Two repeated experiments were conducted to investigate the effects of different stocking densities on broiler performance, carcass characteristics, litter quality and foot lesions using 5224 one day-old Ross 308 broilers. In each experiment, a total of 2612 one day-old broilers were allocated in 16 floor pens at densities of 16, 18, 20 and 22 birds/m² of floor space in an environmentally controlled broiler house. Data collected from the two experiments were pooled together for statistical analysis. The results indicated that body weight gain significantly decreased with increasing stocking density (P<0.05). Density beyond 16 birds/m² significantly decreased the body weight gain at 1 to 21 day of age and density beyond 20 birds/m² significantly decreased the body weight gains at 22 to 42 day of age and in whole experimental period (P<0.05). Body weight gain expressed as kg BW/m² increased linearly with increased stocking density (P<0.05). Treatments had no significant effects on feed conversion ratio, mortality rate, production efficiency index (PEI), carcass dressing measurements (carcass weight and relative weights of liver, abdominal fat, bursa of fabricius, spleen and parameters of the broiler litter (moisture, pH and ammonia level) (P>0.05). Foot pad lesions significantly increased as stocking density increased (P<0.05), but hock burns was not affected by increasing stocking density. In conclusion, based on the obtained results of this study, 20 birds/m² of floor space was an appropriate density for rearing broilers in an environmentally controlled broiler house.

**KEY WORDS** broiler chicken, foot lesion, litter, performance, stocking density.
house considerably intensify negative effects of higher stocking density on birds (Dawkins et al. 2004; Puron et al. 2005; Estevez, 2007; Knizatova et al. 2010). The consequences of increasing bird density on environmental conditions of within poultry house are include of changes in environmental quality such as outbreak of wet and caked lit-
ter, odors and ammonia emissions, etc. (Estevez, 2007).

Increasing stocking density must be done according to the house environmental conditions, equipments, ventilation, cooling and heating systems, drinker and feeder space, etc. otherwise, it can have deleterious impacts on birds welfare, health and performance (Czarick and Lacy, 1990; Lacy and Czarick, 1992; Puron et al. 1997; Estevez, 2007; Yardimci and Kenar, 2008). In other hands, efficacy of higher density in production is dependent on technical parameters includes cooling, heating and feeding systems and management conditions such as ventilation and litter quality. It means that increasing stocking density in environmental conditions fully controlled poultry house in comparison with conventional houses can have less negative impacts on environment of within house and poultry performance as long as the optimal environmental conditions (temperature, ventilation, humidity) are provided (Yardimci and Kenar, 2008). Dawkins et al. (2004) concluded that environmental conditions includes of air and litter quality had more direct impact on broiler welfare than stocking density. In this study, the impacts of increasing broiler density from 16 to 22 birds/m² of floor space on growth performance, carcass characteristics, litter quality as well as footpad lesions and hock burns were investigated in an environmentally controlled broiler house.

**MATERIALS AND METHODS**

**Experimental treatments**

This study was conducted in an environmentally controlled broiler house in Khuzestan Province, Iran. Building characteristics, ventilation system and equipment of the broiler house were presented in Table 1. Average daily dry bulb temperature and relative humidity of ambient and inside of the barn were weekly recorded during both experiments and data of two trials were pooled (Table 2). A total of 16 floor pens were allocated to placing the chickens. Pens dimensions were 2.5 × 3.44 (8.60 m²). Height of the pens was 65 cm. Two repeated trials were conducted using 5224 as-hatched one-day-old broiler chickens (Ross 308) in two 42-day periods. In each experiment, a total of 2612 one-day-old unsexed broiler chicks were provided from a local hatchery and were placed into prepared floor pens at four stocking densities of 16, 18, 20 and 22 birds/m². The pens arranged in the center of the house and to simulate actual conditions, the pens were surrounded by a commercial broiler chicken flock composed of birds of the same origin as those used in the experiments.

<table>
<thead>
<tr>
<th>Table 1 Building characteristics, ventilation design and equipment of the environmentally controlled broiler house</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Characteristics</strong></td>
</tr>
<tr>
<td>House sidewalls (R-value, mm)</td>
</tr>
<tr>
<td>Roof type</td>
</tr>
<tr>
<td>Floor type</td>
</tr>
<tr>
<td>House width × length (m)</td>
</tr>
<tr>
<td>House height (m)</td>
</tr>
<tr>
<td>Ventilation system</td>
</tr>
<tr>
<td>Fan numbers and capacity (m³/h)</td>
</tr>
<tr>
<td>Maximum ventilating capacity (m³/h)</td>
</tr>
<tr>
<td>Evaporative cooling system</td>
</tr>
<tr>
<td>Number and surface area of evaporative cooling system (m²)</td>
</tr>
<tr>
<td>Housing capacity (birds)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Table 2 Temperature and relative humidity (RH) values of the broiler house during the study¹</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Days</strong></td>
</tr>
<tr>
<td>Max°C</td>
</tr>
<tr>
<td>1 to 7</td>
</tr>
<tr>
<td>8 to 14</td>
</tr>
<tr>
<td>15 to 21</td>
</tr>
<tr>
<td>22 to 28</td>
</tr>
<tr>
<td>29 to 35</td>
</tr>
<tr>
<td>36 to 42</td>
</tr>
</tbody>
</table>

¹Data of both experiments were pooled and presented weekly. Max: maximum value; Min: minimum value; Avg: average value and RH: relative humidity.

The numbers of chickens per each density treatments (per each pen) during the two rearing periods were as follows: 1) stocking density 16 birds/m²: 546 birds/treatment (137 birds/pen), 2) stocking density 18 birds/m²: 620 birds/treatment (155 birds/pen), 3) stocking density 20 birds/m²: 688 birds/treatment (172 birds/pen) and 4) stocking density 22 birds/m²: 756 birds/treatment (189 birds/pen). Each treatment was replicated four times. Each pen was covered with fresh unused wood shaving as litter bedding material at about 5 cm depth and equipped with 13 nipple drinker and two tube feeder. All of the pens of each density treatments had equal feeding and drinking space from pen feeders and drinkers. The feeding space in each pen per densities of 16, 18, 20 and 22 birds/m² was 2.37, 2.10, 1.89 and 1.72 cm per bird.

**Bird husbandry**

The conditions and standards of rearing used in this research were approved by the Ethics Committee for Animal Experiments of the Animal Science Research Institute of Iran. All of the broiler chickens were fed with a basal corn-
soybean meal diet during 1-42 d of age. Corn and soybean meal-based diets were formulated to meet nutritional requirements of broilers according to NRC (1994) broiler recommendation. The composition of experimental diets and their nutritive characteristics are presented in Table 3.

Feed and water were provided *ad libitum*. All diets were provided as mash form. In 2 first days of the two experiments, lighting schedule was continuous and afterward, a 23L: 1D lighting schedule was provided. In 2 first days of the experiments, lighting schedule was continuous and afterward, a 23L: 1D lighting schedule was provided. House temperature was maintained at 32 °C at the beginning of the experiments and reduced as the birds progressed in age to around 28 °C at 22 d and thereafter reduced to about 22 °C at 24 day of period and maintained until 42 day.

### Measurements

#### Broiler performance

The broiler chickens were weighed as a group at 1, 21 and 42 day of age. All birds were weighed by pen and live body weight gain and feed intake was determined. Feed conversion ratio (FCR) was calculated after adjusting for daily mortality. Production efficiency index (PEI) was determined according to below equation after calculating liability percentage (Liability % = 100–mortality %) and feed conversion ratio:

\[ \text{PEI} = 100 \times (\text{Liability} \% \times \text{final body weight (kg)} / \text{time period (day)} \times \text{FCR}) \]

#### Carcass characteristics

At the end of each experiment (day 42), four birds with an average weight of each replicate (2 male and 2 female, 16 birds per each treatment) were randomly selected, weighted and killed by cervical dislocation. The carcass without giblets was weighed and expressed as a percentage of its initial live weight and considered as the carcass yield. The weights of abdominal fat pad, liver without bladder-gall, spleen and bursa of fabricius were recorded and their relative weights as percentages of the live weight were determined.

#### Litter quality

At day 21 and 42 of age, litter samples were collected from different positions of each pen for determination of pH (1:10 litter per distilled water) and moisture content (at 105 °C for 24 h). Each litter sample consisted of 6 subsamples of litter. The subsamples were collected from the corners of each pen and from the center. At day 42, emissions of litter ammonia were determined by the farm method described by Chamblee and Yeatman (2003). Briefly, after putting a 3.5 container fitted with a manual air-stirring device, an air-sampling port was inverted on the litter and after 5 min. an air sample was drawn into an ammonia detector tube (Ammonia detector tube, 3La type, Gastec Co, Japan) by a manual pump (Gas sampling pump, GV-100S, Gastec Co, Japan).

#### Foot lesions

At day 42 of both experiments, frequency of footpad lesions and hock burns, based on visible swelling or injuries, were studied objectively on 40 randomly selected birds (20 male and 20 female) from each pen according to method described by Thomas et al. (2004). The foot pad and hock burns were scored using a 3-point scale: 1 = no lesions or burns, 2 = mild lesions or burns and 3 = large and deep lesions or burns.

#### Statistical analysis

Data were analyzed according to the GLM model procedure of SAS (1996) as a complete randomized design with 4 densities of 16, 18, 20 and 22 birds/m² of floor space. Mortality data were transformed to arcsine-square root before analysis. Data of both experiments were pooled and significant differences among treatments were determined by
Duncan’s multiple range test at P < 0.05. Each pen was used as experimental unit. Frequency analysis was conducted for the data on the incidence of footpad lesions and hock burns, using frequency procedure of SAS (1996).

**RESULTS AND DISCUSSION**

**Growth performance**

The effects of different stocking densities (16, 18, 20 and 22 birds/m² of floor space) on broiler performance included of body weight gain, feed intake, feed conversion ratio at day 1-21, 22-42 as well as mortality rate at 1-42 day of age were presented in Table 4. At day 21, body weight gain of broilers grown at densities of 18, 20 and 22 birds/m² significantly decreased compared to the 16 birds/m² stocking density treatment (P<0.05) and there were no significant difference between densities of 18, 20 and 22 birds/m².

Feed intake was not affected by stocking density increases at day 1-21 day of age. At day 22-42, the results of body weight gain were somewhat complicated. By comparing the treatments, the only significant difference observed between 18 and 22 birds/m² stocking density treatments (P<0.05). At this period, feed intake significantly decreased in the broiler chickens grown at 22 birds/m² compared to 16 and 18 birds/m² stocking density treatments (P<0.05). At day 1-42, broilers grown at 22 birds/m² significantly had a lower body weight gain and feed intake compared to 16 and 18 birds/m² treatments (P<0.05). In the present study, reduced body weight gain was in agreement with the results taken by other researchers who reported lower body weight gain as stocking density increased (Shanawany, 1988; Puron et al., 1995; Feddes et al., 2002; Dozier et al., 2005a; Dozier et al., 2005b; Dozier et al., 2006; Mtileni et al., 2007).

At day 21, lack of the significant effect of stocking density on feed intake was in agreement with the results published by Dozier et al. (2006) and Ravindran et al. (2006). But, decrease in broiler feed intake in stocking density of 22 birds/m² compared to 16 or 18 birds/m² at days 22-42 and whole of the experiment was in agreement with those studies which reported that decrease in feed intake was related to less feed consumed by birds grown at higher densities due to inhibited access to feeder spaces (Shanawany, 1988; Estevez, 2007; Dozier et al., 2005b). In the present study, like other studies, feeder space and drinker counts were equal in the all of the pens (Feddes et al., 2002; Guardia et al., 2011; Zhang et al., 2011). while, in some studies, feeding and drinking spaces increased by increasing feeder and drinker counts (Dozier et al., 2005b; Dozier et al., 2006).

Zuowei et al. (2011) showed that adequate feeder space can be beneficial for supporting the feed consumption of broiler chickens under high stocking density, whereas, Haansen and Becker (1960) confirmed that even with maintaining constant feeding space per bird, the deleterious effects of higher stocking density on birds yet be existed. In the current study, feeder and drinker spaces were equal in each stocking density treatments. Feddes et al. (2002) reported that increasing broiler density from 5 to 20 per each nipple drinkers had no substantial effect on performance and carcass characteristics of broiler chickens. They concluded that each nipple drinker was enough for every 20 broiler chickens to 2 kg body weight. In the present study, all of the pens had 13 nipple drinkers which there were about 10 to 14 birds per each nipple for densities of 16 to 22 birds/m², respectively which had no disagreement with experiment conducted by Feddes et al. (2002). Since each tube feeder was provided for 50 birds, the existence of 2 tube feeder in each pen somewhat decreased feeder space in densities of 20 and 22 birds/m² (include 172 and 189 birds/pen, respectively). Furthermore, it seems that providing more feeding or drinking space for birds may intensify negative impacts of high stocking density on birds because of occupied floor spaces by equipments which decrease movement ability of birds and increase stress among birds (Estevez, 2007).

Increasing stocking density had no significant impact on feed conversion ratio and mortality rate at all of the days of age and production efficiency index (PEI) at day 42 of age. The reported results by other researchers indicated that stocking density has no effect on FCR (Proudfoot et al., 1979; Cravener et al., 1992; Martrenchar et al., 2000; Feddes et al., 2002; Ventura et al., 2010; Skeroglu et al., 2011) and these findings were confirmed in this study. In contrast with these results, Houshmand et al. (2012) reported that density of 10 birds/m² of floor space significantly lower FCR compared to 16 birds/m² of floor space. Insignificant effects of higher stocking density on mortality rate was reported in some studies (Proudfoot et al., 1979; Shanawany, 1988; Cravener et al., 1992; Martrenchar et al., 1997; Feddes et al., 2002; Thomas et al., 2004; Skeroglu et al., 2011). Estevez (2007) stated that incidence of greater mortality rate in higher stocking density was related to heat stress created among birds. In higher densities, all of the floor space covers with birds and the heat of the litter cannot be removed effectively by the usual ventilation system.

Therefore, birds may suffer from heat stress even under moderate climatic condition (Lolli et al., 2010). Although it was well established that growth rate decreases as stocking density increases but the increase in stocking density may has the economic advantage of maximum return per square meter of floor space (Proudfoot et al., 1979; Shanawany, 1988; Cravener et al., 1992; Puron et al., 1995). The kg BW/m² of floor space significantly influenced by stocking density (16, 18, 20, 22 birds/m²) treatments (P<0.05) (Table 5).
The kg BW/m² of floor space significantly increased as stocking density increased. Stocking density of 22 bird/m² had the highest (38.16) kg BW/m² compared to other densities of 16 (29.06 kg/m²), 18 (33.40 kg/m²) and 20 (35.67 kg/m²) (P<0.05).

Carcass characteristics
In the present study, stocking density had no significant influence on carcass yield and relative weight of liver, abdominal fat pad, bursa of fabricius and spleen (Table 5). Studies conducted on effects of stocking density on carcass characteristics are scarce. Bilgili and Hess (1995) demonstrated that different stocking density (1.0, 0.9 and 0.8 ft²/broiler) had no significant impacts on carcass yield and abdominal fat pad. Jayalakshmi et al. (2009) by evaluating stocking densities of 900, 750, 600 and 450 cm²/broiler confirmed that abdominal fat pad and relative weight of giblets were not affected by stocking density, while higher stocking density declined carcass yield. In contrast, Castellini et al. (2002) and Simsek et al. (2009) reported a significant increase in body fat reservation by increasing stocking density and attributed this effect to reduced movement ability of birds. Insignificant effect of stocking density with published results of other researchers (Turkyilmaz, 2008; Buijs et al. 2009). In contrast, Heckert et al. (2002) demonstrated that relative weight of bursa of fabricius decreased as increasing stocking density from 10 to 20 birds/m² of floor space.

Litter quality
The results regarding the impacts of different stocking density on pH, moisture and ammonia concentrations of litter were presented in Table 6. Stocking density had no significant effect on litter moisture and pH at days 21 and 42 of age. Published results by Zhang et al. (2011) indicated that stocking density had no impact on litter pH. Litter pH is one of important factors in ammonia production in poultry houses (Coufal et al., 2006) but it seems that litter pH is less affecting by stocking density regarding the results of this study.

Stocking density had no any significant effect on litter moisture. In contrast, Dozier et al. (2005) reported that litter moisture increased as stocking density increases. Ammonia emissions from broiler litter in response to different stocking densities were not affected at day 42 of age as shown in Table 6.
These findings are in agreement with the results reported by those researchers that showed different stocking density had not significant effect on litter ammonia production (Dozier et al. 2005b). It seems that the caked litter under birds at higher stocking density presumably was a main factor in the variability of litter moisture and ammonia production results. As indicated by Dozier et al. (2005b), increasing stocking density elevated litter moisture, but litter ammonia production was not affected likely due to increased amount of caked litter in the pens with high stocking density which suppressed ammonia volatilization.

Foot lesions
The results of incidence and severity of foot pad lesions and hock burns are shown in Table 7. The incidence and severity of foot pad lesions were significantly affected by different stocking densities (P<0.05), as increasing stocking density from 20 to 22 birds/m² significantly decreased the number of broilers with normal foot pad (score 1) and increased the number of broilers with footpad lesions (score 2 and 3), particularly in density of 22 birds/m² compared to other densities. There were no significant difference between densities of 16, 18 and 20 birds/m². The incidence and severity of hock burns was not affected by stocking densities, however, increasing stocking density from 16 to 22 birds/m² numerically increased incidence and severity of hock burns. Ventura et al. (2010) by investigating stocking densities of 8, 13 and 18 birds/m² founded that incidence of foot pad lesions and hock burns increased by increasing broiler density.

Table 6. Effects of different stocking densities on broiler litter pH, moisture and ammonia levels at day 42

<table>
<thead>
<tr>
<th>Stocking density (birds/m²)</th>
<th>Item</th>
<th>16</th>
<th>18</th>
<th>20</th>
<th>22</th>
<th>SEM</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>21 day</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td>7.74</td>
<td>8.05</td>
<td>8.13</td>
<td>8.16</td>
<td>0.088</td>
<td>0.319</td>
<td></td>
</tr>
<tr>
<td>Moisture, %</td>
<td>42.71</td>
<td>39.41</td>
<td>40.12</td>
<td>39.63</td>
<td>1.675</td>
<td>0.902</td>
<td></td>
</tr>
<tr>
<td></td>
<td>42 day</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td>8.12</td>
<td>8.33</td>
<td>8.42</td>
<td>8.39</td>
<td>0.070</td>
<td>0.491</td>
<td></td>
</tr>
<tr>
<td>Moisture, %</td>
<td>40.97</td>
<td>37.67</td>
<td>37.05</td>
<td>40.70</td>
<td>2.302</td>
<td>0.910</td>
<td></td>
</tr>
<tr>
<td>Ammonia, ppm</td>
<td>54.05</td>
<td>62.56</td>
<td>57.59</td>
<td>61.13</td>
<td>3.069</td>
<td>0.779</td>
<td></td>
</tr>
</tbody>
</table>

The means within the same column with at least one common letter, do not have significant difference (P>0.05). SEM: standard error of the means.

Table 7. Effect of different stocking density on foot pad lesion and hock burn scores (%) at day 42

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Foot pad score¹</th>
<th>Hock burn score²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stocking density (birds/m²)</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>16</td>
<td>67.81¹</td>
<td>32.18²</td>
</tr>
<tr>
<td>18</td>
<td>72.81¹</td>
<td>27.19³</td>
</tr>
<tr>
<td>20</td>
<td>65.00³</td>
<td>35.00⁴</td>
</tr>
<tr>
<td>22</td>
<td>44.37⁶</td>
<td>52.50⁷</td>
</tr>
<tr>
<td>P-value</td>
<td>0.0041</td>
<td>0.0046</td>
</tr>
</tbody>
</table>

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

¹² Scoring: on the basis; 0: normal (no lesion or burn) to 3: severe lesion or burn.

Sorensen et al. (2000) and Dozier et al. (2005b) demonstrated that litter moisture content and foot pad lesion scores increased linearly with increasing stocking density. In the present study, although litter moisture was not affected by increasing stocking density, but litter moisture content in all of the pens was high enough to affects foot pads.

Moreover, Heckert et al. (2002) and Arnould and Faure (2003) reported that greater incidence of foot pad lesions and hock burns of broilers at higher stocking densities were related to inhibited movements of broiler chickens. Higher stocking density often makes unsuitable rearing situation by deteriorate the environment of within poultry house (Dozier et al. 2005a; Dozier et al. 2005b; Dozier et al. 2006). Bilgili et al. (2010) reported that wet and caked litter was a primary cause in greater incidence of foot pad lesions and hock burns and proper ventilation was a key factor for decreasing wet litter. Foot pad lesion and hock burn scores increased by increasing stocking density (P<0.05) which can be related to more contact of foot skin with wet litter and reduced movement by increased stocking density.

**CONCLUSION**

Broilers grown at higher stocking density (22 birds/m²) considerably had lower body weight gain, feed intake and greater footpad lesions than lower stocking densities (16 and 18 birds/m²). However, application of stocking densities higher than 20 birds/m² of floor space, regardless its negative impacts on broiler performance, increased kg
BW/m² of floor space. Application of a higher stocking density in broiler production needs more attention to environmental conditions control and adequate feeding and drinking spaces in the house. According to the results taken by this study, density of 20 birds/m² of floor space was an appropriate density for growing the broilers in an environmentally controlled broiler house.

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REFERENCES


Dozier W.A., Lott B.D. and Branton S.L. (2005a). Live performance of male broilers subjected to constant or increasing air velocities at moderate temperatures with a high dew point. Poult. Sci. 84, 1328-1331.


