

The effect of stocking density and crude protein level on performance, carcass characteristics, nitrogen use, and ammonia emissions of the Indonesian native chickens

F. R. P. Hantoro^{1,2,*}, D. Sunarti¹, T. Yudiarti¹, S. Sumarsih¹, and A. Sinurat²

¹Department of Animal Science, Faculty of Animal and Agricultural Sciences,
Universitas Diponegoro, Semarang, 50275, Indonesia

²Research Center for Animal Husbandry, Research Organization for Agriculture and Food,
National Research and Innovation Agency, Cibinong Science Center, Indonesia
Corresponding E-mail : franciscusrudiprasat@students.undip.ac.id

Received October 20, 2023; Accepted May 20, 2024

ABSTRACT

This study aimed to investigate the impact of stocking density and crude protein level on the performance, carcass characteristics, nitrogen (N) use, and ammonia emissions of Sentul Selected (Sensi)-1 Agrinak chickens. A total of 504 chickens were used in the 70-day analysis using a randomized block design with factorial pattern of 3×3 which was arranged in nine treatments and four replicates. The administered treatment covered two factors, including stocking density (A) and crude protein level (B). The treatments were administered when chickens reached 5 weeks of age. The results showed that different stocking density significantly influenced ($p < 0.05$) the average weekly feed consumption per chicken at 6, 7, 8, and 9 weeks, but not at 5, 6, and 10 weeks. However, varying crude protein levels showed no significant effect on carcass weight, giblet weight, and total edible parts. There was also no significant interaction observed between stocking density and crude protein level in relation to carcass weight and edible parts. Furthermore, no significant differences were found in giblet and carcass weights between the two groups ($p > 0.05$). The results showed that feeding Sensi-1 Agrinak chickens with a lower crude protein level of up to 14% did not compromise their performance, meat quality, protein digestibility, or ammonia emissions, but affected carcass characteristics.

Keywords: Crude protein level, Nitrogen use, Sentul Selected (Sensi)-1 Agrinak Chicken, Stoking density

INTRODUCTION

In mid-2020, the global population was around 7.8 billion, up from 7 billion in 2010, with an annual growth rate of 1.1%. By 2030, it is projected to reach 8.5 billion, increasing further to 9.7 and 10.9 billion by 2050 and 2100, respectively (United Nations, Department of Economic and Social Affairs, 2022). Scientists have estimated that this growth may lead to a

50% increase in food demand, potentially rising to 75% (Prosekov and Ivanova, 2018). The expanding population is driving a surge in demand for high-quality protein sources, particularly in the production of farm animal products. Food and Agriculture Organization (FAO) predicts a 15% global increase in meat consumption by 2031. The trend towards meat consumption, particularly chicken, is significant across high, middle, and low-income countries. The preference

for white meat, perceived as easier to prepare, healthier, and more affordable, is contributing to this shift (Food and Agriculture Organization, 2022). To meet the growing demand for chicken meat, particularly chickens, it is necessary to enhance the productivity of purebred and local chickens.

Several production systems aim to maximize chickens' weight per square meter of space, optimizing economic outcomes through high stocking density (Benante and Boateng, 2023). Native chickens play a crucial role in the meat and egg supply chain, offering advantages such as adaptability to local environments, disease resistance, superior meat quality, and distinct taste (Deng *et al.*, 2022). In Indonesia, there is currently no established stocking density standard for chickens. Therefore, investigating the effects of stocking density is essential to enhance productivity.

High stocking density negatively impacts body weight, feed consumption, and efficiency, and may also compromise health and welfare (Sugiharto, 2022). According to Ravindran *et al.*, (2006), higher stocking of chickens can lead to increased disease and mortality rates, as well as impair carcass quality, posing significant economic challenges. A similar result has been observed in France with guinea fowl broilers, where stocking density was found to affect carcass quality (Kryeziu *et al.*, 2018).

Protein is crucial in biological systems, participating in processes such as tissue synthesis, regeneration, and development (Chrystal *et al.*, 2019). Protein requirement varies among chicken species, and maintaining a balance between protein and energy in their diet is essential for optimal growth. A diet with low energy and high protein content can lead to protein deficiency and slow growth. To optimize chickens' performance, it is crucial to consider essential nutrients such as energy, protein, carbohydrates, minerals, and vitamins in the available feed. High stocking density can increase temperatures, causing stress in chickens and affecting their performance. Bilal *et al.* (2021) reported that heat stress resulting from stocking density reduced the efficient use of protein rations during production. The main reason is that a high protein intake increases heat production in the body due to the heat increment during protein digestion. Additionally, the heat generated from digesting food, including carbo-

hydrates and proteins, can overload chickens with heat and reduce metabolic activity. Indrasari *et al.* (2014) observed that an increase in protein digestibility correlated with higher nitrogen (N) retention in the body. Nitrogen retention indicates the absorption and use of nitrogen by chickens (Abun *et al.*, 2023). High retention suggests that protein needs of the livestock are met, potentially leading to increased weight gain. However, converting excreted nitrogen into ammonia (NH₃) can pose environmental and health risks to farmers and animals (Madri Brink *et al.*, 2022). Therefore, this research aims to assess the impact of stocking density and crude protein level on the performance, carcass characteristics, nitrogen use, and ammonia emissions of Indonesian native chickens.

MATERIALS AND METHODS

Research Materials

This research was conducted following the animal ethics guidelines established by the Ethical Clearance Commission of the Faculty of Animal and Agricultural Sciences, Universitas Diponegoro (No. 59-05/A-11/KEP-FPP). Chickens used were Sentul Selected (Sensi)-1 Agrinak, totaling 504 chickens, bred through pure strain formation analysis conducted by the Livestock Research Institute. Designated as Indonesian native chickens' strain by the Minister of Agriculture under Decree number 39/Kpts/PK.020/1/2017, these chickens served as grandparent stock (GPs) and parent stock (PS) for producing day-old chicks (DOC) (Hasnelly *et al.*, 2018). At 10 weeks of age, selected male Sensi-1 Agrinak chickens weighed between 1.015-1.051 g/bird, when provided with a single ration containing approximately 17% crude protein and 2850 kcal/kg. Meeting the nutritional requirements of chickens at this age covered a daily ration of 50-70 g/bird or roughly 350-490 g/bird/week, ensuring a cumulative intake in the range of 1960-2695 g/bird up to the age of 10 weeks. Meanwhile, Sensi-1 Agrinak chickens had a rapid harvest age of 70 days.

The research feed comprised rice bran, distillers dried grain with soluble (DDGS), yellow corn, cassava flour, wheat bran, meat bone meal, fish meal, minerals, DL-methionine, tryptophan, threonine, and lysine. The ingredient composition and nutrient content of the diets were shown

Table 1. Nutrient Content of Ration Ingredients in Dry Air

Feed Ingredients	**ME (kcal/kg)	*Crude Protein (%)	*Crude Fat (%)	*Crude Fiber (%)	*Ca (%)	*p (%)
Rice bran	2462.73	8.78	5.71	11.19	0.04	1.40
Distillers dried grain with soluble	2697.31	20.91	3.91	10.32	0.05	0.00
Yellow corn	3123.12	6.82	2.01	2.01	0.02	0.30
Cassava flour	2681.44	2.58	1.71	12.84	0.30	0.35
Wheat Bran	2935.66	15.14	2.87	3.61	0.14	1.10
Meat Bone Meal	2103.42	44.00	470	14.90	11.00	3.00
Fish meal	2632.71	52.95	11.67	4.46	0.50	2.60

* Results of proximate analysis.

** The results of calculations using the formula Bolton

Table 2. Composition and Nutritional Contents of Research Rations*

Feed Ingredients	Composition (%)	Composition (%)	Composition (%)
Rice bran	12.00	10.00	5.00
Distillers Dried Grain with Soluble	24.00	24.00	28.00
Yellow corn	28.00	27.50	24.00
Cassava flour	12.21	10.21	6.21
Wheat bran	9.00	10.00	15.50
Meat Bone Meal	4.00	6.00	8.00
Fish meal	6.00	7.50	8.50
Mineral	3.00	3.00	3.00
DL-Methionine	0.25	0.25	0.25
Tryptophan	0.27	0.27	0.27
Threonine	0.77	0.77	0.77
Total	100.00	100.00	100.00
Nutritional Content:			
Metabolic Energy (kcal/kg)	2632.33	2605.66	2603.97
Crude Protein (%)	14.44	16.05	18.00
Crude fat (%)	3.58	3.70	3.79
Crude Fiber (%)	7.15	7.28	7.10
Calcium (Ca) (%)	1.63	2.06	2.28
Phosphorus (P) (%)	0.83	0.95	0.96

*Calculation was based on the results of the proximate analysis.

in Tables 1 and 2. During the analysis, 36 stilt cages measuring $1 \times 1 \times 70 \text{ m}^3$ were used, with each plot accommodating 10, 14, and 18 chickens (unsexed).

Research Method

This research adopted a randomized block design with factorial pattern of 3×3 with four repetitions, using body weights as a block. The administered treatments covered two factors, including stocking density (A) and crude protein

level (B). Stocking density (A) was categorized into three levels, comprising 10 birds/m² (A1), 14 birds/m² (A2), and 18 birds/m² (A3). Similarly, crude protein level (B) featured three treatment levels, consisting of 14% CP (B1), 16% CP (B2), and 18% CP (B3), resulting in 9 treatments with 4 repeats, totaling 36 experimental units. The resulting treatments included
A1B1 = 10 birds/m² with 14% crude protein,
A1B2 = 10 birds/m² with 16% crude protein,
A1B3 = 10 birds/m² with 18% crude protein,

A2B1 = 14 birds/m² with 14% crude protein,
 A2B2 = 14 birds/m² with 16% crude protein,
 A2B3 = 14 birds/m² with 18% crude protein,
 A3B1 = 18 birds/m² with 14% crude protein,
 A3B2 = 18 birds/m² with 16% crude protein, and
 A3B3 = 18 birds/m² with 18% Crude Protein.

Research Procedure

This research spanned 70 days, with the starter phase lasting from day 1 to day 28. In this phase, chickens were fed BR-1, a commercial feed with 2986.79 kcal/kg nutritional content, including 20% crude protein, 4% crude fat, 5% crude fiber, and 0.8-1.10% calcium (Ca) and phosphorus (P). Feed treatment adaptation was carried out at the age of 21 to 28 days. By day 29, chickens were divided into 9 treatments, with 504 chickens in total, grouped based on uniform body weight. They were then fed a basal feed with varying crude protein level until reaching 10 weeks of age.

Data Collection

Data related to performance were collected weekly from chickens aged 5 to 10 weeks. In the 10th week, the characteristics of carcass from each treatment were recorded. To prepare for slaughter, selected chickens were fasted for 16 hours and individually weighed. The slaughter process included severing the neck and veins near the first cervical bone, followed by bleeding, and weighing. Subsequently, the calf and head were separated, and the stomach and intestines contents were removed, along with internal organs such as the gizzards, lungs, spleen, liver, and heart. Carcass and chickens' offal, including empty gizzard, liver, and heart, were then weighed separately. Finally, weight proportional to the live chickens' weight, carcass weight, and total edible parts were calculated (El-Shony *et al.*, 2021).

$$\text{Giblets Weight (\%)} = \frac{GW}{LW} \times 100\%$$

$$\text{Edible Parts (\%)} = \frac{EW + GW}{LW} \times 100\%$$

Where, LW = live body weight
 EW = eviscerated weight
 GW = giblets weight

Regarding meat composition, chicken breasts were sliced, bones and skin removed, samples weighed, dried in an oven at 70°C for 72 hours, mashed, and analyzed for protein and fat content (Helrich, 1990).

Protein digestibility and nitrogen retention were measured using the total collection method. Excreta was sprayed with 0.2 N HCl, weighed, homogenized, and sampled for crude protein analysis, with endogenous nitrogen measured using 6 untreated chickens as a correction group. After 2 × 24 hours with access to drinking water, excreta was cleaned, dried, and analyzed for nitrogen and protein content. The Kjeldahl method determined crude protein content (Helrich, 1990), and the digestibility was calculated using the formula by (Wahju, 2014).

(1) The formula for calculating protein consumption included the following.

Protein consumption = protein content of feed X feed dry matter

(2) Protein digestibility measurement

Protein digestibility (%) = {(Protein consumption – excreta protein) / Protein consumption} X 100%

Nitrogen retention was calculated to determine the amount of nitrogen left behind and used in the body (Sibbald and Wolynetz, 1987).

3) Nitrogen retention = N consumption – (N excreta – N endogenous)

The ammonia emissions data was collected using a method created by Marang *et al.*, (2019). In this research, a modified chamber was used to collect excreta samples weighing 20 g. The ammonia level was measured using an AR8500 intelligent sensor ammonia gas detector with 1 ppm accuracy and then converted into ppm units per gram of excreta.

Data Analysis

The obtained data were analyzed using ANOVA with a factorial group random design. When the results showed a significant difference (p<0.05) in the variables measured, Duncan's test was conducted using SPSS 20.

RESULTS AND DISCUSSION

Performance Parameters

The impact of different stocking density and

crude protein level on the performance and mortality of Sensi-1 Agrinak chickens was presented in Table 3. The analysis of stocking density showed no positive effect ($p > 0.05$) on the average live weight of chickens aged 5 and 9 weeks but did at 10 weeks ($p < 0.05$). At week 10, the average live weight of A1 differed significantly from A3 but not from A2. Although the average live weight of A1 was higher than that of A3, it did not substantially differ from A2. The analysis of crude protein level showed a positive effect ($p < 0.05$) on the average live weight of Sensi-1 Agrinak chickens at 6 weeks of age. At this age, the average live weight of B3 did not significantly differ from B2 ($p > 0.05$) but differed ($p < 0.05$) from B1. Furthermore, the average live weight of B3 and B2 was higher than that of B1, and there was no interaction between stocking density and different feed protein levels on chickens' weight.

The analysis of stocking density showed a positive effect on the average weekly feed consumption per head ($p < 0.05$) at 6, 7, 8, and 9 weeks old, but not at 5 and 10 weeks ($p > 0.05$). Although feed consumption of A3 was not significantly different from A2 at 6, 7, and 8 weeks, it differed from A1, with A3 and A4 consuming more than A1. At 9 weeks, A1 and A2 had higher feed consumption compared to A3, with no significant difference between them. The analysis of crude protein level showed a positive effect ($p < 0.05$) on average feed consumption at week 8, but not at 5, 6, 7, 9, and 10 weeks ($p > 0.05$). At week 8, the average feed consumption of B3 and B2 was higher when compared to B1. Therefore, there was an interaction between stocking density and crude protein level on average feed consumption, particularly at density of A1 to A2, where the consumption with B2, B1, and B3 proteins were increased, stabilized, and decreased, respectively.

The analysis of stocking density showed a profound effect ($p < 0.05$) on the feed conversion ratio (FCR) in chickens aged 5, 6, and 10 weeks, but did not ($p > 0.05$) at 7, 8, and 9 weeks. Although the feed conversion ratio of A3 was significantly different from A1 at 5 weeks, it did not differ from A2. At 6 and 10 weeks, the conversion ratios of A3 and A2 were not substantially different but differed from A1. The analysis further showed that the feed conversion ratio of both B3 and B2 was higher than B1. Therefore,

there was no interaction between stocking density and crude protein level on the feed conversion ratio. In the maintenance period, chickens' mortality remained below 1%, with stocking density and different protein level having no significant effect ($p > 0.05$).

This research investigated the impact of stocking density and crude protein level on the performance of Sensi-1 Agrinak chickens. Concurrently, Son *et al.* (2022) investigated the end weight of broilers (Ross 308) under maintenance condition. It was found that the final weight of chickens at density treatments of 18 and 16 birds/m² (2013.7 g and 1988.6 g) were higher than those at 21, 23, and 26 birds/m² (1945.3 g, 1844.0 g, and 1816.2 g). Harn *et al.* (2012) observed no significant difference in the live weight of broilers (Ross 308) when crude protein level was reduced by 1%, 2%, and 3%. Meanwhile, Goo *et al.* (2017) reported that different stocking density positively affected broiler feed consumption ($p < 0.05$), with lower consumption at higher density compared to lower ones. The investigation by Pankaj Chauhan *et al.* (2022) indicated that stocking density differences significantly impacted feed conversion ratio, with higher density resulting in greater conversion. Madilindi *et al.* (2018) reported, stocking density and different crude protein level did not positively influence chickens' mortality.

Carcass Characteristics

Table 4 presented the effect of stocking density and crude protein level on the characteristics of Sensi-1 Agrinak chicken carcass. The results showed that stocking density positively affected ($p < 0.05$) giblet weight, with A1 higher than A3 but not different from A2. Meanwhile, carcass weight and total edible parts were not substantially affected by stocking density. The analysis of different crude protein level positively affected ($p < 0.05$) carcass weight and total edible parts, with B3 carcass heavier than B1 but not different from B2. In the total edible parts, B3 was higher than B1 but not different from B2. It was then concluded that there was no interaction between stocking density and crude protein level in carcass weight, giblet weight, and total edible parts.

Carcass performance was crucial in broiler production economics, determined by final weight, body development, and primary carcass

Table 3. Effect of Different Stocking Density and Crude Protein Levels on Average Live Body Weight (ALBW), Average Feed Intake (AFI), Feed Conversion Ratio (FCR), and Mortality in SenSi-1 Agrinak from 5 Weeks to 10 Weeks of Age

Item	5 weeks			6 weeks			7 weeks			8 weeks			9 weeks			10 weeks			Mortality %
	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	
A1	389.13 ±40.26	42.68 ±4.93	2.46± 0.32b	501.69 ±47.93	48.98± 5.93b	3.03± 0.20b	588.24 ±62.86	49.44± 5.93b	4.08 ±0.7	685.72 ±86.75a	50.08± 9.94b	3.86 ±1.3	782.47 ±92.91	61.29 ±8.13	4.57 ±0.8	855.75± 93.16a	62.81 ±7.91	6.33± 1.47b	0.008 ±0.03
A2	385.28 ±40.86	43.20 ±4.34	0.42a b	494.33 ±59.55	51.85± 7.60a	3.41± 0.43a	578.72 ±70.01	51.85± 7.60a	4.60 ±1.4	672.19 ±87.67a	57.61± 9.23a	4.38 ±0.5	777.99 ±108.6	61.08 ±8.22	4.13 ±0.6	830.75± 108.00ab	62.36 ±8.70	8.65± 2.39a	0.012 ±0.04
A3	381.10 ±35.54	44.23 ±4.24	2.83± 0.54a	489.15 ±45.43	50.76± 5.20ab	3.37± 0.33a	567.97 ±38.94	50.76± 5.20ab	±1.6 3	±47.88 9.20	57.17± 5.90a	±0.6 6	750.47 ±54.03	±5.75 b	±0.5 9	800.95± 57.37b	62.78 ±8.36	9.67± 3.10a	0.005 ±0.17
SEM	3.28	0.64	0.09	5.37	0.95	0.09	7.52	0.87	0.36	9.20	1.69	0.25	11.49	0.87	0.21	12.88	0.91	0.57	0.01
Crude Protein (%)																			
B1	379.10 ±40.08	42.34 ±4.64	2.71± 0.44	480.79 ±51.53	49.44± 6.33	3.43± 0.48	566.90 ±55.63	49.44± 6.33	4.22 ±0.3	655.08 ±75.67	50.94± 4.96b	4.32 ±1.2	754.24 ±85.28	60.50 ±8.60	±0.6 4	812.36± 92.99	61.50 ±8.45	7.93± 2.34	0.008 ±0.03
B2	387.33 ±35.96	43.91 ±4.29	2.67± 0.49	501.26 ±45.69a	50.83± 5.94	3.16± 0.22	581.87 ±56.70	50.83± 5.94	±1.2 8	677.10 ±71.12	54.71± 12.20ab	±0.9 4.39	776.79 ±83.91	59.60 ±8.60	±0.9 0	835.54± 79.79	62.19 ±7.60	8.81± 3.68	0.005 ±0.17
B3	389.07 ±40.34	43.86 ±4.68	2.57± 0.45	503.12 ±54.00a	51.30± 6.87	3.21± 0.32	586.15 ±63.72	51.30± 6.87	±1.5 7	682.41 ±81.99	59.22± 6.85a	±0.5 0	779.90 ±96.28	1±7.1 1	±0.5 9	839.54± 98.95	64.26 ±8.65	7.91± 2.08	0.012 0.03
SEM	3.28	0.64	0.09	5.37	0.95	0.09	7.52	0.87	0.36	9.02	18.35	0.25	11.49	0.87	0.21	12.88	0.91	0.57	0.01
Stoking Density * Crude Protein																			
A1	B	389.50 ±37.17	41.43 ±5.08	2.37± 0.27	496.56 ±38.75	46.25 ±3.33	579.75 ±56.87	46.44± 5.15	4.00 ±0.6	672.17 ±87.06	46.79± 1.24cd	±1.9 1	780.84 ±86.44	59.68 ±6.54	±0.8 0	844.66± 98.62	61.10 ±5.75	6.82± 0.77	0.03± 0.05
B	2	389.19 ±43.49	42.41 ±5.09	2.45± 0.38	500.56 ±54.46	48.52 ±2.02	586.33 ±76.28	48.99± 6.63	±1.2 3	678.67 ±98.07	42.92± 9.64d	±1.4 3	±104.3 1	59.91 ±9.76	±1.0 6	851.75± 92.33	60.43 ±7.63	5.33± 1.93	0.00± 0.00
B	3	388.69 ±51.51	44.19 ±6.68	2.57± 0.36	507.94 ±62.20	50.70 ±4.03	598.63 ±72.05	51.52± 6.42	±0.4 1	706.34 ±97.56	60.53± 6.26ab	±0.6 6	±111.4 2	64.27 ±9.31	±0.5 4	870.83± 114.38	±10.2 2	6.85± 1.25	0.00± 0.00
SEM		21.24	1.10	0.15	27.49	1.65	31.94	1.51	0.62	41.24	2.93	0.43	47.46	1.51	0.37	22.31	1.58	0.99	0.01

Table 3. Effect of Different Stocking Density and Crude Protein Levels on Average Live Body Weight (ALBW), Average Feed Intake (AFI), Feed Conversion Ratio (FCR), and Mortality in SenSi-1 Agrinak from 5 Weeks to 10 Weeks of Age (continued)

Item	5 weeks			6 weeks			7 weeks			8 weeks			9 weeks			10 weeks			Mort ality %				
	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR	ALBW g/bird	AFI g/bird	FCR					
A2	372.11± 52.18	42.21 ±3.50	±0.3 8	470.27 ±73.14	50.11 ±6.50	3.67± 0.65	564.92± 78.26	50.25± 7.76	±0.5 4	3.72 4.74	±0.5 4.74	649.46± 94.80	50.60± 4.29bcd	±0.7 9	4.30 4.26	746.08±1 16.79	62.73 ±8.76	±0.6 5	4.34 3.89	60.64 ±10.5	8.59± 1.70	0 0	
B1	393.21± 36.39	44.43 ±3.87	±0.3 2	512.61 ±52.30	53.95 ±5.07	3.18± 0.19	593.92± 59.57	54.00± 6.31	±0.7 6	4.74 5.35	±0.7 5.35	697.67± 75.71	62.79± 10.03a	±0.4 7	4.26 4.59	814.20±9 2.75	±10.6 4	±0.7 8	3.89 4.18	64.58 ±8.09	9.09± 3.20	±0.0 0	0
B2	390.52± 41.51	42.89 ±6.20	±0.4 5	500.13 ±60.46	52.53 ±8.21	3.37± 0.18	577.34± 78.93	51.29± 10.09	±2.2 2	3.72 4.94	±2.2 4.94	669.46± 109.10	62.79± 9.35ab	±0.4 0	4.59 4.63	773.67±1 33.85	59.09 ±6.99	±0.3 5	4.18 4.49	61.87 ±9.52	8.26± 2.71	±0.0 4	0
B3	21.24 375.71± 39.50	1.10 43.38 ±6.19	0.15 ±0.4 9	27.49 475.54 ±48.69	1.65 51.36 ±4.81	0.16 3.60± 0.06	31.94 556.04± 40.07	1.51 51.64± 6.40	0.62 ±2.1 9	0.62 ±2.1 5.14	0.62 ±2.1 5.14	41.24 643.62± 61.85	2.93 55.44± 4.50abc	0.93 ±1.0 5	0.43 ±1.0 4.57	47.46 735.79±6 3.78	1.51 59.09 ±6.99	0.37 ±0.4 4	0.37 4.49 4.25	1.58 ±10.6 9	0.99 8.37± 3.76	0.01 ±0.0 0	0
SEM	379.58± 37.11	44.91 ±4.68	±0.6 6	490.61 ±40.37	51.43 ±4.83	3.24± 0.23	565.38± 42.88	49.50± 5.03	±1.8 5	±1.8 4.70	±1.8 4.70	654.96± 44.43	58.42± 7.83ab	±0.6 3	±0.6 4.54	751.04±5 8.35	57.41 ±7.18	±0.6 6	±0.6 4.43	787.33± 56.33	12.02 ±2.26	±0.0 3	0
A3	388.00± 39.84	44.42 ±2.85	±0.5 9	501.29 ±55.87	51.53 ±4.29	3.26± 0.48	582.50± 40.44	51.12± 5.45	±1.6 3	±1.6 3	±1.6 3	671.46± 45.64	57.67± 6.29abc	±0.2 7	±0.2 7	764.59±5 1.74	58.13 ±5.75	±0.7 8	±0.7 8	819.34± 56.50	8.62± 2.12	±0.0 0	0
SEM	21.24 379.58± 37.11	1.10 44.91 ±4.68	0.15 ±0.6 6	27.49 490.61 ±40.37	1.65 51.43 ±4.83	0.16 3.24± 0.23	31.94 565.38± 42.88	1.51 49.50± 5.03	0.62 ±1.8 5	0.62 ±1.8 4.70	0.62 ±1.8 4.70	41.24 654.96± 44.43	2.93 58.42± 7.83ab	0.93 ±0.6 3	0.43 ±0.6 4.54	47.46 751.04±5 8.35	1.51 57.41 ±7.18	0.37 ±0.6 6	0.37 4.49 4.25	1.58 ±10.6 9	0.99 8.37± 3.76	0.01 ±0.0 0	0
P-value	NS	NS	**	NS	*	**	NS	NS	*	NS	**	NS	**	NS	NS	*	NS	*	NS	*	NS	**	NS
Stocking Density	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Crude Protein (%)	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Stocking Density x Crude Protein	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

A1, stoking density 10 birds/m²; A2, stoking density 14 birds/m²; A3, stoking density 18 birds/m²; B1, crude protein 14%; B2, crude protein 16%; A3, crude protein 18%; SEM, Standard error of means; LBW, live body weight; FI, feed intake; FCR, feed conversion ratio; **= highly significant (P<0.01); * = significant (P<0.05); ns = non-significant (P>0.05)

Table 4. Effect of Differences in Stocking Density and Crude Protein Levels on Average Carcass Characteristics of Sensi 1 Agrinak Chicken

Item	Carcass Traits						
	CW		GW		EP		
	g/bird	%	g/bird	%	g/bird	%	
Stoking Density							
A1	608.25±64.87	64.07	114.83±9.73 ^a	12.11	723.08±70.58	76.17	
A2	566.08±43.14	64.73	105.75±14.94 ^{ab}	12.24	671.83±121.86	76.99	
A3	631.25±109.43	65.04	100.25±8.18 ^b	12.68	683.58±58.28	76.71	
SEM	16.01		3.25		17.27		
Crude Protein (%)							
B1	533.00±97.98 ^b	62.89	106.67±16.34	12.23	659.67±109.41 ^b	75.12	
B2	587.25±52.14 ^{ab}	64.77	105.67±8.56	11.63	692.91±55.53 ^{ab}	76.40	
B3	613.08±77.66 ^a	66.18	112.25±12.22	12.17	725.92±85.74 ^a	78.35	
SEM	16.01		3.25		17.27		
Stoking Density * Crude Protein							
A1	B1	598.25±47.79	63.82	116.50±12.66	12.41	714.75±59.70	77.86
		595.25±43.14	63.16	108.00±8.85	11.45	704.00±45.78	74.85
		631.25±101.10	65.22	114.83±9.73	12.48	750.50±104.99	77.11
SEM		27.73		5.63		19.94	
A2	B1	505.00±144.45	61.70	97.00±20.46	12.12	602.00±163.08	75.79
		595.75±83.73	66.25	108.00±8.04	11.10	703.63±86.61	75.57
		597.50±92.55	66.28	112.25±13.04	12.52	709.75±102.80	78.94
SEM		27.73		5.63		19.94	
A3	B1	555.75±78.96	63.17	100.25±16.34	12.17	662.25±72.39	76.08
		570.75±23.51	64.89	105.67±8.56	11.37	671.00±27.39	76.41
		612.25±50.61	67.05	112.25±12.22	11.51	671.00±27.31	77.45
SEM		27.73		5.63		19.94	
P-value							
Stoking Density		NS		*		NS	
Crude Protein (%)		*		NS		*	
Stoking Density x Crude Protein		NS		NS		NS	

A1, stoking density 10 birds/m²; A2, stoking density 14 birds/m²; A3, stoking density 18 birds/m²; B1, crude protein 14%; B2, crude protein 16%; A3, crude protein 18%; SEM, Standard error of means; CW, carcass weight; GW, giblets weight; EP, edible part; **= highly significant (P<0.01); * = significant (P<0.05); ns = non-significant (P>0.05)

yield. The analysis showed no effect of different stocking density on carcass weight, consistent with Raffaella Castro Lima *et al.* (2018) which observed no differences ($p > 0.05$) in carcass results between 11 m² and 13 m² densities. Stocking density had a significant effect ($p < 0.05$) on giblet weight, contrasting El-Shony *et al.* (2021), who found the effect of stocking density on offal weight. It was evident that higher stocking density resulted in heavier weight for both carcass and edible parts. Additionally, different crude protein levels positively affected

carcass weight and edible parts, with carcass weight 19% higher when chickens were fed higher contents of protein. The results were in line with the observation of Songsee *et al.* (2020), who showed the significant effect of crude protein level on carcass weight.

Meat Composition

Table 5 presented the effect of stocking density and crude protein level differences on meat composition. The analysis showed that different stocking density and protein level had no signifi-

Table 5. The Effect of Differences is Stocking Density and Crude Protein Levels on The Meat Composition of Sensi 1 Agrinak Chicken

Item	Meat composition			
	Moisture	Crude Protein	Fat	
	%	%	%	
Stoking Density				
A1	71.30±1.67	21.10±3.14	1.84±0.46	
A2	72.08±1.48	24.23±1.88	1.32±1.01	
A3	72.15±1.41	21.17±4.58	1.43±0.75	
SEM	0.60	1.62	0.33	
Crude Protein (%)				
B1	72.23±1.83	22.45±3.08	1.80±1.11	
B2	72.20±1.04	20.53±3.40	1.31±0.22	
B3	71.10±1.43	23.53±3.93	1.48±0.74	
SEM	0.60	1.62	0.33	
Stoking Density * Crude Protein				
A1	B1	72.20±1.98	20.61±4.12	1.87±0.39
	B2	72.09±0.45	20.12±5.03	1.49±0.25
	B3	69.61±1.07	22.57±0.37	2.16±0.64
SEM	1.02	2.61	0.63	
A2	B1	73.23±2.12	23.34±0.23	1.91±1.83
	B2	71.42±1.37	23.06±2.14	1.15±0.13
	B3	71.57±0.74	26.29±0.38	0.88±0.70
SEM	1.02	2.61	0.63	
A3	B1	71.26±2.11	23.39±4.49	1.62±1.57
	B2	73.08±0.75	18.40±2.38	1.28±0.21
	B3	72.11±1.26	21.72±7.31	1.39±0.38
SEM	1.02	2.61	0.63	
P-value				
Stoking Density	NS	NS	NS	
Crude Protein (%)	NS	NS	NS	
Stoking Density x Crude Protein	NS	NS	NS	

A1, stoking density 10 birds/m²; A2, stoking density 14 birds/m²; A3, stoking density 18 birds/m²; B1, crude protein 14%; B2, crude protein 16%; A3, crude protein 18%; SEM, Standard error of means; **= highly significant (P<0.01); * = significant (P<0.05); ns = non-significant (P>0.05)

cant effect ($p>0.05$) on moisture and fat in Sensi-1 Agrinak chickens' meat. Therefore, there was no interaction between stocking density and protein level in meat composition.

The composition of animal carcass varied depending on genetic factors, age, sex, nutrition, and environment. Similarly, the composition of different species varied significantly in terms of carcass weight, fat percentage, muscle mass, and bone mass. The age at which the animal was slaughtered also affected crude protein level of chicken meat. In conventional heavy strains of

chickens, fillet protein content increased from 23.5% to 24.9% between 35 and 63 days (Baéza *et al.*, 2022). This research found no significant effect ($p>0.05$) of different stocking density and crude protein level on water content, protein, and fat of Sensi-1 Agrinak chickens' meat. In contrast, Milanino chickens' proximate analysis showed that stocking density affected crude protein and meat moisture content ($p<0.05$), with lower protein level at low density (Cerolini *et al.*, 2019). The analysis on Arbor acre chickens suggested that stocking density significantly influ-

Table 6. Effect of Differences in Stocking Density and Crude Protein Levels on Consumption Protein, Crude Protein Digestibility, Nitrogen Retention, Ammonia Emission of Sensi 1 Agrinak Chicken

Item	Consumption	Crude protein	Nitrogen	Ammonia	
	Protein	digestibility	retention	Emission	
	g/bird/day	%	g	rpm	
Stoking Density					
A1	11.23±2.14 ^b	79.20±2.61 ^a	1.77±0.41 ^b	4.14±2.52 ^b	
A2	11.88±2.58 ^b	75.76±4.08 ^b	1.76±0.51 ^b	5.18±2.90 ^b	
A3	14.79±2.46 ^a	79.36±4.49 ^a	2.22±0.48 ^a	8.98±7.02 ^a	
SEM	0.59	1,04	0.14	1.20	
Crude Protein (%)					
B1	10.96±2.14 ^b	73.99±5.79	1.69±0.42	4.88±4.15	
B2	13.03±2.71 ^a	75.89±1.33	1.89±0.44	7.00±7.24	
B3	13.91±2.81 ^a	77.38±4.24	2.17±0.57	6.41±2.44	
SEM	0.59	1,04	0.42	1.20	
Stoking Density * Crude Protein					
A1	B1	10.04±2.17	79.16±4.18	1.62±0.33	4.68±3.87
	B2	12.19±1.78	80.17±1.87	1.86±0.44	3.15±1.77
	B3	11.22±2.41	78.28±1.27	1.84±0.51	4.60±1.79
SEM		1.03	1.80	0,24	2.08
A2	B1	10.61±1.98	73.99±5.79	1.59±0.46	2.65±1.16
	B2	11.08±2.02	75.89±1.33	1.58±0.08	4.16±0.45
	B3	13.95±2.77	77.38±4.24	2.13±0.68	8.70±1.69 ^b
SEM		1.03	1.80	0,24	2.08
A3	B1	12.22±2.17	79.97±6.34	1.89±0.48	7.30±5.69
	B2	15.82±1.84	76.30±1.78	2.23±0.48	13.68±9.94
	B3	16.34±0.82	81.79±2.66	2.54±0.37	5.97±2.05
SEM		1.03	1.80	0,24	2.08
P-value					
Stoking Density		**	*	*	*
Crude Protein (%)		**	NS	NS	NS
Stoking Density x Crude Protein		NS	NS	NS	NS

A1, stoking density 10 birds/m²; A2, stoking density 14 birds/m²; A3, stoking density 18 birds/m²; B1, crude protein 14%; B2, crude protein 16%; A3, crude protein 18%; SEM, Standard error of means; **= highly significant (P<0.01); * = significant (P<0.05); ns = non-significant (P>0.05)

enced meat crude protein, but water and fat content remained unchanged (Adelegume *et al.*, 2020). Usturoi *et al.* (2023) found that different crude protein levels had no significant effect on water, protein, and fat contents.

Consumption Protein, Crude Protein Digestibility, Nitrogen Retention, Ammonia Emission

The effect analysis of differences in stocking density and crude protein level on protein consumption, protein digestibility, nitrogen re-

tention, and ammonia emissions were presented in Table 6. Different stocking density positively affected (p<0.05) protein consumption, protein digestibility, nitrogen retention, and ammonia emissions. Additionally, A3 significantly differed (p<0.05) from A2 and A1, with A3 showing higher consumption. The analysis of protein digestibility showed no difference between A3 and A1 but a significant difference (p <0.05) with A2. Regarding nitrogen retention, there was a substantial difference (p<0.05) between A3 and A2 but not with A1. Moreover, ammonia emis-

sions were significantly higher in A3 compared to A2 and A1.

Different crude protein level had a significant effect ($p < 0.05$) on protein consumption but did not affect crude protein digestibility, nitrogen retention, and ammonia emissions. The analysis showed that B3 and B2 had the same protein consumption, which was higher than B1. Therefore, there was no interaction between different stocking density and crude protein levels on protein consumption, protein digestibility, nitrogen retention, and ammonia emissions.

The results showed that different stocking density positively influenced protein consumption, protein digestibility, nitrogen retention, and ammonia emissions. Silas *et al.* (2014), on the other hand, reported that the difference in stocking density (0.25 m²/bird, 0.17 m²/bird, and 0.13 m²/bird) did not affect protein digestibility ($p > 0.05$). Mendes *et al.* (2010) stated, stocking density significantly impacted ammonia emissions, with chickens at density of 3.10 m²/bird (high density) emitting more ammonia due to increased excreta production. The analysis found that varying crude protein levels affected nitrogen consumption but not protein digestibility, nitrogen retention, and ammonia emissions. Based on the observation of Roberts *et al.* (2007), a 1% decrease in crude protein concentration substantially affected nitrogen consumption, while nitrogen retention remained unaffected. Lambert *et al.* (2022) reported that differences in protein level affected both digestibility and nitrogen retention. Reducing crude protein level in broilers could gradually decrease nitrogen consumption, with nitrogen retention resembling that of diets containing crude protein. A reduction of 1% point in protein resulted in a 10-11% decrease in nitrogen excretion, while a 2%-point reduction led to a 26% decrease. The response to crude protein reduction mirrored that of nitrogen measured in the litter. Ullrich *et al.* (2018) found that a 2-3% decrease in protein level significantly reduced nitrogen, thereby improving the environmental impact of animal production.

CONCLUSION

In conclusion, high stocking density negatively impacted the performance, protein digesti-

bility, and ammonia emissions of chickens, but did not affect carcass characteristics or meat quality. Although feeding Sensi-1 Agrinak chickens with a lower protein level of up to 14% did not compromise their performance, meat quality, protein digestibility, or ammonia emissions, it affected carcass characteristics.

ACKNOWLEDGMENTS

The author would like to thank the Agricultural and Food Research Organization for funding this research project and the National Innovation Research Agency for granting a Degree by Research scholarship for doctoral studies at Universitas Diponegoro.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in the manuscript.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in the manuscript.

REFERENCES

- Abun, A., R.R.O. Putra, and K. Haetami. 2023. Metabolizable energy and nitrogen retention of local chickens fed rations containing fermented catfish waste. *GSC biol. pharm. Sci.* 25(03):123-148.
- Ademulegun, T.I., G.O. Adeyemo, and J.A. Olupona. 2020. Carcass traits of broiler chickens as influenced by stocking density, calorie: protein ratio and season. *Niger. J. animal sci.* 22(2):155-162.
- Baéza, E., L. Guillier, and M. Petracci. 2022. Review: Production factors affecting poultry carcass and meat quality attributes. *Animal*, 16:100331.
- Benante, V., and E.A. Boateng. 2023. Effects of stocking density and environmental condi-

- tions on performance, immunity, carcass characteristics, blood constituents, and economical parameters of Cobb 500 strain broiler chickens. *Am. Lit.* 19(1):524-535.
- Bilal, R. M., F. Hassan, M.R. Farag, T.A. Nasir, M. Ragni, H.A.M Mahgoub, and M. Alagawany. 2021. Thermal stress and high stocking densities in poultry farms: Potential effects and mitigation strategies. *J. Therm. Biol.* 99:102944.
- Chrystal, P.V., A.F. Moss, A. Khoddami, V.D. Naranjo, P.H. Selle, and S.Y. Liu. 2019. Effects of reduced crude protein levels, dietary electrolyte balance, and energy density on the performance of broiler chickens offered maize-based diets with evaluations of starch, protein, and amino acid metabolism. *Poult. Sci.* 99(3):1421-1431.
- Deng, S.T. Xing, C. Li, X. Xu, and G. Zhou. 2022. The effect of breed and age on the growth performance, carcass traits and metabolic profile in breast muscle of Chinese indigenous chickens. *Foods*, 11(3):483.
- El-Shony, S.A., G.M. El-Gendi, H.R. Samak, and O.H. El-Garhy. 2021. Effect of Stocking density, dietary vitamin D3 and probiotic supplementation on carcass traits and blood parameters of broiler chickens. 5th International Conference on Biotechnology Applications in Agriculture (ICBAA), April, 51–60.
- Food and Agriculture Organization. 2022. OECD-FAO Agricultural Outlook 2022-2031. Food and Agriculture Organization. Paris
- Goo, D., J.H. Kim, H.S. Choi, G.H. Park, G.P. Han, and D.Y. Kil. 2017. Effect of stocking density and sex on growth performance, meat quality, and intestinal barrier function in broiler chickens. *Poult. Sci.* 98(3):1153-1160.
- Harn, J.V., M.A. Dijkslag, and M.M. Krimpen. 2012. Effect of low protein diets supplemented with free amino acids on growth performance, slaughter yield, litter quality, and footpad lesions of male broilers. *Poult. Sci.* 98(10):4868-4877.
- Hasnelly, S. Iskandar, T. Sartika. 2018. Qualitative and quantitative characteristics of SenSi-1 Agrinak chicken. *Indonesian J. Anim. Vet. Sci.* 22(2): 68.
- Helrich, H. 1990. Official methods of analysis of the association of official analytical chemists. Association of Official Analytical Chemists Inc. Virginia.
- Indrasari, F.N., V.D. Yudianto. and I. Mangisah. 2014. Evaluation of crude protein digestibility and nitrogen retention of broiler with different levels of crude protein diet and acetic acid. *Anim. Agric. J.* 3(3):401-408.
- Kryeziu, A.J., M. Kamberi, S. Muji, N. Mestani, and S. Berisha. 2018. Carcass traits of broilers as affected by different stocking density and sex. *Bulgarian J. Agric. Sci.* 24(6):1097-1103.
- Lambert, W., T.C. Demersay, R. Bouvet, J.L.C. Grandmaison, and S. Fontaine. 2022. Reducing dietary crude protein in broiler diets does not compromise performance and reduces environmental impacts, independently from the amino acid density of the diet. *J. Appl. Poult. Res.* 31(4):100300.
- Madilindi, M.A., A. Mokobane, P.B. Letwaba, T.S. Tshilate, C.B. Banga, E. Bhebhe, and K. Benyi. 2018. Effects of sex and stocking density on the performance of broiler chickens in a sub-tropical environment. *S. Afr. J. Anim. Sci.* 48(3):459-468.
- Madri, B., P.J.J. Geert, D. Peter, B. Özer, D. Evelynne. 2022. Reduction of dietary crude protein and feed form: Impact on broiler litter quality, ammonia concentrations, excreta composition, performance, welfare, and meat quality. *Anim. Nutr.* 9:291-303.
- Marang, E.A.F., L.D. Mahfudz, T.A. Sarjana, and S. Setyaningrum. 2019. Quality and levels of ammonia litter effects of addition synbiotic in broiler diets. *Indonesian J. Anim. Sci.* 21(3):303-310.
- Mendes, L.B., H. Xin, and H. Li. 2010. Ammonia Emissions of Pullets and effects of stocking density. Iowa State University Animal Industry Report, 7(1).
- Pankaj, C., D.S. Kunwar, S.P. Partha, K. Subodh, K.S. Ashish, K.T. Krishna, N. Durgesh. 2022. Impacts of stocking density on broiler chicken performance stress reaction and mortality in broiler chicken. *Indian J. Vet. Sci. Biotechnol.* 18(4):37-39.
- Prosekov, A.Y., and S.A. Ivanova. 2018. Geoforum food security: The challenge of the present. *Geoforum*, 91:7-77.
- Raffaella, C.L., R.F. Ednardo, M.G. Herbenson,

- E.B.C. Carlos, and R.F. Danilo. 2018. Performance of broiler chickens reared at two stocking densities and coir litter with different height. *Rev. Cienc. Agron.* 49(3):519-528.
- Ravindran, V., D.V. Thomas, D.G. Thomas, and P.C.H. Morel. 2006. Performance and welfare of broilers as affected by stocking density and zinc bacitracin supplementation. *Anim. Sci. J.* 77(1):110-116.
- Roberts, S.A., H. Xin, B.J. Kerr, J.R. Russell, and K. Bregendahl. 2007. Effects of dietary fiber and reduced crude protein on nitrogen balance and egg production in laying hens. *Poult. Sci.* 86(8):1716-1725.
- Sibbald, I.R., and M.S. Wolynetz, 1987. Effects of dietary fat level and lysine: energy ratio on energy utilization and tissue synthesis by broiler chicks. *Poult. Sci.* 66(11):1788-1797.
- Silas, A.F.A., A.O. Ayorinde, E. Daisy, S.O. Mark, O. Oluwatoyin, and E.G. Nwakaegho. 2014. Research article effect of stocking density and quantitative feed restriction on growth. *J. Anim. Health. Prod.* 2 (4):60-64.
- Son, J., H. Kim, E. Hong, and H. Kang. 2022. Effects of stocking density on growth performance, antioxidant status, and meat quality of finisher broiler chickens under high temperature. *Antioxidants* 11(871):1-12.
- Songsee, O., S. Tangtaweewipat, B.C. Isarakul, and T. Moonmanee. 2020. Proper dietary crude protein and metabolizable energy levels on growth performance, carcass characteristics and meat quality of Royal Project Bresse capon. *Agric. Nat. Resour.* 54:121-129.
- Sugiharto, S. 2022. Dietary strategies to alleviate high-stocking-density-induced stress in broiler chickens – a comprehensive review. *Arch. Anim. Breed.* 65(1):21-36.
- Ullrich, C., M. Langeheine, R. Brehm, V. Taube, D. Siebert, and C. Visscher. 2018. Influence of reduced protein content in complete diets with a consistent arginine – lysine ratio on performance and nitrogen excretion in broilers. *Sustainability*, 10:1-13.
- United Nations, Department of Economic and Social Affairs, Population Division. 2022. *World Population Prospects 2022: Summary of Results.* United Nations Publication. New York.
- Usturoi, M.G., R.M. Radu-Rusu, A. Usturoi, C. Simeanu, M.G. Doliş, R.N. Raţu, and D. Simeanu. 2023. Impact of different levels of crude protein on production performance and meat quality in broiler selected for slow growth. *Agriculture*, 13(2):1-16.
- Wahju, J. 2014. *Poultry Nutrition Science.* Gadjah Mada University Press. Yogyakarta.