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Original Article

■Author(s)

Shaheen MS ^I	ip https://orcid.org/0000-0002-9314-4075
Mehmood S ¹	(ip) https://orcid.org/0000-0001-8229-7343
Mahmud A ⁱ	(D) https://orcid.org/0000-0002-2106-4113
Riaz A ^{II}	(ip) https://orcid.org/0000-0002-0963-7118
Mehmood A ^{III}	(iD) https://orcid.org/0000-0003-0935-1299
Ahmad S ¹	(iD) https://orcid.org/0000-0002-0390-3083

- Department of Poultry Production, Faculty of Animal Production and Technology, University of Veterinary and Animal Sciences, Lahore, 54000, Pakistan.
- Department of Theriogenology, Faculty of Veterinary Science, University of Veterinary and Animal Sciences, Lahore, 54000, Pakistan.
- Veterinary Research Institute, Lahore, 54000, Pakistan.

■Mail Address

Corresponding author e-mail address Muhammad Shabir Shaheen Department of Poultry Production, Faculty of Animal Production and Technology, University of Veterinary and Animal Sciences, Lahore-Pakistan. Phone: +92-300-0999907 Email: shabir.shaheen@uvas.edu.pk

■Keywords

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ABSTRACT

Objectives of the study were to investigate influence of artificial insemination (AI) in caged and floored flock in comparison to natural mating (NM) on broiler breeders' welfare, productive performance and economic appraisal. To execute this experiment, a total of 1440 pullets of Ross-308 Parent Stock along with 168 males were picked from a commercial flock and divided into three groups i.e., AIC=AI in cages, AIF=AI on floor and third was NM=NM on floor. Each group carried 480 females (HH) while 168 males were divided into 41, 48 and 58 for AIC, AIF and NM, respectively. During both phases all procedures and studied parameters were the same except the sperm dose rates which were changed during post peak. According to the results, significantly $(p \le 0.05)$ higher levels of serum corticosterone, glucose and cholesterol were recorded in birds of AIC and respectively as compared to birds being allowed to mate naturally. Whereas, the experimental males and females of AIF and AIC kept for AI exhibited ($p \le 0.05$) higher body weight, depletion % and feed consumption particularly in post peak phase. However, among the birds subjected to AI, the birds kept in the cages had better performance ($p \le 0.05$) than the birds kept in the floor. Moreover, frequency rate of insemination in females and milking of males found ($p \le 0.05$) reciprocal to the depletion, feed intake and body weight during peak but particularly in post peak. Perhaps, various sperm doses remained inert to implicit any impact on studied parameters. Similarly, egg weight was neither affected by housing systems nor by mating strategies during both phases. However, various insemination and milking frequencies noticeably swaved the productive traits under this study. AI in floored flock was found ineffective even in comparison to natural mating. In conclusion, AI in cages brought forth the better productive performance and lesser male depletion, hence, can be recommended.

INTRODUCTION

Housing systems and mating strategies have become a global focus of concern as these are being referred one of the most cogent factors in overall performance of broiler parent stock (PS) in progressive poultry (Whitehead *et al.*, 2016). Hence, now a days, the scientists are striving to probe the exact impact of keeping place of birds on their productive and reproductive performance. Despite of many advantages and disadvantages, two housing systems i.e., floor and cage are being attached with commercial laying birds (Layers and breeders) (Valkonen *et al.*, 2008).

Even though, deep litter floor is more common and a cheaper housing system than cages (Aviagen, 2016), yet its' extraordinary wetness or dryness can make it inappropriate bedding material which may negatively disrupt the welfare and performance of a PS flock (De



Jong et al., 2014; Petek et al., 2014). However, floor rearing system to an extent satisfies the natural behavior of the bird (Ericsson et al., 2016) with lesser capital cost in consort with natural mating (NM) and artificial insemination (AI) as compared to cages. However, in addition to higher ratio of dirty eggs (De Reu et al., 2009), higher need of feed consumption owing to liberty of movement to some extent along with vices can be linked with deep litter floor housing system. Contrarily to floor, cages not only maintain quality of eggs (Gianenas et al., 2009), but also prevents the nutrient wastage by restricting movement of birds thus better production with less feed consumption has been noticed (Hetland et al., 2004). In cages, the birds are bolted from several vices especially cannibalism in males which is probably the main cause of mortality during production. Resultantly, better livability has been recorded in cages as compared to floor where the aggressive males fight to kill the recessive ones. However, installation of cage system escalates the capital cost (Valkonen et al., 2008) as well as the welfare requirements of birds are also being compromised by two folds (Matthews and Sumner, 2015; Campbell et al., 2019) i.e., one by restricting movement and secondly by adopting AI which is almost compulsory in such housing systems. Indeed, any kind of management of mental stress experienced during life can exert short and long-term negative impressions on birds (Janczak and Riber 2015). Conclusively, nutrition and behavioral expression along with the ability of the birds to adapt to environmental stress are being determined by the production systems which influence bird welfare and productive performance. That's why conventional battery cage system (caged) has been criticized and banned in some countries (e.g., European), because it prevents birds from exhibiting their natural behavior (Mugnai et al., 2011).

Reproduction technique is another commanding factor affecting the poultry production (Koohpar *et al.*, 2010). Although, males (hereafter can be termed as males) of commercial strains used in Pakistan like Ross (308), Cobb (500), Arbor Acres and Hubbard classic are genetically competent to maintain fertility across the production cycle (25 to 64 weeks) through natural mating yet a sharp decline in fertility has been observed after peak phase of production (29 to 45 weeks), particularly after 50 weeks of age. Actually, males' management at PS farm, it looks phenomenal target to control body weight with acceptable uniformity (above 80%@±7.5%) particularly it turns out to be a herculean

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task after 50 weeks (post peak) in floored flocks. While, failure in controlling the body weight can be blemished as the defect cause of decline in fertility in post peak phase which is being cater with AI in which males are used more efficiently (Villaverde-morcillo et al., 2015) with ease (Kharayat et al., 2016). However, ultimate results of AI depends upon appropriate sperms dose rate at regular intervals (Douard et al., 2003; Mohan et al., 2018). On the other hand, handling stress along with labor cost owing to AI should be reduced by improving insemination and milking intervals (Froman et al., 2011). Albeit, AI leads to better reproductive traits yet it may disrupt the productive performance and welfare aspects which can cause changes in blood biochemistry (Chloupek et al., 2011). So far, single AI frequency along with one sperm dose has been attempted in most studies conducted earlier. Therefore, this study can be the part of this endeavor to smidgen some appropriate insemination and milking frequencies with required sperm according to age of a PS flock. As combinations of different AI frequencies along with various sperm doses need to be tried to get some efficient and bird friendly AI protocols in caged and floored flock. Similarly, the utilization of semen can be used even more efficiently by précising the quantity of sperms/insemination according to requirement of hen with progression of age. Keeping this in view, the present study has been planned with the objective to investigate the effect of different AI frequencies and semen dose rates on productive and reproductive traits of broiler breeders during peak and post- peak phases in cage and floor production systems.

MATERIALS AND METHODS

The present study was a part of a PhD research work which was executed at a commercial broiler breeder farm of "Bird Inn Poultries (Pvt. Ltd)" (N = 30.912, E = 73.354) with collaboration of Pakistan Poultry Association (PPA, North Zone) and University of Veterinary and Animal Sciences Lahore (UVAS), Pakistan. The experiment was performed in two phases (Peak = 29 to 45 weeks and post peak = 45-62 weeks) to investigate the effect of various interventions being strived in AI in caged as well as in floored flock in comparison to natural mating (NM) on bird welfare, productive traits and economic appraisal. All the birds were maintained under experimental animal care procedures approved by the Ethical Review Committee (vide letter No. DR/1053) of University of Veterinary and Animal Sciences, Lahore, Pakistan.



A total of one thousand, six hundred and eight (1608) Ross (308) birds (1440 pullets along with 168 males) of 18-weeks of age were randomly picked from a commercial broiler PS flock. After a week of light stimulation, the experimental pullets were divided evenly (480) into three flocks i.e., AIC=AI in caged flock, AIF= AI in floored flock and NM= natural mating in floored flock while these flocks were allotted 41, 48 and 58 males respectively and 13 males were kept as spare stock for replacement.

To exercise AI in cages (AIC), 480 females (hereafter can be termed as females) were placed in 96 colony cages $(0.2 \times 0.4 \text{ m})$ having 5 females each (8 females /m²), while, 41 males (8.5% of females) were kept in individual cages (5males /m²). Similarly, 480 females for AI on floor (AIF), were placed in 16 pens having 30 females each (5 females /m²) with 48 males (10%) in 8 replicates having 6 males each (3 males /m²). While, for NM, same size of females flock with 58 males (12 % of females) were placed on deep litter floor. The flocks intended for AI i.e., AIC and AIF were further divided into 4 groups (480/4=120 females in each) to apply 4 AI frequencies, and these 4 groups had 120 females. After this arrangement, four different frequencies/intervals of AI i.e.,4, 6, 8 and 10 days were applied on both AIC and AIF from 29 to 62 weeks of age. Each group assigned for an insemination frequency was further fragmented into 4 sub-groups (120/4=30 females), having a total of 30 females which were kept in the 6 replicates (5×6=30 females). Finally, one of these 4 subgroups were subjected to one of the aforementioned Al frequency with 4 different semen doses (100, 125, 150 and 175×10⁶ sperms/insemination) during peak. While, this regimen of sperm doses was replaced with higher concentration i.e., 200,225,250 and 275×10⁶ during post- peak phase. Perhaps, all other procedures and parameters remained the same as practiced in peak phase. Although, semen volume/insemination was adjusted after every 6 weeks to assert the required sperm concentration/dose by using ONGO machine (working on CASSA principle) at experimental site (Bird Inn breeder farm), yet these results were further substantiated after every 6 weeks with CASSA machine present in Theriogenology Department of UVAS. Feed quantity and recipe were adjusted to get required body weight gain and egg weight provided in Ross 308 parent stock: Nutrition Specifications & Performance Objective (2016). AI was started in AIC and AIF on the 27th weeks of age while males were mixed in hens on achieving 5% of egg production. The collected semen was diluted and inseminated in

volume carrying concerned number of sperms within few minutes (5-8 minutes) with micropipette. To gauge the influence of housing systems and mating methods on bird's well-being, the blood samples (2 ml) were taken from 10 females and 2 males of each treatment promptly after AI and NM and proceeded to laboratory for serum extraction (Rubbani *et al.*, 2001) at the age of 36 weeks (peak) and at 55 weeks (post- peak) to monitor the serum level of corticosterone (CS), glucose (GLU) and cholesterol (CHOL) through ELISA kit. CS, GLU

Parameters evaluated

Productive performance

Feed intake: calculated daily feed was offered to females and males which was added for a week. Cumulative feed consumption during a phase was recorded through addition of offered feed in17 weeks of a phase and divided by HH to get feed consumed/ HH while, the feed consumed by males bestowed to HH.

Body weight: weight of individual females and males of each replicate was taken on alternative weeks across the experiment tenure. Average body weight of each phase of treatment was measured for further comparison. Uniformity of body weight was calculated at $\pm 7.5\%$ of each replicate and treatment.

Depletion: Dead female and male of every replicate was registered on daily basis. Aggregate of dead birds of 7 days was divided by balance birds and multiplied by 100 to record the weekly mortality (%). Weekly (%) mortality was added to get cumulative mortality of a phase independently.

Egg weight: Fortnightly all eggs of a treatment were weighed and their average was taken by dividing the total weight of eggs by the total number of eggs.

Serum chemistry

To gauge birds' serum chemistry, the serum corticosterone, glucose and cholesterol levels were detected through Chicken ELISA kit of Corticosterone, glucose and cholesterol, respectively (Wein *et al.*, 2016).

Economic appraisal

At the end of the experiment, chick's cost of production was analyzed by calculating just running expenditure of all three flocks (NM, AIC & AIF) from 29 to 62 weeks of age in USD (\$) along with Pakistan rupees (PKR). Total expense of HH was divided by its total produced chicks to calculate chick cost.



Statistical analysis

Effect of different mating strategies, production system, insemination and milking frequencies in broiler breeders during peak and post peak on productive performance and serum chemistry were analyzed through factorial ANOVA. GLM procedure was used in SAS software (version 9.1). Significant treatment means were compared by Duncan's New Multiple Range test considering probability at $p \le 0.05$.

RESULTS AND DISCUSSION

Serum chemistry

Apart from economy, globally the importance of bird's well-being has been recognized as a mammoth issue to be addressed in commercial poultry, although, multiple steps have been taken for the comfort of birds. Stress can be defined as any biological response elicited when a bird perceives a threat to its homeostasis which cause a negative impact on the welfare of a bird (Mugnai, 2011). The extent of stress inflicted by housing and mating methodology were monitored during both trials by evaluating the concentration of CS, GLU and CHOL levels in blood serum once in each phase. According to the results of both trials, the elevation of CS, GLU and CHOL levels were found significantly (p<0.0001) higher in caged flock (AIC), followed by floored (AIF) and the least were in birds of NM, respectively during peak (Table 1) while this pattern of said entities became more profound in postpeak. Additionally, the experimental birds subjected to AI exhibited (p < 0.0001) higher level of CS, GLU and CHOL as compared to birds which were let to mate naturally during both trials. While, during comparison of age factor, the older flock (post peak) experienced (p<0.0001) more stress than younger experimental birds (Table 1).

 Table 1 – Combined effect of housing systems and mating strategies on blood biochemistry.

		5,		, 5		,	
			Peak phase			Post peak	
Treatment		CS (ng/ml)	GLU (mol/L)	CHOL (g/L)	CS (ng/ml)	GLU (mol/L)	CHOL (q/L)
Mating	Al	0.72±0.10 ^a	13.96±2.30 ^a	2.89±0.10 ^a	1.29±0.20ª	14.46±1.16 ^a	2.97±0.20 ^a
strategies	NM	0.63±0.17 ^b	13.89±2.19 ^b	2.43±0.19 ^b	0.79±0.12 ^b	13.95±1.10 ^b	2.63±0.11 ^b
Housing system	Cage	0.86±0.09ª	14.08±2.16 ^a	2.96±0.12 ^a	1.36±0.08ª	14.58±0.11ª	3.13±0.12ª
	Floor	0.75±0.14 ^b	13.19±3.11 [♭]	2.60±0.19 ^b	0.67±0.15 ^b	13.52±0.19 ^b	2.69±0.18 ^b
				AN	OVA		
Mating Strategies		0.005	0.007	0.003	< 0.001	< 0.001	< 0.001
Housing System		< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Superscripts on means within column show significant results at p≤0.05; CS = Corticosterone, GLU = Glucose, CHOL = Cholesterol AI = Artificial insemination, NM=Natural mating

These results are indicative that cages and AI somehow would have exerted some physical and physiological stress that has been validated by the elevation of serum CS, GLU and CHOL. These findings could be explained by changes in the hypothalamus, adrenal cortices and corticosterone secretions under severe stress conditions and ultimately enhancing the process of glycogenesis. These findings are in-line with the reports of Lin et al. (2004) who found a stimulatory response for glucogenesis by corticosterone. On the other physiological phenomenon, stress conditions would have expedited lipolysis, which usually led to raise in serum triglycerides and fatty acids concentrations, thereby, up surging the level of cholesterol in serum would be logged in birds of AIC and AIF respectively. Some other studies also have observed similar changes in blood biochemistry during uncomforted scenario. Results of this experiment are also consistent with the work of Ozhan et al. (2016) who stated that serum cholesterol, glucose and uric acid levels were higher in

birds reared in cages as compared to floor. However, Johnson (2014) mentioned that the increase in level of these blood biochemistry entities depends on the duration and extent of stress stimulation. A number of new techniques in modern poultry industry have been introduced like cage system, feed restriction and AI. Due to these techniques the welfare of birds might be suppressed to measurable extent leading to changes in blood biochemistry of birds (Fraser, 2008). Therefore, it can be stated that higher levels of studied parameters are imperative that AI and cages would have impaired the welfare aspects of birds and these arguments have also been legitimated by some earlier study (Chloupek *et al.*, 2011).

Productive performance

Feed intake

Although, productive and reproductive performance of PS can be intervened by manipulating the feed quantity and recipe yet its' cost is main expenditure



to produce a chick. It has to be adjusted according to the productive and reproductive performance of a PS flock. When feed consumption of studied flocks was compared, the experimental hens of AIC consumed less feed quantity closely followed by hens of NM which was subjected to natural mating, while, the highest (p<0.0001) feed consumption was chronicled in AIF being inseminated artificially during both phases (Table 2). However, when cumulative feed consumption was compared among the flocks, AIC consumed (p<0.0001) the least quantity of feed followed by NM and AIF, respectively. While, under both trials, the hens (p < 0.0001) consumed more feed which were inseminated at intervals of 4 and 6 days, while, the least quantity of feed was consumed by experimental hens being inseminated with the intervals of 8 and 10 days in AIF and AIC, respectively. However, it was suggested that various sperm doses remained inert to influence the feed intake in this very study (Table 2). It was quite logical that AIC (females) consumed the least total feed in peak as well as in post-peak as there was restricted movement in cages, which would have induced less feed requirement to lay along with keeping up the pace of body weight gain as compared to other two flocks. These findings are largely consistent with those of Yan Li et al. (2018) and Khan & Khan (2018) who also noticed less feed consumption in caged birds. The maximum feed intake by birds of AIF might be due to more liberty of movement along with additional exercise during AI handling and forced crowding.

While among the floored flocks, NM consumed less feed as compared to AIF, this difference might be attributed to the fact that NM flock was being bolted from hectic AI procedure. It was revealed on analysis of weekly data that peak feed withdrawal was slower for AIF & AIC to maintain hen's body weight and egg mass as compared to NM. Thus, it could be avowed from the results that AI might have exerted no direct effect on requirement of feed of experimental hens rather it was magnitude of exercise, hens have to face during AI, which might have ascended the need of some extra feed to compensate the wasted energy by movement by due procedure. These annotations are in line with the findings of some other researchers like Banga-Mboko et al. (2010) who convinced that movement did matter in need of feed and caged birds produced well with less feed consumption in comparison to floored hens. The hectic procedure of AI was legitimated when feed consumption was found the highest in those hens which were inseminated frequently (4&6th days) as compared to those which were inseminated with far a partly intervals (8&10th days).

Usually feed consumed by male is bestowed to HH while calculating the economics of a PS flock. In addition to housing system, the reproductive performance of a male is strongly driven by its feed (quality and quantity) and vice versa. So, feed of males was documented for each applied treatment in the study. Where, the highest (p<0.0001) feed consumption was recorded by roosters which were

Table 2 – Effect of housing systems, mating strategies, insemination frequencies and sperm doses on feed consumption, mortality % and body weight of female.

Treatment		Feed intake (g)		Morta	Mortality%		Body weight (g)	
		Peak	Post peak	Peak	Post peak	Peak	Post peak	
HS	AIC	18193.81±0.00 ^b	18683.55±5.72 ^b	6.02±0.40	6.12±0.64 ^b	3580.33±3.93 ^b	3707.67±2.33 ^b	
	AIF	18730.45±5.83ª	19617.25±6.66ª	6.40±0.66	8.0±0.66 ª	3698.08±2.05ª	3853.42±2.05ª	
MC	AIF	18730.50±5.83	19617.25±6.66ª	6.40±0.66	8.0±0.66 ª	3698.08±2.05ª	3853.42±2.05ª	
MS	NM	18662.65±0.00	18559.80±6.00 ^b	5.98±0.52	6.78±0.52 ^b	3665.00±2.49 ^b	3722.00±4.61 ^b	
	4	18972.78±50.25ª	18472.78±50.25ª	7.20±0.91ª	8.20±0.91ª	3635.56±2.51 ^b	3754.11±5.63 ^b	
IF	6	18439.90±50.29 ^{ab}	18473.56±50.29 ^a	6.92±1.13 ^{ab}	7.72 ± 1.13^{ab}	3697.56±2.48 ^{ab}	3770.11±3.37 ^b	
IF	8	18185.27±50.29 ^b	18473.82±50.29 ^a	5.78±0.81 ^b	6.28±0.87 ^b	3719.89±2.48ª	3857.44±9.33 ^{ab}	
	10	18094.38±42.67 ^b	18385.18±42.67 ^b	6.16±0.75 ^b	7.06±0.75 ^b	3727.22±2.50 ª	3893.78±7.59ª	
	К	18559.77±48.17	18459.45±48.17	5.04±0.66	8.63±1.10	3569.21 ± 3.33	3796.54 ± 3.33	
SD	L	18465.45±48.17	18459.51±48.17	6.16±1.09	6.88±1.23	3577.96 ± 2.27	3805.29 ± 2.27	
20	Μ	18500.39±48.17	18459.43±48.17	6.33±0.80	7.20±1.14	3578.58 ± 2.38	3805.92 ± 2.38	
	Ν	18492.26±53.28	18432.33±53.28	6.66±0.29	7.51±1.30	3573.26 ± 2.96	3800.60 ± 2.96	
		ANOVA						
HS		<0.0001	0.0044	0.5010	0.0005	0.0005	0.0041	
MS		0.2754	<0.0001	0.4700	<0.0001	0.0068	0.0005	
IF		<.0001	0.0410	0.0186	<0.0001	0.0005	0.0038	
SD		0.2820	1.0000	0.3470	0.3470	0.0710	1.0000	

Superscripts on means within column show significant results at $p \le 0.05$; HS = Housing System, MS = Mating Strategies, AIF= AI in cages, AIF= AI on floor, NM=natural mating on floor, IF = Insemination Frequency, SD = Sperm Dose, K=100, L=125, M=150, N=175 (during peak phase) and K=200, L=225, M=250, N=275 (during post peak phase)



subjected to force milking for AI as compared to males of NM which were let to mate naturally (Table 3). While, among the males subjected to AI, the highest (p<0.0001) quantity of feed was consumed by AIF males as compared to males of AIC during both trials. Similarly, higher (p<0.0001) feed consumption was noticed by males which were being milked frequently. Conclusively, it is deducted from results of both trials that less feed required for natural mating might be owing to liberty to mate without any compulsion of semen production as compared to those males being pushed to produce many times more volume of semen in a week (Nahak et al., 2015). This margin of difference in offered feed among experimental males increased as milking continued till the end of trial could be explainable that older male would have required more feed (energy, Amino acids) to produce higher volume of semen per milking. Thus, peak feed intake was recorded up to 172, 165 and 145 g/day/ male of AIF, AIC and NM, respectively. These results

are in line with the study of Villaverde-morcillo et al. (2105) who also found the higher requirement of feed for males subjected to AI as compared to which are meant for natural mating. While by shortening the milking frequency, enhanced the requirement of feed is guite rational as frequent forced milking might have negatively affected the body weight, fleshing and ultimately on semen quality and quantity which would be compensated by lavishly offering feed as compared to those males which were milked after a bit longer intervals (Mohan et al., 2017). The results further indicated that although heavier males logically were not good enough for natural mating yet they were found to be able to produce better volume of semen after eating higher quantity of feed (175 & 163 g). Conversely, a special feed for males of AI need to be designed rather just giving feed in bulk as this huge ingesta in Gastrointestinal tract would be encumbering the optimum reproductive potential along with economy.

Table 3 – Effect of housing systems, mating strategies, insemination frequencies and sperm doses on feed consumption, mortality % and body weight of male.

Treatment		Feed intake (g)		Mortality %		Body weight (g)	
		Peak	Post peak	Peak	Post peak	Peak	Post peak
HS -	AIC	16776.63±16.23 ^b	19379.08±33.8 ^b	6.33±0.76 ^b	11.90±0.96 ^b	4328.47 ± 3.34	4655.44 ± 3.18 ^b
	AIF	18420.80±9.78ª	20750.73±51.17ª	7.70 ±0.44 ^a	15.51±0.13ª	4369.09 ± 2.29	4844.62 ± 2.99^{a}
MC	AIF	18420.80±9.78ª	20750.73±51.17ª	7.70 ±0.44	15.51±0.13ª	4369.09 ± 2.29	4844.62 ± 2.99^{a}
MS	NM	16065.43±11.00 ^b	18955.90±60.00 ^b	7.12±0.72	11.56±0.27 ^b	4366.85± 2.22	4704.58 ± 3.34 ^b
IF	2	18776.59±10.27ª	19935.15±30.00 ^a	7.35±0.2	17.55±1.2ª	4352.47 ± 5.53	4608.88 ± 5.53°
	3	18765.43±11.00 ^a	19555.22±37.00 ^b	7.00 ±0.20	15.81±1.15 ^b	4346.22 ± 5.61	4621.75±5.61°
	4	18520.80±19.69 ^b	18940.19±30.00 ^{bc}	6.80±0.20	11.78±0.27 ^c	4350.97 ± 5.40	4726.50± 5.40 ^b
	5	18188.63±15.23°	18870.25±35.05°	6.33±0.23	9.78±0.96 ^d	4342.08 ± 5.43	4867.61±5.43ª
		ANOVA					
HS		0.005	<0.0001	0.0021	0.0040	0.071	<0.0001
MS		<0.001	<0.0001	0.8100	0.0001	1.000	<0.0001
IF		<0.001	<0.0001	0.0669	0.0158	0.900	<0.0001

Superscripts on means within column show significant results at $p \le 0.05$; HS = Housing System, MS = Mating Strategies, AIF= AI in cages, AIF= AI on floor, NM=natural mating on floor, IF = Insemination Frequency, SD = Sperm Dose, K=100, L=125, M=150, N=175 (during peak phase) and K=200, L=225, M=250, N=275 (during post peak phase).

Depletion percentage

Health status and quality of management can be appraised by depletion % of a flock. On the other hand, housing systems and mating strategies can exaggerate the depletion. Thus, during this study, statistically less depletion was noticed in the females of NM flock followed by AIC and AIF. Similarly, it was revealed that while, there was non-significant difference in depletion % between the flocks being subjected to AI where AIC was a bit better than AIF during peak, but mortality % in AIF was significantly ahead of AIC during post peak. When the combined effect of mating strategies and housing type on depletion was compared, significantly (*p*<0.0001) the highest depletion % was found in flock of AIF followed by AIC and the least was recorded in NM flock's hens with the progression of age as found in weekly trend of mortality (Figure 1).

Vis-à-vis influence of various AI frequencies, the highest (p<0.0001) depletion was at the 4th day frequency followed by the 6 and 8th, while, the least mortality was observed among the experimental females when inseminated at the interval of the 10th day in both flocks (AIC &AIF) particularly in the phase of post- peak production (Table 2).

It could be inferred from the study that AI played some role to elevate the depletion of females that might be associated with traumatic and accidental reasons during procedure of AI. As there was continual



handling and forced crowding during insemination that would have led to more chances of mortality. But in cages the chances of crowding were abandoned that might rendered to better livability as found in AIC hens. During post peak, it was evident that weekly mortality was the highest in females of AIF as compared to AIC and NM, which could be attributed to the aging factor as older females could have been more prone to death during handling for AI. Moreover, on postmortem some lesions of injuries in reproductive tracts were noticed. These lesions might be of the glass straw used to deposit semen or of forceful vaginal douching that is vital, unavoidable, but hygienically chancy segment of AI (Hudson et al., 2017). These results are similar to the findings of Yan Li et al. (2018) who also found much higher average weekly mortality in females subjected to AI as compared to hens which were allowed to inseminate naturally. However, higher rates of leg fracture incidence could be observed when Al is being proceeded in conventional cages where a worker pulled out a female for insemination (DEFRA, 2006). Contrary to findings of this study, some workers recorded higher mortality in laying females when raised in litter-based housing as compared to cages (Michel & Huonnic, 2003; Rodenburg et al., 2008; Sherwin et al., 2010). Conclusively, the current results alluded that depletion% was significantly lower in laying females of NM than of AI and these results are largely consistent with those of Koohpar et al. (2010) who considered that AI can enhance the mortality.

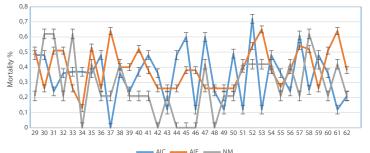


Figure 1 – Cumulative effect of housing systems and mating strategies on weekly mortality % of female during peak (29-45 weeks) and post peak (46-62 weeks) phases; AIC=AI in cages, AIF= AI on floor, NM = natural mating on floor.

Perhaps, mortality of males in a PS flock is economically more important than females as causality of a single male would spoil the fate of more than 10 hens by letting them to lay about 1700 unfertile eggs (ROSS 308: Performance objective guide, 2016). Secondly the number of males is already limited (14-52% of total flock), so the menace of paucity of males is always oscillate in mind of a manger. While, the gravity of competitiveness and aggression among males are far extensive which inclined to higher depletion in males than females in commercial PS. So, when data of mortality of experimental males were analyzed, the least (p<0.0001) depletion % was noticed in the males of AIC followed by NM and AIF, respectively even during peak phase. Although, depletion % varied nonsignificantly during peak, yet it diverged to significant (p<0.0001) in post-peak among the experimental males of AIC and AI. While the comparison of treatments during post peak revealed the highest (p < 0.0001) depletion in AIF, followed by NM and the least was recorded again in males of AIC (Table 3). Among the paraphernalia of various milking frequencies/intervals, the highest depletion (p < 0.0001) was recorded in the males which were subjected to frequent milking i.e., 2nd, 3rd day milking frequencies followed by 4th and 5th day in both flocks, respectively, particularly during post- peak phase of production as recorded in weekly pattern of mortality (Figure 2).

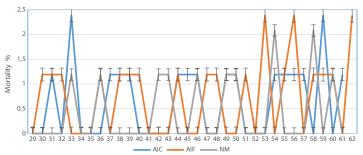


Figure 2 – Cumulative effect of housing and mating strategies on weekly mortality % of male during peak (29-45 weeks) and post peak (46-62 weeks) phases; AIC=AI in cages, AIF= AI on floor, NM = natural mating on floor.

Therefore, it is imperative from the results that higher depletion % of males of floored flocks could be attributed to certain management of mental pathological etiologies e.g., cannibalism, and traumatic fractures of leg and wings, bumble foot and staphylococcus infections. Whereas, the highest mortality rate in experimental males of AIF might be linked with traumatic injuries, crowding along with internal injuries due to abdominal massage in addition to curse of cannibalism. While the least mortality in male of AIC is logical as there was individual male in each cage, thus they remained safer from the above said reasons of mortality except forced milking which might be a cause of death as it would have led to internal injuries. These findings are in line with those of Khan & Khan (2018) who found less mortality in caged flock. According to postmortem findings and personal observations, cannibalism and accidental mortality during procedure of AI could be blemished as the foremost cause of depletion in males kept on floor. Similarly, on postmortem examination, a bit more cases of internal injuries and infection were observed



in males subjected to AI as compared to males of NM while, cage fatigue was also observed to be the cause of death in males of AIC. On the other hand, it was observed that uniformity of male body weight became poorer with progression of age in all the experimental males particularly males of AIF. In addition to stress of forced milking, males of AIF were kept in a separate pen without females, where weak males (regressed, under fleshed and health compromised male) might have turn out to be easy victims of rampant aggressive males as compared to males of NM, where they were mixed in females and as a combined flock (male + female) there would be lesser intra-gender competition for survival. These results could be legitimated by the findings of impact of milking frequencies as there were more frequent milking led to more mortality and deterioration of body fleshing was recorded which is inconsistence with some other work like (Nahak et al., 2015). Contrarily, there was no such struggling scenario as males were kept in individual cages thus the least mortality was noticed in males of AIC.

Body weight

Managing the female body weight is critical for sustainable production performance in commercial poultry. Uniform and stead weight gain is controlled by manipulating feed quantity and recipe with progression of age of a breeders flock. When female average body weights of all experimental flocks were analyzed, significantly (p < 0.0001) higher body weight was exhibited by the hens of AIF as compared to NM and AIC, respectively, under both trials (Table 2). On the other hand, higher body weight was noticed in hens which were inseminated far a partly (8th &10th days). These results are explainable, although the body weight of all experimental hens tended to make well-ordered through manipulation in the offered feed to keep average body weight according to the given standards in Ross 308 PS management Handbook (2018). However, more feed was offered to experimental hens of AIF to gain required body weight as compared to hens of NM which might have led to more weight gain as compared to other. Feed distribution couldn't have been so uniform in manual grilled feeders, which might be another contributory factor due to which AIF hens attained higher body weight than its competitors. While, the body weights of caged hens were conveniently managed.

Although hen's body weight is critical for production, yet, appropriate male body weight is necessary for sustainable fertility % which makes the production

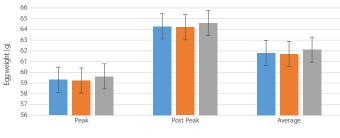
worthy in a PS flock. During this experiment, when data of male body weight were analyzed, the body weights of AIF males were although statistically higher followed by males of NM and AIC respectively during peak yet this difference became significant (p < 0.0001) in the said pattern during post- peak phases (Table 3). Moreover, milking frequency exerted noticeable impact on body weight, thus the male being milked at every 5 and 4^{th} days expressed higher (p < 0.0001) body weight than that of males being milked frequently i.e., 2 and 3rd days of AIF and the same array was observed in males of AIC. The results also indicative that males in cages possessed lesser body weight as compared to males kept on floor across the experiment. On another hand, it was also revealed that higher body weight was exhibited by males which were subjected to AI as simply they were offered more quantity of feed to compel them for more semen production as compared to males which were let to mate naturally (Silveira et al., 2014). It is deducted that more feed was required to produce more semen thus more feed was offered which resulted in heavier weight in males (Karaca et al., 2002). Contrarily, heavy males would be unfitted for mounting which is basic need in natural mating, thus less feed was offered to males of NM (Ross PS management Handbook, 2018), which kept the males smarter as compared to males of AI (AIC&AIF). Poor uniformity in body weight could be associated with AI as it progressed which further might have increased feed requirement.

These results could be explained on the basis of weekly data of feed offered to males, where the feed allowance for AI males was increased quickly (5g/male/ week) after 33 weeks until it reached to the peak feed intake i.e., 172/day and 165/day/males for AIF and AIC, respectively, and was maintained till the end of production cycle. While it was increased slowly i.e., 1-2g/male/week to the peak feed intake i.e., 145/day/ male for male being let to mate without copulation. These arguments are strengthened by the work of Schramm (2005) who stated that body weight has to be increased while uniformity of flock is deteriorated when Artificial Insemination is practiced.

Although, egg weight is critical in commercial poultry, as it determines the weight of day- old chick (DOC), yet it is regulated through nutrition (Aviagen, 2014). During both trials of experiment, feed was adjusted to get similar egg weight in all flocks. However, AIF flock yielded non-significantly heavier eggs as compared to AIC and NM, respectively, in the study.



It is deducted from the results that mating strategies along with production systems inflicted minor or no role of interfering in egg weight of broiler PS (Figure 3). These results are in line with the work of Yan Li *et al.* (2018) who could not find any difference owing to housing systems. These findings are contrary to those reported by Habibullah *et al.* (2015) and Duru *et al.* (2017) indicating that mating strategies as well as housing systems influenced egg weight. However, nutrition (energy) and genetics appear to play the key role in managing the egg weight rather management.



■ C ■ F ■ N

Figure 3 – Combined effect of housing system and mating strategies on egg weight during peak (29-45 weeks) and post peak (46-62 weeks) phases; C=AI in cages, F= AI on floor, N = natural mating on floor.

Economic appraisal

Indeed, poultry is primarily a commercial activity thus ultimately productive and reproductive outputs of a flock are being gauged through economic appraisal. Therefore, it seemed necessary to compare economic worth of natural mating and AI when being conducted in two housing systems i.e., cages (AIC) and floor (AIF). Resultantly, AI in cages proved to be the best as the greatest number of chicks/H.H were attained with the least cost production (Table 4). These findings are quite logical as there were less feed consumption and labor cost along with better production performance (Habibullah *et al.*, 2015). However, higher capital cost of cages discouraged the farmers particularly in developing countries like, Pakistan, where poultry industry suffers market turmoil now and again. While, bird welfare aspect and its' basic freedom are being impaired in cages for which the use of cages already have been abandoned in Europe (El-Deek & El-Sabrout, 2019).

Although AI on floor (AIF) yielded more chicks than natural mating (NM), yet the highest cost/chick was noticed in AIF among all the experimental flocks. It may be concluded from the calculations (Table-9) that although AI in floored flock led to more chicks than natural mating, yet, it seems to be retro-productive due to handling stress (Janczak & Riber, 2015), in addition to being expensive in terms of labor cost and feed consumption/HH.

CONCLUSIONS

It was concluded that although the consortium of cages and AI were economical as less; depletion and feed consumption per HH was chronicled as compared to its competitor birds, yet it hampered bird's wellbeing with utmost gravity. While, AI on floored flock seemed futile even in comparison to natural mating on floor. Conceivably, the highest mortality, feed consumption and body weight were recorded in the birds (male & female) being subjected to AI particularly in floored flock while the margin of difference extended further with progression of age. Insemination and milking frequencies also meddled in productive performance of experimental birds respectively. While the both mating strategies and housing systems inflicted inert impact on egg weight.

Table 4 – Cumulative effect of housing systems and mating strategies on economic appraisal.

5,7	5 5		
Particular	NM	AIF	AIC
Feed/HH (kg) (Male + Female)	42.622kg/H.H	45.86Kg/H.H	42.21kg/H.H
Feed cost/H.H; Feed@0.4\$/Kg (US)	17.049	18.344	16.884
Al equipment	0.00	0.05	0.05
Al procedure (cost/H.H)	0.00	1.70	1.02
Labor cost	0.48	0.48	0.32
Other expense	0.066	0.066	0.066
Total expense (US \$/H.H) 29-62 Weeks	17.64	20.64	18.34
Total expense (PKR) 29 to 62 Weeks	2646	3094	2751
Chicks/H.H*	125.75	128.29	131.51
Cost of production/ chick (US \$)**	0.139	0.160	0.138
Cost of production/ chick (PKRS)	21.041	24.12	20.91

*AIC produced the highest chicks/HH followed by NM and AIF, respectively; AIF= AI in cages, AIF= AI on floor, NM=natural mating

** It was just running expenditure from 29 to 62 weeks



DECLARATION OF INTEREST

It is declared by the authors that this research does not have any conflict of interest with any private or governmental organization.

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