (8) procedure. Experimental treatments consisted of control diet (without mustard seed) and diets contained treated mustard seed at 5, 10 and 15% levels. The experimental period lasted up to the age of 35 days. The rearing and management conditions were the same for all groups. Experimental diets were formulated to meet the nutrients requirements of the Japanese quails (20). Chicks had free access to feed and water during the experimental period. Live body weight and feed intake were recorded weekly.

On days 35 of the experimental period, blood samples of one male bird per cage (four birds per treatment) were collected to determine the blood metabolites. After slaughtering, breast muscle was separated and kept frozen at -18°C for 30 days. Breast muscle lipid peroxidation was assessed as thiobarbituric acid-reactive substance concentrations in samples by the method of Tarladgis et al. (27). The breast muscles samples were also used for pH (16), water holding capacity, drip loss (7) and cooking loss (4) tests.

Results and discussion: Percentage of crude protein, ether extract, ash, moisture, and AME_n (Kcal/Kg) of the experimental mustard seed were 28.6, 40.3, 5, 7, 14.8 and 4630, respectively. Using different levels of processed mustard seed had no significant effects on growth performance compared to the control. Using untreated mustard seed (10) and mustard meal (6, 21) at more than 10 percent had detrimental effects on performance. However, it seems that using 15% treated mustard had no adverse effect on growth performance.

Relative weights of the carcass traits (thigh, breast, heart, testis, gizzard, cecum and small intestine) were not significantly affected by the treatments. However, the weight of pancreas tended to be heavier in the birds fed mustard seed ($P < 0.05$). Mustard seed contains considerable amounts of phenolic compounds like sinapic acid and sinapin (10, 11). The phenolic compounds can decrease protein digestion and induce pancreas enlargement. Moheb-Ali (19) reported that weight of pancreas increased linearly as the level of untreated mustard seed increased in the diet. However, few studies have reported canola meal and or mustard meal had no effect on pancreas weight (6, 25, and 26).

Dietary treatments had no effect on TBA, pH and other meat quality parameters. Dietary treatments had also no effect on blood metabolites (triglyceride, LDL and HDL), except glucose. Birds fed diet with 15% mustard seed showed significantly lower glucose level compared to the birds fed diet with 5% mustard seed ($P < 0.01$). In another study, adding untreated mustard seed at 15% level increased LDL and decreased glucose level without any significant effect on other blood metabolites (19). According to Bhattacharjee et al (5), substitution of untreated mustard meal with peanut meal lowered serum protein and increased cholesterol concentration.

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Conclusion: Overall, using processed mustard seeds at %10 level were more economic and can be used as a protein and energy source for quails.

Key word: Blood parameters, Growth performance, Japanese quail, Processed mustard seed.



Effect of *in ovo* injection of different nutrients and 36 h starvation after hatch on hatchability, blood metabolites, intestinal morphology and growth performance of broiler chicks

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Introduction: Many birds do not have access to feed until 48 h after hatching (27). *In ovo* injection technology is a practical means for safe introduction of nutrients into developing embryos, including amino acids, carbohydrates, vitamins, L-carnitine, and hormones which may benefit post-hatch growth and BW gain (19, 36). The objective of this study was to evaluate the effects of *in ovo* injection of amino acids and dextrose, on hatchability, growth performance, blood metabolites, immune organs and intestinal morphology of the broiler chicks.

Materials and methods: The experiment was conducted in a completely randomized design with 5 treatments, 3 replicates of 16 fertile eggs from Ross 308 breeders (28 Week old). Treatments consisted of control (no injection) and injection of 0.7 ml of different nutrients into the amniotic sac of fertile eggs on 17.5th day of incubation including: distilled water (sham), amino acids, dextrin 10% and dextrin 20%. The injection point from the broad end of the egg which was disinfected with alcohol and then 0.7 ml of each solution was injected into the amnion, using a 23- gauge needle with depth of 25 mm. The holes were then sealed using commercial glue. Hatched chicks were fasted for 36 hours. Body weight, feed intake and feed conversion ratio were recorded weekly. On days 1 and 3, blood samples were collected from one chick per replicate to determine serum metabolites (glucose, triglyceride, cholesterol, high density lipoprotein (HDL) and low density lipoprotein (LDL)). On days 1, 3, 7, 14 and 42, one bird per replicate was slaughtered and the relative weight of the immune organs (bursa of Fabricius, spleen and thymus) was determined. On day 3, villus height, crypt depth and villus height to crypt depth ratio were also measured.

Results and Discussion: The results showed that *in ovo* injection of amino acids can lead to heavier birth weight compared to sham and control treatments ($P=0.05$). Chicks hatched from control eggs (no injection) showed the lowest significant weight gain and feed intake. Different treatments had no significant effect on feed conversion ratio. Improved growth performance could be attributed to increase in glycogen stores during the prenatal period (39). Because the late-term embryo, orally consumes the amniotic fluid (comprised primarily of water and albumen protein) prior to pipping, *in ovo* injection of dextrose, amino acids or albumin may help to overcome any nutrient deficiency that may limit embryonic growth. Thus, it was hypothesized that administration of carbohydrates to the amnion may improve the energy level of the broiler embryo and reduce internal energy consumption (proteins and lipids) during pipping, thereby increasing chick BW (45). Glucose is the major energy source in living organisms. Maintenance of glucose homeostasis during few days pre and post hatch is a great challenge in the chick's life. The frequent activity of embryos implies a large amount of energy consumption, and higher glucose demand for fuel (45, 46).

Serum glucose level after hatch was significantly higher in treatment dextrin 20% compared to sham treatment ($P<0.05$). Serum triglyceride level was also higher in control compared to amino acid, dextrose 10% and sham treatments ($P<0.05$). No significant differences were observed for serum metabolites after 36 h starvation. *In ovo* injection of amino acids and dextrin 10% and 20% led to a higher relative weight of thymus at 1 and 3 days of age ($P<0.05$). The relative weight of bursa in 1 day old birds was also significantly higher than control, sham and amino acid treatments ($P<0.05$). Jejunal villus height was significantly higher in chicks injected by dextrose 20% at 3 d of age ($P<0.01$).

The immune system of birds is partly developed at hatch and thereafter grows rapidly in the same way as the development of immune function in mammals (44). For a few days of post hatch, the bursa of fabricius increases in weight and during this time the chick does not have the capability to produce IgG. As a result, after hatching,

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the chick is dependent upon maternal antibody for humeral immune protection. Chicken embryos have limited ability to digest and absorb nutrients prior to hatch (39). This rapid intestinal growth is due to accelerated processes of enterocyte proliferation and differentiation (13). In addition, the intestinal crypts, which begin to form at hatch, are clearly defined several days post hatch, increasing in cell numbers and size (13). Previous studies have shown that feeding immediately post hatch leads to an acceleration of small intestinal morphological development (40), whereas late access to external feed results in delayed development of the small intestine mucosal layer (13).

Conclusion: In conclusion, it seems that *in ovo* injection of nutrients especially 20% dextrin solution may be helpful for improvement of growth performance and immunity in hatched broiler chicks.

Key words: Amino acid, Amniotic sac, Broiler, Dextrin, *In-ovo* injection.



Comparative effects of *Scrophularia striata* with synbiotic and antibiotic on performance and immune response of broiler chickens

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Introduction: Following the ban on antimicrobial growth promoters in poultry nutrition in EU and growing pressure on poultry producers in other parts of the world, there is an increasing interest in searching for growth promoting and immune system-strengthening alternatives. Among the possible alternatives, probiotics, prebiotics, synbiotics (combination of probiotic and prebiotic) and more recently phytogenic products are considered interesting because they have acquired more reliability and acceptability among consumers as safe and natural additives. *Scrophularia striata* (SS) is a plant which grows in the northeastern part of Iran and their immunomodulatory activities of some species of *Scrophularia* have also been reported by other investigators. Due to the lack of study for *Scrophularia striata* effects on broilers performance, and the importance of improving the immune status of broilers, this study was conducted to evaluate the effect of *S. striata* on male broilers growth performance, heterophil to lymphocyte ratio and immunity; and also to compare them with virginiamycin as a well-documented antibiotic growth-promoter.

Materials and methods: Two-hundred and fifty of one-day-old male (Ross 308) broiler chicks were classified into 25 groups. Each group included 10 chicks (five treatments and five replicates per treatment). The five experimental treatments were as follow: basal diet with no additives (control diet) and basal diet containing virginiamycin antibiotic, synbiotic, 0.4 or 0.8 % SS. Feed intake (FI) and body weight gain (BWG) were recorded in different periods and feed conversion ratio (FCR) was calculated. To study the effects of different treatments on blood leukocyte subpopulations, blood samples of two birds from each replicate were collected from the wing vein at the end of experiment. EDTA-containing blood samples were stained subsequently, 100 leukocytes per samples were counted by an optical microscope. The ratio of heterophil (H) to lymphocyte (L) was calculated. Vaccination was carried out according to the routine regional vaccination program and was based on optimal timing of the maternal antibody level. On 7th and 14th days after last vaccination, blood samples were collected from brachial veins, and the sera were used to determine the humoral immune response derived from vaccination against Newcastle disease and infectious bursal disease. Haemagglutination inhibition tests and enzyme-linked immunosorbent assays were used to determine antibody titers of the chickens against Newcastle disease and infectious bursal disease, respectively. The data were analyzed in a completely randomized design by ANOVA using the General Linear Model (GLM) procedure of SAS Institute.

Results and Discussion: The BWG of broilers receiving the antibiotic diet was higher ($P<0.05$) than the control and 0.8% SS groups at age of 11 to 24 days. Compared to the control, feeding synbiotic, 0.4% SS and 0.8% SS also decreased ($P<0.05$) FI at age of 11 to 24 days. However, there were no significant differences in BWG and FCR among treatments during the overall experiment. When broilers were 25 to 42 days of age, supplementing the diet with antibiotic or synbiotic significantly decreased FCR compared with control and 0.4% SS treatments. Moreover, all dietary treatments reduced ($P<0.05$) overall FCR compared with control treatment, where the broilers fed antibiotic diet had best FCR. The results of the present study also showed that blood lymphocyte percentage was greater and heterophil to lymphocyte ratio was lower in antibiotic, synbiotic and 0.8% SS groups compared to control group. Supplementing the basal diet with 0.8% SS increased ($P<0.05$) the relative weights of thymus and bursa of Fabricius at age of 42 days. The birds fed synbiotic or 0.8% SS treatments had also higher secondary antibody titers against Newcastle and Gaboru vaccines ($P<0.05$). However, relative spleen weight and primary antibody response were not affected by dietary treatments. A higher BWG observed in broilers fed antibiotic during the days 11–24 which was not reflected at slaughter age

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might be due to this fact that nutrient requirements of older birds decrease with age and also they have a well-developed digestive tracts and organs. The most efficient FCR in broilers fed diets supplemented with all additives revealed that the impact of growth promoter substances, such as synbiotic and phytogetic products, on performance could be associated with a more efficient use of dietary nutrients, which in turn resulted in an improved FCR. The improvements in broilers FCR by synbiotic and 0.4% SS had similar magnitude to the ones determined for treatment containing virginiamycin, known as a well-documented antibiotic growth promoters. The heterophil to lymphocyte (H/L) ratio is a reliable indicator of the stress response in chicken. The lower H/L ratio observed in the broilers fed the antibiotic, synbiotic and 0.8% SS reflects lower stress levels. Another prominent result from this study is the highest relative thymus weight and secondary antibody response that observed in birds fed synbiotic and 0.8% SS. This could be a main factor for the lack of effect of these treatments on BWG. By stimulation of immune response, nutrients will be applied for production of immunoglobulin antibodies and thus growth rate will be retarded (Khodambashi Emami et al., 2012).

Conclusion: From the results of the present study it could be concluded that the addition of 0.8% *Scrophularia striata* to the broiler chickens diet improve FCR and immune response, which could be considered as effective alternative for antibiotic in broiler chickens nutrition.

Key words: Broiler, Heterophil:lymphocyte, Immune response, Performance, *Scrophularia striata*.



Phenotypic and genotypic analysis of age at first calving in Iranian Holstein dairy cows

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Introduction: Age at first calving (AFC) has an important effect on profitability and reproductive management of dairy cattle. Every month increase in AFC beyond 24 months increases the cost of production. The time between birth and first calving represents a period in which replacement heifers are not generating income. Instead this rearing period requires considerable capital expenditures including feed, housing, and veterinary expenses. These expenses constitute 15% to 20% of the total expenses related to milk production. A basic approach to reduce this cost is to decrease the time between birth and her first freshening. Worldwide recommendations for one particular AFC might be an incorrect management goal for all of the cattle on all of the farms, since the recommendation might not represent the management goals and/or capabilities of a particular production system or farm. We realize that each dairy has its own set of unique management and environmental conditions, which makes a universal AFC and BW after first calving, a difficult goal to achieve. The AFC has a profound influence on the total cost of raising dairy replacements in which older calving heifers are more expensive to raise than younger ones.

Materials and methods: A total of 19499 calving records belonged to 96 herd from 1996 to 2008 were used to estimate genetic components and genetic trend for age at first calving in Holstein dairy cows of Iran. Data were analyzed using a univariate model and Wombat software. Linear regression of estimated breeding values on calving year was used to estimate genetic trend.

Results and Discussion: Estimated genetic trend was positive for some years and was negative for others and showed that reducing age at first calving has not been considered in the selection strategies; however, the phenotypic trend was decreased. The age at first calving for Yazd, Markazi, and southern Khorasan provinces were the highest and for Kermanshah, East Azarbayjan, and Ardebil provinces were the lowest compared to the other provinces. Most analysis shows that the financial benefit afforded to heifers that freshen at a low AFC seems to at the least offset any milk lost in the first lactation. The NRC (2001) suggests a post weaning BW equal to 82% of her mature body weight. This can be attained with a maximal pre-pubertal ADG of 2.0 lbs/d if a traditional pre-weaning program is employed or 1.8 lbs/d if an intensified pre-weaning program is employed. Due to the well-defined link between insufficient BW at calving and increased mortality and disease in first calf heifers, attaining this aim post calving BW is of critical importance. Ettema and Santos (2004) conducted an economic analysis of the AFC study that was discussed above. Rearing prices for the medium and high AFC groups were \$40.34 and \$107.89, respectively, more than that of the low AFC collection. Income for each AFC collection was adjusted for the cost of rearing, assessed feed to increase milk yield, stillbirths, diseases, open days, culling, mortality, labor cost, and the value of milk and calf produced as well as the value of a cow at the end of the 310 day studies. Adjusted income was \$119.73 and \$9.08 more for the medium and high AFC, respectively, than for the low AFC. These values were not significantly diverse implying no single AFC had an economic benefit over another. Nevertheless, these authors (Ettema and Santos, 2004) did not study the net present value of money in their analysis as St-Pierre (2002) did. If this had been considered, it would presumably shift the economic improvement to the low AFC heifers.

Conclusion: Good climatic and weather conditions can be effective factors for reducing the age at first calving and cause to increase the fertility of heifers. However, management methods had a significant effect on this trait in some provinces. The primary benefits of reducing AFC include reducing rearing costs as well as reducing the amount of time

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in which the heifer is only a capital drain on farm resources. The primary disadvantage of reducing AFC is that it is frequently associated with a reduction in first lactation milk yield. Despite this reduction in first lactation milk yield, production per year of herd life is usually increased by reduced AFC. First lactation may be influenced by AFC, future lactations are definitely not. Furthermore, stay ability and health of cows is not influenced by reduced AFC as long as first calf heifers freshen at an adequate weight.

Keywords: Age at first calving, Dairy cattle, Genetic trend, Phenotypic trend, Profitability.



Estimation of population stratification in crossbred and inbred dairy cattle using genome wide association by simulation

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Introduction: Domestic animals are invaluable resources to study the molecular architecture of complex traits. Although the mapping of quantitative trait loci (QTL) underlying economically important traits in domestic animals has achieved remarkable results in recent decades, not all of the genetic variation in the complex traits has been captured due to the low density of markers used in QTL mapping studies. The genome wide association study (GWAS) utilizing high-density single-nucleotide polymorphism (SNP), provides a new way to tackle this issue. Genetic association tests identify differences in allele frequency between cases and controls. Population stratification can be a problem in association studies, such as case-control studies, where the association found could be due to the underlying structure of the population.

Material and Methods: In current research, Genome wide association technique (Case-control design) was used to evaluate population stratification. Historical population and genome of 10000 cattle were simulated along 100 generations by Mutation-Drift Equilibrium (MDA) technique. By using historical population, 800 inbred and cross bred cattle with ~50000 SNPs on 30 chromosomes were simulated. Genomic control was performed to survey markers with a low prior probability of association with trait ("null markers") and to estimate population stratification by Q-Q plot and lambda statistics.

Results and discussion: Deviation of cases/controls ratios between inbred subpopulations causes increasing lambda and population stratification; as lambda was estimated 0.42, 11.31 and 97.77 in additive genetic model with case/control ratios 1.00, 0.77 and 0.33, respectively and 0.47, 8.21 and 57.40 in co-dominant genetic model. Therefore, the more disparate composition cases/controls the more population stratification. When cases and controls were drawn from different randomly mating breeding populations, allele frequencies were different, but these differences may not be related to disease status or complex trait. This means that the assumption of independence of observations is violated. Often this will lead to an overestimation of the significance of an association but it depends on the way the sample is chosen. Population stratification was surveyed between two random groups of crossbred population (400 cases, 400 controls). There was no population stratification among subpopulations of crossbreds in current research; as lambda was estimated 0.55, 0.66, 0.89, 0.76 and 0.41 in co dominant, dominant, recessive, over dominant, additive genetic models, respectively.

Conclusion: The main GWAS problem in inbred cattle is population stratification. When cases and controls are drawn from different inbreeding populations, Population Stratification occurs. Lambda and PS is related to Cases/controls ratio among inbred lines, as more deviation ratio of one, more population stratification. It is suggested to control population stratification inbred cattle should not be used unless exactly equal ratio of cases/controls between inbred subpopulations can be achieved and it is better to use of crossbred cattle for genome wide association studies.

Key words: Association Studies, Population Stratification, SNPs.

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Study of pedigree structure and effects of inbreeding depression on growth traits in Lorestan native sheep

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Introduction: Inbreeding is defined as the probability that two alleles at any locus are identical by descent and occur when related individuals are mated to each other. The initial consequence of inbreeding is inbreeding depression reducing the performance of growth, production, health, fertility, and survival traits. This concern has become more serious in animal breeding nowadays, in which selection responses are maximized using animal model best linear unbiased predictors (BLUP) of breeding value. The use of these breeding values alone may result in more closely related selection candidates preferred for selection, with increased levels of inbreeding since they share most of their familial information. The unavoidable mating of related animals in closed populations leads to accumulation of inbreeding and decreased genetic diversity. Inbreeding has deleterious effect on additive genetic variance as well as on phenotypic values. Heterozygosity and allelic diversities can be lost from small, closed, selected populations at a rapid rate. The rates of inbreeding must be limited to maintain diversity at an acceptable level so that genetic variation will ensure that future animals can respond to changes in environment. Inbreeding depression has been found in a large proportion of species examined. Lori sheep breed is mainly raised in Lorestan Province in Iran. This study was conducted to identify pedigrees and inbreeding depression on growth traits in Lori sheep population from 2001 to 2010. The rate of inbreeding needs to be limited to maintain diversity at an acceptable level so that genetic variation will ensure that future animals can respond to changes in the environment and to selection. Without genetic variation, animals cannot adapt to these changes.

Material and Methods: For this study pedigree information and body weight at different ages (birth weight, weaning weight, 6-month weight, and 9-month weight) of 6440 lambs from 273 rams and 1955 ewes during the years 2001 to 2010 from Lorestan Agricultural and Natural Resources Research Center were used. The pedigree corrections were done by the Excel program and the estimation of inbreeding coefficients was done by pedigree software. To study the pedigree, Pedigree, program, or estimate inbreeding coefficient, CFC program, and for calculate the amount of inbreeding citizenship traits, wombat software were used.

Results and Discussion: In total population the number of inbred animals were 2126 (33%) of the herd. The average total population inbreeding coefficient and average inbreeding coefficient inbred of population were 0.69% and 2.24%, respectively. Annual changes in inbreeding coefficient was 0.21, which was statistically significant ($p < 0.0001$). The maximum and minimum amounts of inbreeding were 26.85% and 0% respectively. In this pedigree, low amount of inbreeding coefficient was owing to some unknown pedigree of animals, management changes, and partially inhibit intercourse near relationship. With increasing every 1 percent in inbreeding, 4 g for birth weight, 20.3 g for weaning weight, 247 g for month 6 and 4.5 g for month 9, was decreased, respectively. The results of this study, confirm the low level of inbreeding in the herd. Applying a designed mating system like crossbreeding could be a suitable method to avoid inbreeding depression via keeping the level of inbreeding under control. The utilization of a program for directed mating in the present flock is an appropriate alternative to keep the level of inbreeding under control.

Conclusion: The results of present experiment showed that the rate of inbreeding is very low and it is not worrying in Lorestan native sheep.

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Keywords: Animal model, Growth traits, Inbreeding depression, Inbreeding trend, Lori sheep.



A study on accuracy of predicted breeding value for body weight at eighth week of age in Khorasan native chickens

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Introduction: Genetic resources in any country are valuable materials which needed to be conserved for a sustainable agriculture. An animal phenotype is generally affected by genetic and environmental factors. To increase mean performance in a population under consideration not only environmental conditions, but also genetic potential of the animals should be improved. Although, environmental improvement could increase the level of animals' production in a more rapid way, it is not a permanent and non-cumulative progress. In any breeding schemes prediction breeding value of the candidate animals is needed to be obtained with a high precision and accuracy for making a remarkable genetic gain for the traits over the time. The main objective of the present research was to study accuracy of predicted breeding value for body weight at eighth week of age in indigenous chickens of Khorasan Razavi province.

Materials and methods: A set of 47,000 body weight (at the age of eight weeks) records belonging to 47,000 head of male and female chicks (progeny of 753 sires and 5,154 dams) collected during seven generations (2006-2012) was used. The data were obtained in Khorasan Razavi native chicken breeding center. An animal model was applied for analyzing the records. In the model, contemporary group of generation*hatch*sex (GHS) as a fixed effect, weight at birth as a covariable, as well as direct and maternal additive genetic random effects were taken into account. In an initial analysis using SAS software, all fixed and covariate factors included in the model were detected to be significant for the trait. All additive genetic relationships among the animals in the pedigree file (47,880 animals) were accounted for. Variance and covariance components of direct and maternal additive genetic effects were estimated through restricted maximum likelihood (REML) method. Breeding value of the animals was obtained by best linear unbiased prediction (BLUP). Selection accuracy was then calculated based on prediction error variance (PEV). The model was fitted to the data using DMU package. Post analysis of breeding values (genetic trend estimation and statistical comparison of groups using student's *t* test) was also undertaken using SPSS software.

Results and Discussion: Average and standard deviation of body weight at the age of eight weeks were 607.93 g. and 127.347 g., respectively. As expected, males (668.98 g.) were generally heavier than females (549.86 g.) chickens. Additive and maternal genetic variance components were 3183.9253 and 350.8929, respectively. Based on genetic covariance (-363.8555) the correlation between direct and maternal genetic effects was revealed to be -0.3442. Direct and maternal heritability for the trait were found to be 0.4387 and 0.0483, respectively. Mean direct and maternal breeding values were 76.65 g. and -7.91 g., respectively. The corresponding figures for the direct and maternal accuracies were 0.741 and 0.427, respectively. Genetic trends for direct and maternal breeding value were 26.951 g. (SE=1.344 g.) and -2.252 g. (SE=0.199 g.), respectively and statistically significant ($P < 0.0001$). For the sires, mean direct breeding value (90.77 g.) was greater than the mean maternal breeding value (-9.18 g.) and the same pattern was also found for mean selection accuracy (0.89 vs. 0.61). For the sires, a high positive and significant Pearson correlation was obtained between the number of progeny and selection accuracy for direct (0.885) and maternal (0.683) breeding values. However, both direct (-0.071) and maternal (0.052) breeding values had non-significant correlation with the number of progeny. A high significant positive correlation was also found between direct and maternal selection accuracies (0.724).

Conclusion: The findings of the current research indicate that body weight at eighth week of age in

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indigenous chickens of Khorasan Razavi province has genetically been evaluated with a rather high degree and acceptable precision. High estimate of direct heritability of the trait indicates that there is a substantial genetic potential in the population to be improved over the generations. Moreover, the results also suggest that the number of progeny is an important factor for increasing selection accuracy.

Keywords: Animal model, Body weight, Indigenous chickens, Selection accuracy.



Effects of garlic (*Allium sativum*) extract on growth performance, survival rate, some hematological and biochemical indices of Gourami (*Trichogaster trichopterus*)

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Introduction: Nowadays, the healthiness and high quality as well as quantity of aquatic are of great importance and the role of diet components is well recognized (20). Fish needs energy and nutrients in order to achieve desirable level of growth and development and reproduction (11). In recent years application of immune system stimuli has been widely adopted in order to enhance the immune system of fish and non-specific immune responses and protect against disease. So it seems that using stimulator of the immune system is the perfect solution for the control of aquatic animal diseases. Using chemical drugs and antibiotics in aquaculture have consequences, including the risk of pathogens resistant to these drugs, the drugs persistence in the meat and fish as well as environmental pollution. Taking herbs and medicinal plants such as garlic fraught with same effect as antibiotics can be served as an alternative to medications and antibiotics.

Materials and methods: In this study, 180 gourami fish with average weight 0.10 ± 4.33 were prepared from reproduction and breeding center of ornamental fish in Golestan province. To add the garlic oil to the diets, the standard meal was purchased from commercial fish feed company of Mazandaran province and were ground using a mortar. Then, garlic oil at levels of 0, 0.10, 0.15 and 0.20 g/kg was added to water and mixed. Fish were fed for 8-weeks in three times by 3% body weight per day at hours 8-14-20. Fish diets and their growth rate monitored by conducting bio-survey at the beginning and during the growth for every 15 days. 10 fish were sampled from each replicate to take serum. After anesthesia using 0.7 ml 2-phenoxyethanol, fish were dried, and about 2 ml of blood was taken by cutting the tail. In hematology laboratory, number of white blood cells, red blood cells, hemoglobin and hematocrit were measured according to standard methods. Glucose levels and serum protein were measured by spectrophotometry using a diagnostic kit (Sigma, America). Data analysis was performed using one-way analysis of variance (ANOVA) in SPSS software. Significant differences ($P < 0.05$) between means (mean separation) were obtained using Duncan test. All data were calculated as the mean \pm standard deviation.

Results and Discussion: The treatment characterized with 0.20 g garlic/kg had the highest weight gain and the lowest specific growth rate, respectively. The results showed that the final weight, weight gain and feed rate were increased especially using the diets with garlic oil. The results are consistent with findings of others who reported the final weight, weight gain and feed rate, especially in the Nile tilapia fish especially in garlic treatments significantly increased compared to the control group. The highest hematocrit, hemoglobin, white blood cells and red blood cells were found in the highest dose of garlic essential oil (0.20) which significantly differed between the control group and the other treatments, however, this was not true for MCV, MCH and MCHC among the treatments. This study showed that treatment with garlic supplement caused a significant increase in blood parameters, this is confirmed by Martins et al. (12). In another study Shalaby et al. (15) reported similar results with Nile tilapia fish that were fed by diets containing garlic. The lowest blood glucose levels was found using the highest dose of garlic oil by a significant difference between the control group and the experimental treatments, whereas the maximum amount of protein in blood of fish at the same treatments were significantly different from the control group and the other treatments. In the present research, the

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concentration of glucose in the groups fed diets containing garlic treatments was lower than the control group, which is in agreement with Shalaby et al. (15) in the Nile Tilapia. Significant increase was found in blood plasma proteins in fish which fed by different dose of garlic similar to results of Hussein (8) who reported that the total amount of protein in the blood plasma of rats fed diets containing garlic oil increased.

Conclusion: Using garlic oil in spot gourami fish diet plays a positive role in growth and enhance immune function so that 0.20 g/kg dose of garlic oil in the diet may be recommended for gourami fish.

Keyword: Feed Conversion Ratio, Glucose, Hematocrit, Protein.