



ISSN 2228-7701

Online Journal of Animal and Feed Research



An International Peer-Reviewed Journal which Publishes in Electronic Format

Volume 7, Issue 1, January 2017

Online Journal of Animal and Feed Research

An international peer-reviewed journal which publishes in electronic format (online)

Online J. Anim. Feed Res., 7 (1): January 25, 2017

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Nutrition - Ruminants

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PhD, Assistant Prof., Dept. Anim. Sci., I.A.U.-Shabestar, IRAN
(Website; Emails: maherisis@iaushab.ac.ir; nama1349@gmail.com)

Nutrition - Ruminants, Nutritive Value, Utilization of Feeds

Nilüfer SABUNCUOĞLU ÇOBAN

PhD, Professor, Department of Animal Science and Production, Faculty of Veterinary Medicine, Atatürk University, TURKEY
(Website; Email: ncoban@atauni.edu.tr)

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Ömer ÇOBAN

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Animal-feed interactions, Nutritive value

Saeid Chekani Azar

PhD, Dept. Anim. Sci., Facult. Vet. Med., Atatürk University, TURKEY
(Emails: saeid.azar@atauni.edu.tr; schekani@gmail.com)

Physiology, Product Quality, Human Health and Well-Being,

Shahin Eghbal-Saeid

PhD, Associate Prof., Dep. Anim. Sci., I.A.U., Khorasgan (Isfahan), IRAN (Email: shahin.eghbal@khuisf.ac.ir)

Animal Genetics and Breeding

Shahin Hassanpour

Dept. Physiology, Facult. Vet. Med., I.A.U., Shabestar, IRAN (Email: shahin.hassanpour@yahoo.com)

Physiology and Functional Biology of Systems

[Shigdaf Mekuriaw](#)

Andassa Livestock research center, ETHIOPIA ([Email: shigdafmekuriaw@yahoo.com](mailto:shigdafmekuriaw@yahoo.com))

[Animal production and Nutrition](#)

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PhD Student, Dep. Anim. Sci., I.A.U., Khorasgan (Isfahan); Tarbiat Modares University, Tehran, IRAN

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[Tohid Vahdatpour](#)

PhD, Assistant Prof., Department of Physiology, I.A.U.-Shabestar, IRAN ([Website](#); [Scopus](#); [Google](#)

[Scholar](#); [Emails: vahdatpour@iaushab.ac.ir;tvahdatpour@gmail.com](mailto:vahdatpour@iaushab.ac.ir))

[Physiology and Functional Biology of Systems](#)

[Ümit Acar](#)

Research Asistant and PhD, Department of Aquaculture, Faculty of Fisheries, Muğla Sitki Koçman University, TURKEY ([Email: umitacar@mu.edu.tr](mailto:umitacar@mu.edu.tr))

[Aquaculture, Fish nutrition, Alternative Feed ingredients](#)

[Vassilis Papatsiros](#)

PhD, Department of Porcine Medicine, University of Thessaly, Trikalon str 224, GR 43100, GREECE

([Email: vpapatsiros@yahoo.com](mailto:vpapatsiros@yahoo.com))

[Dietary input, Animal and Feed interactions](#)

[Wafaa Abd El-Ghany Abd El-Ghany](#)

PhD, Associate Prof., Poultry and Rabbit Diseases Department, Cairo University, Giza, EGYPT

([Email: wafaa.ghany@yahoo.com](mailto:wafaa.ghany@yahoo.com))

[Poultry and Rabbit Diseases](#)

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MSc, DVM, College of Veterinary, Medicine, State University of Ceará, Av. Paranjana, 1700, Fortaleza, BRAZIL

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PhD, Department of Plant Biology, Atatürk University, Erzurum, TURKEY ([Email: zohreh.yousefi12@ogr.atauni.edu.tr](mailto:zohreh.yousefi12@ogr.atauni.edu.tr))

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Volume 7 (1); 25 January 2017**Research Paper****Culling in dairy cattle farms of Khartoum, Sudan.**

Karrar MH, Osman KhM, Sulieman MS.

Online J. Anim. Feed Res., 7(1): 01-08, 2017; pii: S222877011700001-7**Abstract**

The study aimed to determine the causes and rates of voluntary and involuntary culling in dairy cattle farms in relation to some management factors in five dairy cattle farms with an average farm size of 264.8±153.1 cow/farm in Khartoum State over one year. Monthly visits were performed to each farm to collect data either by reviewing the farm records or directly from animal owners or attendants. The overall culling rate was 15.0% (71.8% voluntary and 28.2% involuntary). The most common causes of voluntary culling were economic reasons (29.1%), low milk yield (23.0%) and aging (19.7%). The common causes of involuntary culling were infertility (17.7%), chronic mastitis (8.5%) and foot injuries (2.0%). In farms where the veterinary supervision was practiced, the overall culling rate (26.4%) was higher than the rate (11.3%) in farms which did not. The highest culling rate (41.9%) where the veterinary supervision was practiced was due to aging, whereas, where the veterinary supervision was not practiced, economic reasons (38.9%) were prevailing. In farms where houses were constructed from fixed materials with adequate shade, the overall culling rate was 13.6% and almost due to low milk yield (35.5%). However, in farms where houses were constructed from local materials the overall culling rate was 17.7% with prevalent culling rate due to economic reasons (53%). In farms where feed was provided from expert companies, the most culling cases were due to economic reasons (53%) whereas when using feed which was prepared within the farm, the most cause of culling was low milk yield (35.5%). It can be concluded that the voluntary culling was the most prevalent type of culling in dairy cattle farms and animals mostly culled for economic reasons.

Key words: Voluntary and Involuntary Culling, Dairy Cattle, Khartoum, Sudan[PDF](#) [XML](#) [DOAJ](#)**Research Paper****Epidemiological investigation on outbreak of brucellosis at private dairy farms of Sindh, Pakistan.**

Yousaf A, Abbas M, Laghari RA, Hassan J, Rubab F, Jamil T, Haider I, Abbas U, BiBi N.

Online J. Anim. Feed Res., 7(1): 09-12, 2017; pii: S222877011700001-7**Abstract**

Brucellosis is one of the drastic diseases of zoonotic significance. Brucellosis is a global challenge not limited to Asia. As a result, economic losses are escalating due to the burden posed by Brucellosis in the investigated area. The present study was conducted to estimate the prevalence of brucellosis in cattle and buffaloes of Sindh, Pakistan. Blood samples were collected from (n=1200) animals (dairy cattle and buffaloes) of different age (2-7 years) and sex from 10 different districts of Sindh, Pakistan where no vaccination against brucellosis is practiced and were subjected to indirect ELISA for detection of Brucella antibodies. The overall mean prevalence was 18.16% with prevalence in unorganized is higher organized farms. In absence of any vaccination presence of circulating antibodies against Brucella in all age group of animals indicated the natural circulation of infection in the state. Based on the findings, the disease is seems to be endemic in the area, perhaps, due to partial or no vaccination. Also, the area presents poor bio-security measures and management. It is therefore, warranted to adopt good surveillance system for early identification of the brucellosis outbreak and appropriate measure for further control transmission of the brucellosis. Study indicated an urgent need of policy for prevention and control of brucellosis in dairy animals.

Keywords: Brucellosis, Prevalence, Privet dairy farms, Bio Security, Sindh[PDF](#) [XML](#) [DOAJ](#)**Research Paper****Effect of age wise incubation programme on broiler breeder hatchability and post hatch performance.**

Jabbar A., Yousaf A.

Online J. Anim. Feed Res., 7(1): 13-17, 2017; pii: S222877011700003-7**Abstract**

Temperature and humidity are most important environmental factors during incubation. The age of birds affect the eggs, its internal as well as external quality, that's why dissimilar conditions require for incubation. The aim of this study was to investigate the effect of age wise incubation profile on hatchability and chick's performance. For this experiment, eggs were collected from Ross-308 breeders which were divided into four groups according to the age of breeders having equal number of eggs in all groups (n=538560 eggs). Group A (Young, 24-31 weeks), B (Prime, 32-50 weeks), C (Old, 50+weeks) and D (control). For groups, A, B and C duration of incubation in setter machine was 456 hours (19th day) while for D (control), incubator duration was 449 hours (18.7 days). Fertility of eggs were performed through candling and shifted to hatchers for next 50 hours for A, B and C while 56 hours for D. Group B was significantly better (P< 0.05) as compare to A in term of hatchability. Candling was significantly better for group B (P< 0.05) than C. Group C was significantly (P< 0.05) better for candling than A and D which contain same candling i.e. A and D. ...[view pdf](#).

[PDF](#) [XML](#) [DOAJ](#)**Research Paper****Effect of dietary feed additives on haematological and serum biochemical parameters of broiler chickens.**

Abstract

The effect of dietary feed additives on haematological and serum biochemical parameters of broiler chickens was evaluated. 180 day-old Arbor acre broiler chicks were weighed and randomly allotted to five dietary treatments with 3 replicates of 12 birds each. Broiler starter diet (2855.7 kcal/kg ME; 23.01%) and finisher diet (2911 kcal/kg; 20.71% CP) were formulated. Dietary treatments were control diet (basal diet without additives), OXYT diet (basal diet with oxytetracycline at 600 ppm as antibiotic, GRO-UP diet (basal diet with probiotic at 500 ppm), MOS-500 diet (basal diet with mannan oligosaccharide at 500 ppm) and MOS-1000 diet (basal diet with mannan oligosaccharide at 1000 ppm). Feed and water were supplied ad libitum. At the end of weeks 4 and 8, blood samples were collected and analyzed. The haematological and serum biochemical parameters of broiler chickens fed diets containing feed additives at the starter phase were not statistically significant ($P > 0.05$). At the finisher phase, there were no significant ($P > 0.05$) differences in all the parameters measured except in the heterophils and eosinophils where birds fed the control diets had the lowest value among all treatments. Serum globulin values were significantly ($P < 0.05$) different as birds fed diets containing OXYT (antibiotics) recorded the lowest value among all treatments. The inclusion of prebiotics and probiotics in the diets of broiler chickens elicited no adverse effect on haematological and serum biochemical parameters, thus, they can be used as replacement for antibiotics.

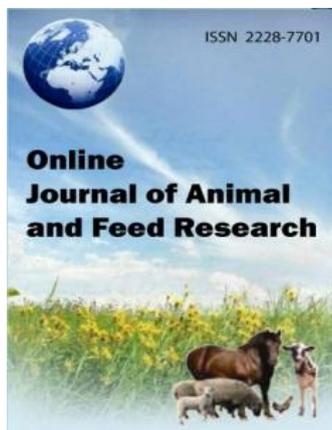
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Online Journal of Animal and Feed Research



ISSN: 2228-7701

Frequency: Bimonthly

Current Issue: 2017, Vol: 7, Issue: 1 (January)

Publisher: [SCIENCELINE](http://www.science-line.com)

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CULLING IN DAIRY CATTLE FARMS OF KHARTOUM, SUDAN

Moatasim Hassan KARRAR¹, Khadiga Mohammed OSMAN², Manal Sulum SULIEMAN³✉

¹Department of Preventive Medicine and Veterinary Public Health, Faculty of Veterinary Medicine, University of Khartoum, P.O. Box 32, Khartoum North, Sudan

²Assistant Professor, Department of Pharmacology and Toxicology, Faculty of Veterinary Medicine, University of Khartoum, P.O. Box 32, Khartoum North, Sudan

³Assistant Professor, Department of Clinical Medicine, Faculty of Veterinary Medicine, University of Khartoum, P.O. Box 32, Khartoum North, Sudan

✉E-mail: vetuk1995@yahoo.com

ABSTRACT: The study aimed to determine the causes and rates of voluntary and involuntary culling in dairy cattle farms in relation to some management factors in five dairy cattle farms with an average farm size of 264.8±153.1 cow/farm in Khartoum State over one year. Monthly visits were performed to each farm to collect data either by reviewing the farm records or directly from animal owners or attendants. The overall culling rate was 15.0% (71.8% voluntary and 28.2% involuntary). The most common causes of voluntary culling were economic reasons (29.1%), low milk yield (23.0%) and aging (19.7%). The common causes of involuntary culling were infertility (17.7%), chronic mastitis (8.5%) and foot injuries (2.0%). In farms where the veterinary supervision was practiced, the overall culling rate (26.4%) was higher than the rate (11.3%) in farms which did not. The highest culling rate (41.9%) where the veterinary supervision was practiced was due to aging, whereas, where the veterinary supervision was not practiced, economic reasons (38.9%) were prevailing. In farms where houses were constructed from fixed materials with adequate shade, the overall culling rate was 13.6% and almost due to low milk yield (35.5%). However, in farms where houses were constructed from local materials the overall culling rate was 17.7% with prevalent culling rate due to economic reasons (53%). In farms where feed was provided from expert companies, the most culling cases were due to economic reasons (53%) whereas when using feed which was prepared within the farm, the most cause of culling was low milk yield (35.5%). It can be concluded that the voluntary culling was the most prevalent type of culling in dairy cattle farms and animals mostly culled for economic reasons.

Key words: Voluntary and Involuntary Culling, Dairy Cattle, Khartoum, Sudan

ORIGINAL ARTICLE
 pii: S222877011700001-7
 Received 20 Sep. 2016
 Accepted 10 Jan. 2017

INTRODUCTION

Maintenance and optimization of a dairy herd profit and avoidance of economic losses are a continuous challenge to dairy herd farmers especially when dairy cattle are reared under stressful conditions. To achieve this goal, farmers have to imply good dairy management practice for their herd by improving the overall health indices and increasing milk yield and reproductive performance. One of these practices is culling. Culling is the removal and disposal of an individual from the herd due to sale or death. It is classified as either voluntarily, when the farmers have the choice to remove the animal for example for low milk yield or aging, or involuntarily when the farmers have no choice to remove certain individuals from the herds for example due to infertility or infectious diseases (Dohoo and Dijkhuizen, 1993; Gröhn et al., 1998). Culling is one of the important management practices to be adopted in dairy herds to maximize the profit and to minimize the economic losses. However, culling will not be effective when it is made in non-systematic and non-programmed models (Lehenbauer and Oltjen, 1998). The decision to remove a cow from the herd is based on economic considerations (Van Arendonk et al., 1988). Information about the reasons for culling in dairy cattle farms is abundant (Pinedo et al., 2010; Ahlman et al., 2011; Lari et al., 2012; Pinedo et al., 2014). Optimum herd profitability is achieved by minimizing the proportion of the herd culled for health (involuntary culling) reasons and by maximizing the proportion culled for voluntary or economic reasons (Stevenson and Lean, 1998; Lari et al., 2012). A high number of involuntary culling indicates potential health and welfare problem in a herd. The rate of the profitable culling is varying with regard to many considerations. Farmers should make strategies to minimize the rate of involuntary culling in expense of voluntary culling which, the latter, is important and is used as a positive economic tool to make a balance between inputs and outputs of a farm (Stevenson and Lean, 1998; Weigel et al., 2003). Studies suggested that profit would be increased with culling rates below the approximate average of 35% (Allaire, 1981). Other studies suggested 25-

40% as profitable culling rates (McCullough and Delorenzo, 1996). Identifying causes for culling is important and can be helpful in defining management status of a herd (Penido et al., 2014). The involuntary culling, which is often be due to diseases or poor reproductive performance, is one of the factors which negatively affect the profitability of a dairy herd particularly when it is being in a high rate. Beaudeau et al. (1993) reported that more than 50% of culling cases were due to health problem. Mastitis as a cause for culling of dairy cows has been reported by many authors (Bascom and Young, 1998; Whitaker et al., 2004; Lari et al., 2012; Penido et al., 2010). Rajala-Schultz et al. (2000) reported that a total replacement percentage was 26 with the highest frequency of voluntary culling in Finish Dairy herds. Mohammadi and Sedighi (2009) reported 13.1% (98.5% voluntary and 1.5% involuntary) as an average culling rate in 23 commercial Holstein dairy cows in Neishaboor area in Iran. Lari et al. (2012) found infertility (32.6% of all culls) was the most prevalent reason of culling followed by mastitis (6.5%). In Sudan, Elimam et al. (1999) reported overall culling rate of 11.95% in Elneshesheba dairy farm at Medani, Sudan. Saeed and Fadel Elseed (2015) reported that 79% of farmers were culling their animals on the basis of aging and decline of production. It is important to link the causes of culling with Farm management practices and individual characteristics as these factors can help explain why and when cows are culled (Bascom and Young, 1998; Whitaker et al., 2004). In Sudan, there are very few descriptive studies that have examined culling and its reasons in dairy farms (Elimam et al., 1999; Saeed and Fadel Elseed, 2015).

Therefore, limited information is available about culling and reasons for culling in dairy farms in Sudan. This available information did not critically describe the rates of voluntary and involuntary culling, so that the objectives of this study were to determine the rates and causes of voluntary and involuntary culling in five dairy herds in Khartoum State in relation to some farm management factors.

MATERIALS AND METHODS

Dairy farms and animals

The study was conducted in dairy cattle farms (N = 5) with a total number of 1324 dairy cows (milking or dry) and primiparous heifers in three localities in Khartoum State. The average farm size was 264.8±153.1 cow

Experimental design

Monthly visits were performed to each farm during the period from October 2009 to September 2010 to collect data by reviewing the records of these farms, by direct observations or directly by asking the farm owners or animal's attendants. The data included some management practices such as adoption of veterinary supervision, the type of houses, the type of provided feed and the information regarding the disposal of milking cows or primiparous heifers from the farm and the reasons behind this disposal. Culling due to economic reasons, aging or low milk yield was identified as voluntary culling, whereas, the involuntary culling involved culling cases due to health problems (Chronic mastitis, infertility and foot injuries). The term, economic causes, was used when the primiparous heifers were sold to maintain the expenses of the farm. Death cases were not considered. The causes and rates of culling were interpreted with regard to different management data.

RESULTS

Management practices:

Number of cows in each farm, adoption of some management practices, the overall percentages of culling (voluntary or involuntary) on the selected farms are shown in Table1. A total of 199 cows or primiparous heifers out of 1324 (15.0%, ranged from 5.7% to 45.1%) were culled due to different causes. 10.8% was voluntary culling and 4.2% was involuntary culling.

Causes and rates of voluntary and involuntary culling:

The most common causes of voluntary culling reported in this study were economic reasons (29.1%), low milk yield (23.0%) and aging (19.7%). The most common causes for involuntary culling were infertility (17.7%), chronic mastitis (8.5%) and foot injuries (2.0%), (Figure1). The highest overall percentage (29.1% of total culled) in this study was due to economic reasons and the lowest percentage (2.0% of total culled) of culling was due to foot injuries.

Causes and rates of culling in the different selected farms:

The causes and rates of voluntary and involuntary culling are shown in Figure 2. The highest rates of overall, voluntary and involuntary culling rates were reported in farms No. 3, 4 and 3 respectively.

Culling rates in relation to some farm management factors:

The overall, voluntary and involuntary culling rates in relation to some farms management factors are shown in Figure 3. The rates of voluntary culling and involuntary culling were higher in farms which practiced the veterinary supervision (18.7% and 7.7% respectively) than the rates in farms which did not (8.2% and 3.1% respectively). The voluntary and involuntary culling rates in farms constructed from fixed materials with adequate shade were lower (10.6% and 3.0% respectively) than the rates in farms which constructed from local materials (11.1% and 6.6% respectively). The voluntary and involuntary culling rates (10.6% and 3.0% respectively) were lower in farms used feed which was prepared within the farm than the rates (11.6% and 6.1% respectively) in farms which purchased feed from a feed company.

Causes of culling with regard to some farm management factors

Rates of causes of culling with regard to veterinary supervision: The causes of culling in relation to practice of veterinary supervision are shown in Figure 4. The highest rate of causes of voluntary culling in farms where the veterinary supervision was practiced was 41.9% for aging, whereas, the highest rate in farms which did not practice the veterinary supervision was 38.9% for economic causes and no case of involuntary culling due to foot injuries in farms which practiced the veterinary supervision.

Rates of causes of culling with regard to type of houses: The rates of causes of culling in relation to type of houses are shown in Figure 5. The prevalent causes of voluntary and involuntary culling in farms where the houses were constructed from fixed materials with adequate shade were aging (31.0%) and infertility (17.3%) respectively. Whereas, the prevalent causes of voluntary and involuntary culling in farms where the houses were constructed from local materials with inadequate shade were economic reasons (53.0%) and infertility (18.1%) respectively.

Rates of causes of culling in relation to type of feed: The rates of causes of voluntary and involuntary culling in relation to type of feed are shown in Figure 6. Culling due to low milk yield (35.3%) and infertility (17.2%) was prevalent in farms which used farm made feed, whereas, culling due to economic reasons (35.0%) and infertility (18.1%) was prevalent in farms used feed which purchased from feed companies.

Table1 - Some management practices adopted in dairy cattle farms in Khartoum State

Farm number	No. of animals	Veterinary supervision	Type of houses	Type of feed	% Culling		
					Overall	Voluntary	Involuntary
1	164	Practiced	Fixed materials with shade	Prepared within the farm	7.9	2.4	5.5
2	530	Not practiced	Fixed materials with shade	Prepared within the farm	5.7	5.7	0.0
3	162	Practiced	Fixed materials with shade	Prepared within the farm	45.1	35.2	9.9
4	214	Not practiced	Constructed from local materials	Purchased from a feed company	31.3	22.9	8.4
5	254	Not practiced	Constructed from local materials	Purchased from a feed company	6.3	1.2	5.1
Total	1324				15.0	10.8	4.2

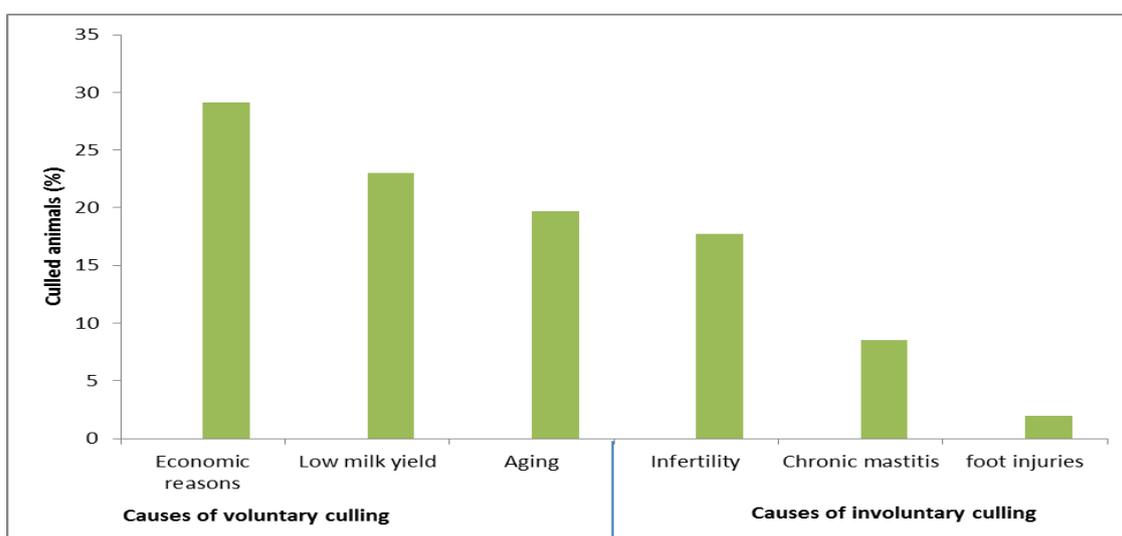


Figure 1 - Causes and rates of voluntary and involuntary culling in dairy cattle farms in Khartoum State, Sudan

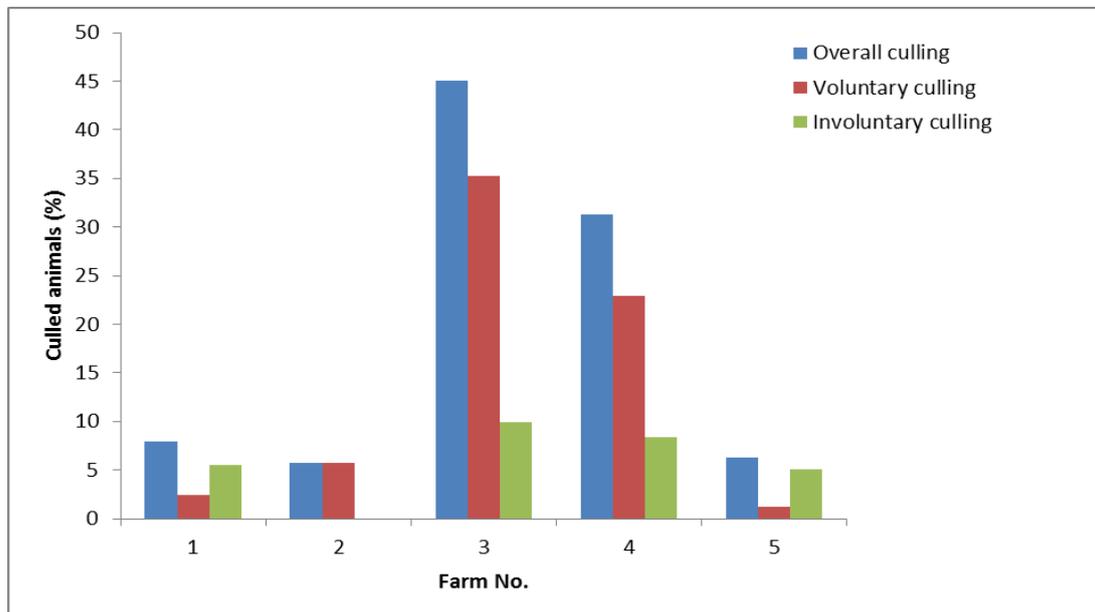


Figure 2 - Overall, voluntary and involuntary culling rates in different dairy cattle farms in Khartoum State, Sudan

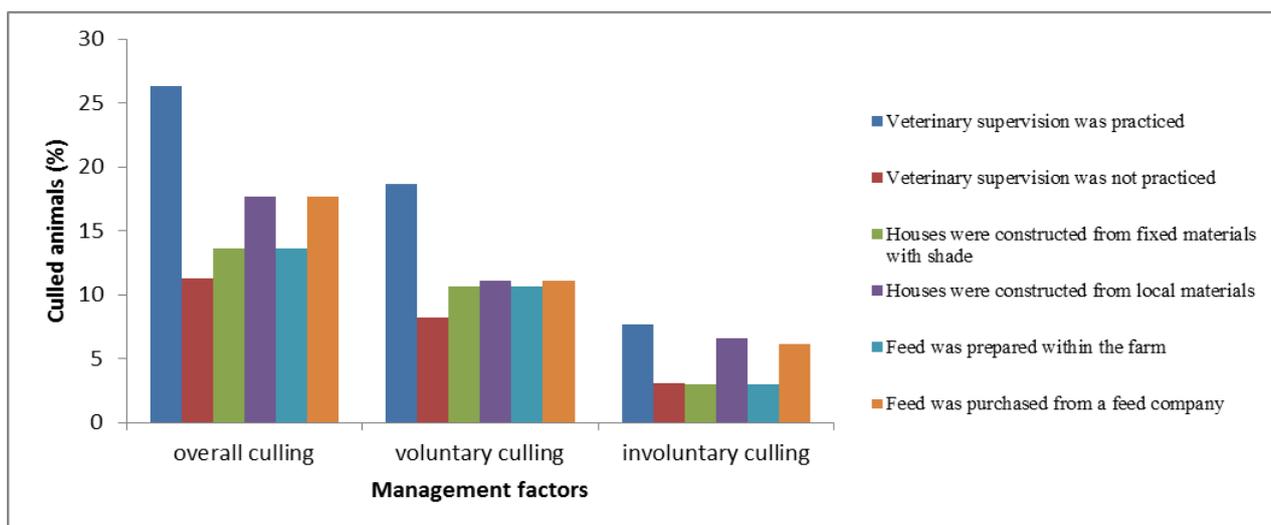


Figure 3 - Overall, voluntary and involuntary culling rates in relation to some farm management factors in cattle dairy farms in Khartoum State

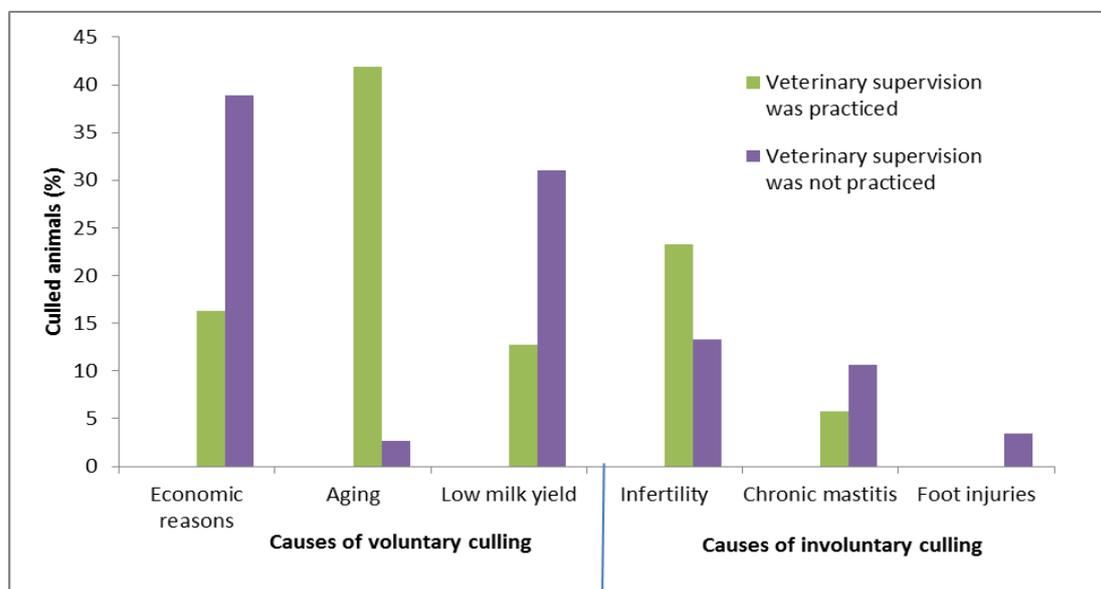


Figure 4 - Causes of culling in relation to practice of veterinary supervision in dairy cattle farms in Khartoum State

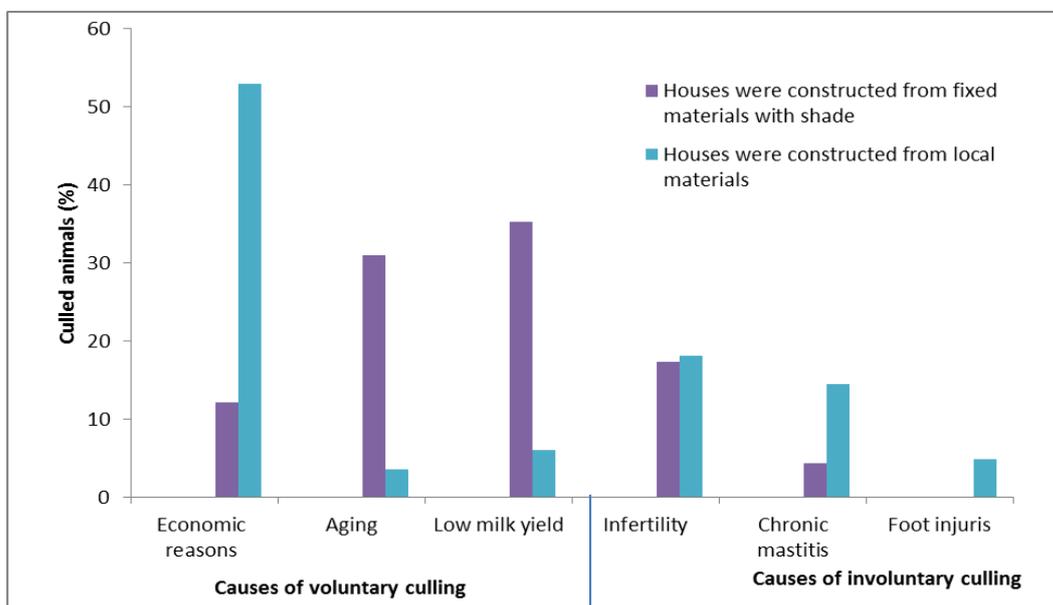


Figure 5 - Causes and rates of voluntary and involuntary culling in relation to type of houses in dairy cattle farms in Khartoum State

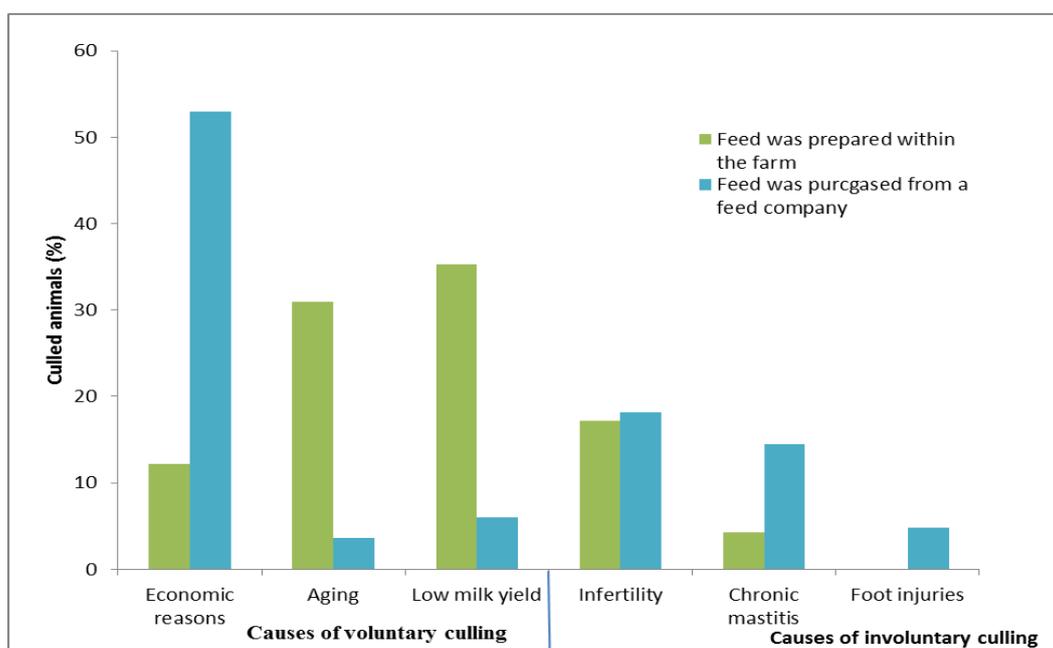


Figure 6 - Causes and rates of voluntary and involuntary culling in relation to type of feed in dairy cattle farms in Khartoum State

DISCUSSION

To make a right decision to remove an animal from the herd, many factors should be taken into considerations. These factors are the health status, age, reproductive performance, milk yield and stage of lactation (Allaire et al., 1977). The overall culling rate in this study was 15.0%. Voluntary culling represented 10.8% and involuntary culling represented 4.2%. The overall rates of culling and the proportions of voluntary and involuntary culling were varying in the five farms. This can be explained by the attitude of farmers towards culling process in different farms. The rate of overall culling in this study is near to that reported by Elimam et al. (1999) in Sudan (11.95%), Maher et al. (2008) in Ireland (19.6%) and Mohammadi and Sidighi (2009) in Iran (13.1%), however, this rate is slightly different from rates reported by others. Lari et al. (2012) reported 25.1%, including death cases, as overall culling rate in dairy cows in Shiraz, southern Iran. In this study death cases were not considered, the fact which may contribute to the low culling rate in this study. The proportions of voluntary and involuntary culling in this study constituted 71.8% and 28.2% respectively. These proportions were in agreement with Stevenson and Lean (1998) who reported that the voluntary culling was the most prevalent type of culling in New South Wales, Australia

and in agreement with the results of Rajala-Schultz et al. (2000) who reported that a total replacement percentage was 26 with the highest frequency of voluntary culling in Finish dairy herds. However, the findings disagreed with those of Mohammadi and Sidighi (2009) who reported overall culling rate of 13.1% with 98.5% for involuntary culling and 1.5% for voluntary culling in 23 Holstein dairy herds in Iran. Also this result disagreed with the results of Lari et al. (2012) who reported 74% for involuntary culling and 26% for voluntary culling. The increased percentage of voluntary culling as general is considered as a sign of good management practice (Stevenson and Lean, 1998; Lari et al., 2012). However, the high rate of voluntary culling in this study can be explained by the non-systematic and non-programmed culling practice in these farms as the owners just cull cows to be sold to maintain the financial needs. This explanation can be indicated by the high rate of culling (29.1% of total culled) due to economic reasons (sale of primiparous heifers to meet the financial needs) reported in this study. The reasons for voluntary culling reported in this study represented the most common reasons reported by many authors (Beaudeau et al., 1993; Mohammadi and Sidighi, 2009; Pinedo et al., 2010; Lari et al., 2012). Age has been reported by many authors as a factor which increases the risk of culling in dairy herds (Mohammadi and Sidighi, 2009; Saeed and Fadel Elseed, 2015; Gross et al., 2016). Saeed and Fadel Elseed (2015) reported that 79% of farmers in Sudan were culling their animals on the basis of aging and decline of production. Gross et al. (2016) reported that 15.5% were culled in Switzerland due to high age. Culling due to low milk yield is categorized with the voluntary type of culling, but low milk can be the end result of many factors like metabolic diseases, infertility and subclinical mastitis. These factors are interrelated and the discrimination between them is difficult. Low milk yield was the second most prevalent cause of culling in this study and was responsible for culling of 23.0% of total culled animals. This rate is relatively higher compared to rates reported by many authors. Anderson (1985) reported 3.7% culling rate due to low milk production. Mohammed and Sidighi (2009) reported 0.4% culling rate due to low milk yield and 1.1% due to aging. These low rates reported by these authors can be explained by the fact that owners keep the cow with low milk yield if it is fertile to complete the lactation cycle and then culled to benefit from their calves. High milk yield is always associated with deteriorated health and fertility and consequently, decreased milk yield and an increased culling rate (Espesito et al., 2014; Raboisson et al., 2014). Therefore, this high rate of culling due to low milk yield can be attributed to interrelations and strong links between low milk yield, infertility and health problems.

The most common causes of involuntary culling reported in this study were infertility (17.7%), chronic mastitis (8.5%) and foot injuries (2.0%). These reasons are common and have been reported by many authors. Beaudeau et al. (1993) reported that more than 50% of culling cases were due to health problems. Gross et al. (2016) reported that 28.4% dairy cows were culled in Switzerland due to infertility, 16.4% due to udder health and 10.4% due to claw health (lameness).

Infertility has been reported as an important cause of culling in dairy cows by many authors (Stevenson and Lean, 1998; Mohammadi and Sidighi, 2009; Lari et al., 2012). Esslemont and Kossaibati (1997) reported that poor fertility was the most important reason for culling of dairy cows in 50 dairy herds in England. Causes of infertility in dairy herds are of multiple origin such as genetic factors (Veerkamp et al., 2001), nutrition and management factors (Dubson et al., 2007), metabolic disorders (Wathes et al., 2009; Esposito et al., 2014). Infertility in this study may be genetic or due to other contributing factors such as reproductive system disorders, metabolic diseases, nutritional deficiencies or management factors. To explain and to define the decision of culling due to infertility as a right decision, it is important to evaluate the time of culling with regard to calving-culling interval, parity, and the fertility traits. Interpretation of culling due to infertility needs more elaborative studies to critically explain the reasons behind culling of dairy cows due to infertility. Farmers may keep highly producing infertile cows for longer time.

Losses due to mastitis can be regarded as a general problem in the dairy sectors worldwide (Bell et al., 2006; Huijps et al., 2008) and in Sudan (Saeed and Fadel Elseed, 2005; Mohammed and El Zubeir, 2015). Chronic mastitis represented 8.5% of total culled in this study. Many studies reported that mastitis is among factors which increase the risk of culling in dairy farms (Lari et al., 2012; Gross et al., 2014).

Lameness is reported as a problem in dairy cattle farms and as a cause for culling by many authors (Bell et al., 2006; Gross et al., 2016). In this study foot injuries represented 2.0% of total culled animals. The rate is fairly close to rates reported by many authors (Lari et al., 2012) who reported 3.5% culling rate due to lameness. Lameness or foot injuries are important detrimental factors of animal health as cows with foot injuries or lameness undergo low milk yield, infertility and other health problems due to restricted or reluctant movement required to achieve the normal daily activities.

Farm characteristics can help explain why cows are culled (Bascom and Young, 1998). The management practice of dairy farms selected for this study was poor as 75.4% (889 out of 1324 dairy cows) did not receive any type of veterinary supervision during the study period (September 2009 to October 2010) and only cows in two farms (326 cows) received the service (24.6%). In 40% of the farms, houses were constructed from local materials

and inadequate shade was provided and only 40% of the farms used feed which was made by expert feed companies. The poor veterinary service provided to dairy farms in Khartoum State has already been reported. Mohammed and El Zubeir (2015) stated that veterinary services in 60.8% of dairy farms in Khartoum State were provided by animal's owners or animal's keepers. Saeed and Fadel Elseed (2015) confirmed the poor veterinary services provided to dairy farms in Khartoum state and they found that, in a questionnaire based study, 63% of the respondents declared poor veterinary supervision in dairy farms in Khartoum State. This poor veterinary services status was, to our opinion, due to high cost of this service as it is provided by a private sector rather than the governmental one. The overall culling rate was higher in farms which received veterinary supervision (26.4%) compared to farms which did not (11.4%). The voluntary culling was most prevalent in the two situations. Culling due to aging represented the most prevalent cause of culling in farms received the veterinary supervision (41.9% of total culled). Whereas, culling due to economic reasons was prevalent in farms which did not. These findings can be poorly explained by the fact that the veterinarian could persuade the farm owners to dispose the senile cows, but as general, the scientific explanation and interpretations are difficult and further detailed studies are needed.

Regarding the type of houses and provided feeds, the findings were not greatly different and the links between these factors and the culling rate, the reasons for culling and rates of reasons of culling were not clear. These results are a true reflection of the fact that culling practice in these farms was not based on systematic or programmed models.

CONCLUSION

It can be concluded that the voluntary culling was the most prevalent type of culling in dairy cattle farms in Khartoum State during the period from September 2009 to October 2010 and animals mostly culled for economic reasons. Further studies are needed to critically evaluate and describe the strategies of culling in dairy farms in Sudan by studying the reproductive and productive characteristics of culled animals.

Competing Interest

The authors have declared that no competing interest exists.

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EPIDEMIOLOGICAL INVESTIGATION ON OUTBREAK OF BRUCELLOSIS AT PRIVATE DAIRY FARMS OF SINDH, PAKISTAN

Adnan YOUSAF^{1✉}, Muhammad ABBAS¹, Riaz Ahmad LAGHARI², Junaid HASSAN¹, Faiza RUBAB³, Tahseen JAMIL¹, Irfan HAIDER¹, Uzma ABBAS¹, Naila BIBI¹

¹Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandoajm, Pakistan

²Department of Veterinary Medicine, Sindh Agriculture University Tandojam, Pakistan

³Department of Animal Reproduction, Sindh Agriculture University Tandojam, Pakistan

✉Email: dr.adnan011@gmail.com

ABSTRACT: Brucellosis is one of the drastic diseases of zoonotic significance. Brucellosis is a global challenge not limited to Asia. As a result, economic losses are escalating due to the burden posed by Brucellosis in the investigated area. The present study was conducted to estimate the prevalence of brucellosis in cattle and buffaloes of Sindh, Pakistan. Blood samples were collected from (n=1200) animals (dairy cattle and buffaloes) of different age (2-7 years) and sex from 10 different districts of Sindh, Pakistan where no vaccination against brucellosis is practiced and were subjected to indirect ELISA for detection of *Brucella* antibodies. The overall mean prevalence was 18.16% with prevalence in unorganized is higher organized farms. In absence of any vaccination presence of circulating antibodies against *Brucella* in all age group of animals indicated the natural circulation of infection in the state. Based on the findings, the disease is seems to be endemic in the area, perhaps, due to partial or no vaccination. Also, the area presents poor bio-security measures and management. It is therefore, warranted to adopt good surveillance system for early identification of the brucellosis outbreak and appropriate measure for further control transmission of the brucellosis. Study indicated an urgent need of policy for prevention and control of brucellosis in dairy animals.

Keywords: Brucellosis, Prevalence, Privet dairy farms, Bio Security, Sindh

ORIGINAL ARTICLE
 pii: S222877011700002-7
 Received 25 Dec. 2016
 Accepted 20 Jan. 2017

INTRODUCTION

Bovine brucellosis is the one of the most dangerous zoonotic disease which prevalence is throughout all over the world (Makita et al., 2011), and after this the most important is rabies (Shafee et al., 2011). This disease is distributed in livestock almost all over the world (Anka et al., 2014). Annually approximately 60 thousand new cases are reported at hospital in human, worldwide, is re-emerging with significant veterinary and public health concern (Manish et al., 2013). In cattle, brucellosis is generally caused by the *B. abortus* and *B. melitensis* (Anka et al., 2014). Most of the *Brucella* strains are highly pathogenic for the human population (Goni et al., 2008) as indicated by the presence of *Brucella melitensis* and *Brucella abortus* in pregnant women (Khan et al., 2001) and *Brucella* in man (Mai et al. 2013). It is also called bang diseases (Durrani et al., 2015). Bovine brucellosis causes serious economic losses in the cattle and buffalo, as a consequences of the late abortion, stillbirths, slaughtering of the infected animals, reduction in meat and milk production (Calistri et al., 2013), low fertility and cost of replacement of the animals (Shafee et al., 2011).

Cattle and buffalo are large ruminants which are mostly considered for dual purpose milk and meat purpose. Buffaloes are also known as "black gold" (Soomro et al., 2014). Cattle and buffaloes play key role in GDP of Pakistan via producing milk and meat (Soomro et al., 2014). Such animals are also play a significant role in draught. Due to very poor hygiene condition and poor management and care these animals are facing very serious several health problems, such as brucellosis and colibacillosis diseases, which are economical loss (Mailk et al., 2013). An aerobic gram-negative bacterium is the causative agent of brucellosis which belongs from genus *brucella* (Yousaf et al., 2015). In human being the brucella organism was affirmed as the primary cause of brucellosis, which are transmitted from cattle/buffalo or goat, sheep's through contamination or other air borne pathogens (Buhari et al., 2013). In males it causes orchitis and epididymitis while abortion in the females (Anka et al., 2014). Preventive measure can be adopted by minimizing animal to animal transmission. The predominant sources of transmission occurs via mucous membrane, contact with the contaminated material, inhalation, milk etc (Mai et al., 2013). In farm animals the main reported genus cause of brucellosis are *brucella melitensis* and *brucella abortus* (Karaca et

et al., 2007). Compared to other microbes, *Brucella* has the capability to localize in supra mammary lymph nodes and mammary glands of infected animals (Calistri et al., 2013). This pathogenic process is a powerful signal for down regulation of the immune system.

In Sindh, cattle and buffaloes are indigenous animals. Different breeds of cattle and buffaloes are kept in large and small scale. Sindh is a border Province, where animals are brought from other cities/provinces (i.e., Karachi, Quetta and Punjab Province etc) without bio security or appropriate measure of quarantine, it may be cause of very serious and several infectious diseases such as Tuberculosis, Anthrax and Brucellosis. Accurate diagnosis, quarantine and proper culling of the infected animals from the herd are required for control and prevention of the disease.

MATERIALS AND METHODS

Study Area

Cross-sectional studies were conducted in 10 districts (Hyderabad, Tando Muhammad Khan, Karachi, Mir Pur Khas, Khair Pur, Larkana, Thatta, Jamshoro, Rani Pur and Kashmor) of Sindh, Pakistan. The climate of the investigated area is humid subtropical with dry winter type and the vegetation is tropical dry and low rainfall of 750-1,100 mm/year, with RH (relative humidity) (25-45%), and normal annual temperature (20 °C to 48 °C).

Collection of samples

In this study, blood samples from (n=1200) animals (of various sex, age and place) including (n=600) from unorganized and (n=600) from organized farms. Animals were kept in separate groups include adults, heifers, and young stock. All the animals at the farm were stall fed. No animal at the farm was vaccinated against the brucellosis. It was ensured very hygienic condition during blood sample collection by applying antiseptic solution on jugular vein of animals. About 5cc blood sample were collected from each and every animals and it was brought to laboratory on ice. The serums were collected by centrifugation at 3,000 rpm for 5 min. Samples were cooled immediately in ice jar as it was collected to transport for diagnostic laboratory. While at laboratory such samples were stored at -20 °C for as described by (Soomro et al., 2014).

Epidemiology Study

Descriptive epidemiology and retrospective cohort study was conducted. In descriptive epidemiology, the herd demographic and management practices were recorded. In retrospective cohort study risk factors associated with the *bovine brucellosis* were investigated. A questionnaire was designed, pre-tested and interviews of owner/employees were conducted. The attendants at farm were also investigated for any sign and symptoms related to the *brucellosis* and farm record was reviewed.

Sero Isolation of anti brucella antibodies

Rose Bengal Plate Test (RBPT) is standard key test for brucellosis in blood (Soomro et al., 2014). The serum samples were analyzed by Rose Bengal Plate Test and using commercially available kit (IDEXX brucellosis, USA) of enzyme linked immune sorbent assay-ELISA (OIE Manual 2004).

Analyzing Results

The data were analyzed descriptively and analytically by using the Epi Info version 7. For the univariate analysis, frequencies of variables and measures were calculated. $P < 0.05$ was considered to be statistically significant.

RESULT AND DISCUSSION

A total (n=1,200) blood samples were collected throughout 10 districts of Sindh, n=76 (12.67%) were positive for brucella *abortus* antibodies in organized dairy farm, whereas 142 (23.67%) were regarded as positive in unorganized dairy farm for *B. abortus* antibodies. Hence overall prevalence percentage in Sindh was documented too much (18.16%) in 10 districts (Table 1), while unorganized farm are much more infected as compare to the organized dairy farm due to their best management, stress free environment, proper medication and well diet ration. As noted, there was no quarantine measures adopted for new animals that are likely to introduce into the farm. The bovine brucellosis outbreak occurred in the month of May 2014. Brucellosis cases were high among human and cattle population between months of March and September (Lee et al., 2013).

In each district total 120 samples were collected from organized (n=60) and unorganized (n=60) dairy farm. District wise details of prevalence of brucellosis in dairy farms are as showed in (Table 2).

Table 1 - Comparison of Organized Dairy Farms & Un-organized Dairy Farm of Sindh.

Farms Details	Total Sample	Positive Result Sample	% +Ve Sample Result
Organized Dairy Farms	600	76	12.67%
Un-Organized Dairy Farms	600	142	23.67%
Total	1200	218	18.16%

Table 2 - Comparison of Organized Dairy Farms & Un-organized Dairy Farm districts wise of Sindh

District	Organized Dairy Farm		Unorganized Dairy Farm	
	No. of Positive Sample	Positive Result %	No. of Positive Sample	Positive Result %
Hyderabad	07	11.66	12	20
Tando M. Khan	12	20	13	21.67
Karachi	03	05	15	25
Mir Pur Khas	05	8.33	11	18.33
Khair Pur	04	6.67	17	28.33
Larkna	03	05	09	15
Thatta	09	15	18	30
Jamshoro	08	13.33	13	21.67
Rani Pur	12	20	16	26.67
Kashmor	13	21.67	18	30
TOTAL	76	12.67	142	23.67

The farm management (Biosecurity and housing) was very poor because the waste material was not properly disposed especially the aborted material which is the source of the infection. It's may be due to lack of awareness. There was no proper disposal of the aborted material at farms in district Hyderabad (Soomro et al., 2014). There were no proper fences or boundary wall around the farm. Rodents and dogs were also present at the farm.

The prevalence with multiple risk factors and different herds always has the possibility of false positive results affecting the final outcome of the study. However, the sensitivity of particular test in any study model reduces the chances of false positivity. It is evaluated from the current study that a wide spread presence of brucellosis in dairy cattle and buffalos are documented in these selected districts of Sindh. Presences of *brucella* antibodies are mostly found in non-vaccinated. The prevalence in un-organized farms is at alarming level. Moreover, animal owners/farmers in this area are in close contact with these animals, and consumption of raw milk and improper handling of aborted materials is frequent. Thus brucellosis is not only the cause of reproductive and production losses but also may be the potential biohazard in this region. Thus, from the current study the more study regarding brucellosis in this area are suggested epidemiological investigations to find out the link between animal and human brucellosis. To get control on this disease and can get control against such dangerous disease.

CONCLUSION

Present study is summarized that the management conditions of the farm were not organized. The area was not fenced for stop the rodents and stray dogs, waste material not properly disposed, no separate pens for aborted and sick animals, poor quality of drinking water, feed not stored properly and no proper treatment of the animals. The workers in the farm working without biosecurity measures. There was no surveillance system working in the areas to identify the brucellosis outbreak.

A good surveillance system is important to early identify brucellosis outbreak and take immediate measure for further control and transmission of the brucellosis. Vaccination and quarantine of newly introduced animals should be regularly practiced. A strict biosecurity measures should be adopted at the farm. The waste materials should be properly handled to reduce the challenges of brucellosis challenges from the potential source of infection.

Acknowledgments

The authors are thankful to Dr. Wazhat Hussain Bangash, for their full support and encouragement during the whole period of study. We are also grateful to Engr. Shahzad Yousaf and technical staff of private dairy farm Sindh for courteous co-operation.

Conflict of interests

The authors declare that they have no conflict of interest with respect to the research, authorship, and/or publications of this article.

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EFFECT OF AGE WISE INCUBATION PROGRAMME ON BROILER BREEDER HATCHABILITY AND POST HATCH PERFORMANCE

Adnan JABBAR, Adnan YOUSAF✉

Sadiq Poultry (Pvt) Limited, Rawalpindi, Pakistan

✉Email: dr.adnan011@gmail.com

ABSTRACT: Temperature and humidity are most important environmental factors during incubation. The age of birds affect the eggs, its internal as well as external quality, that's why dissimilar conditions require for incubation. The aim of this study was to investigate the effect of age wise incubation profile on hatchability and chick's performance. For this experiment, eggs were collected from Ross-308 breeders which were divided into four groups according to the age of breeders having equal number of eggs in all groups (n=538560 eggs). Group A (Young, 24-31 weeks), B (Prime, 32-50 weeks), C (Old, 50+weeks) and D (control). For groups, A, B and C duration of incubation in setter machine was 456 hours (19th day) while for D (control), incubator duration was 449 hours (18.7 days). Fertility of eggs were performed through candling and shifted to hatchers for next 50 hours for A, B and C while 56 hours for D. Group B was significantly better (P<0.05) as compare to A in term of hatchability. Candling was significantly better for group B (P<0.05) than C. Group C was significantly (P<0.05) better for candling than A and D which contain same candling i.e. A and D. Groups A and B have significantly (P<0.05) same dead in shell (DIS) (6.18±0.29 and 6.20±0.37 respectively) as compare to C (7.13±0.60). Group D (6.70±0.67) was same for DIS as groups A, B and C. Excellent performances of chicks with mortality (P<0.01) for A (2.93±0.60), B (2.77±0.49), C (2.85±0.53), D (3.10±0.82), weight gain (gram/bird) (P<0.01) for A (1972.66±0.33), B (2012.33±35.92), C (1996±14), D (1985.33±18.58), FCR (P<0.01) for A (1.55±0.12), B (1.51±0.15), C (1.54±0.13), D (1.57±0.11) and feed intake (gram/bird) (P<0.01) for A (3146.92±189.13), B (3138.63±203.4), C (3139.75±201.55), D (3166.72±154.84) all were same in all groups. Water loss was different for A (11.93±0.60) then B (12.34±0.76), C (12.24±0.65) as compare to D (10.17±0.55). Chick yield was same for A (69.28±0.18), B (69.51±1.12) and C (69.28±0.88) then D (71.46±1.54) had more yield and low water loss. Group A, B, and C were better as compared to D but overall group B has significantly better which mean the eggs from different ages of birds require different conditions of incubations and it will not affect the chick's performance at the farm.

Keywords: Broiler, Age Wise Incubation Regimen, Hatchability, Dead in Shell, Candling, Performance

INTRODUCTION

Hatching egg quality and incubation conditions influence broiler performance. Growth rates of broiler chickens have been increasing day by day due to genetic selection and latest technology incubation system (Hulet, 2007; Baghbanzadeh and Decuyper, 2008). Numerous factors have been documented to influence fertility and hatchability of chicken eggs (Alsobayel and Albadry, 2012). Age of the hen appears to have an influence on fertility, hatchability and embryonic mortality (Alsobayel and Albadry, 2012). Fertility and hatchability of chicken decreased the age (Insko et al., 1947). Newly-hatched chicks can have their thermoregulatory capability affected by incubation temperature (Tzschentke and Rumpf, 2011), which can also affect their blood hormones levels (Christensen et al., 2001) and development after hatching (Molenaar et al., 2011).

Temperature and Humidity play a key role which is one of the most important environmental factors during incubation (Lourens et al., 2005). At end of incubation period, the eggshell temperature increases due to the higher heat production of the embryo (Lourens et al., 2005). Different temperatures degrees are required to the embryo at different stages and ages (Lourens et al., 2005). Modern broiler chickens are extra sensitive to metabolic disorders such as ascites because of the genetic selection for quick growth, low feed conversion ratio (FCR), and high meat yield (Balog, 2003; Arce-Menocal et al., 2009), which has resulted in decreased visceral organ development (Havenstein et al., 2003).

The aim of the research was to study the impact of broiler breeders' age on its eggs incubation Programme.

MATERIALS AND METHODS

Site Selection

The current study was designed at Sadiq Poultry (Pvt) Limited, Chakri Hatchery Rawalpindi (Pakistan), which is situated 5km from chakri interchange on M2 motorway (Rawalpindi-Lahore). It is fulfilled from the latest automation and ISO 9001- 2000 certified hatchery. There were selected three category age wise breeder flocks, to assess the comparison of hatchability, chick's quality and post hatched performance of broiler chicks.

Eggs selection and Classification

Ross-308 breeder's four groups classified according to breeder's age. Each experimental group was consisting of (n=4, 03,920 eggs) with three replicates of (n=134640 eggs). Group D (Setter) was served as control group, while group A, B and C were treated with accordingly their age wise incubation regimen. Group A (Young age breeder eggs 24-31 weeks), Group B (Prime age breeder eggs 32-50 weeks), Group C (Old age breeders eggs 50+weeks) and Group D (control) having eggs of different age breeders. High-quality hatching egg shells were smooth, without ridges or small lumps of calcified material (pimples). The color of eggs within a flock was uniform. Young flocks produce eggs with thicker shells and when the flock older the shell becomes thinner and the incidence of abnormal shells increases. The eggs were graded on the basis of their quality and stander, all the hatchable eggs were graded through egg grading machine MOBA 9A. While the poor shell, crack, bloody stained, elongated eggs were rejected (Khan et al., 2016), only oval shape and good quality of eggs were selected. All group's eggs were fumigated with 20 g KMnO₄ and 40ml formalin40%, and 40 ml of water for 100ft³ areas for 15 minutes.

Incubation Regimen

Groups A, B and C incubation duration in setter machine was 456 hours (19th day) while for D (control) in incubator duration was 449 (18.7 days). Fertility of eggs were performed through candling then shifted to hatcher for next 50 h for A, B and C while 56 h for D. All of these stage regimens were recommended by chick master USA. All the groups were treated for full time incubation period 21 days (506 hours). Body weights of chick were determined immediately after chick collection. Candling and DIS (dead in shell) were recorded for each group individually.

Chicks Counting/Grading

Hatch pull out was performed through shell separator and grading was performed on conveyer and grading table. Only good quality chicks having soft legs, shining eyes soft feathers, nose good naval were selected for the experiment. Chicks were shifted to box after counting, while dead, week, and unhealed naval chicks were removed.

Delivery to Poultry House

Each experimental group was consisting of (n=74,000) day old chicks, with 4 replicate (each replicate n=18,500). Environmentally control vehicles (75 °F temperatures, 65% humidity) were used to deliver the chicks up to poultry houses with 102 chicks/box packing to Sadiq Broiler Farm Khilri chakri. Continuous light was provided during whole study. Poultry house condition was same for all groups as shown as below (Table 1). Chicks were fed with starter diets from 1 to 12 d (3020 Kcal ME/kg, 22% CP), grower diets from 13 to 22 d (3185 Kcal ME/kg, 20% CP) and finisher diets from 23 to 35 d of age (3230 Kcal ME/kg, 18% CP). The diet was formulated according to the recommendations of the NRC (1994) using WUFFDA formulation software program. Intake of feed and water was taken daily, while body weight and total feed consumed was recorded on weekly basis. Viper Touch (Big Dutchman, Co., Germany) system was installed.

Table 1 - Environmental condition of Poultry House

Parameters	1 st Week	2 nd Week	3 rd Week	4 th Week	5 th Week
Temperature °F	95-86	86-83	83-77	77-75	75
Humidity %	65	65	65	65	65
Ventilation (m ³ /hour/bird)	0.07	0.25	0.40	0.59	0.87

Statistical analyses

All data were analyzed by using Statistical Analysis System package software (SAS version 9.2, SAS Institute Inc., Cary, NC, USA). All means were compared using Duncan's Multiple Range test and results were presented as mean ± SEM (standard error of mean). Results were considered significant if exist P<0.05.

RESULTS

After ten successful hatches out for Individual group, hatch abilities for individual groups were recorded. Hatchability was significantly ($P<0.05$) better for group B (89.02 ± 0.41) than A (86.66 ± 0.33 , C (86.80 ± 0.65) and D (86.25 ± 1.22). Candling was significantly better ($P<0.05$) for B (4.77 ± 0.17) than C (6.05 ± 0.67). Group C was significantly ($P<0.05$) better for candling than A (7.15 ± 0.33) and D (7.03 ± 0.76) which contain significantly same in term of candling i.e. Group A and D. Group A and B have significantly ($P<0.05$) same dead in shell (DIS) (6.18 ± 0.29) (6.20 ± 0.37) respectively as compare to group C $P<0.05$ (7.13 ± 0.60). Whereas, D significantly ($P<0.05$) (6.70 ± 0.67) same in term of DIS as group A, B and C shown (Tables 2 and 3).

During 35 d trail period, mortality, feed intake, weight gain and FCR was recorded and results were presented in Table 3. Interestingly, the effect of age wise regimen on broilers performance was also found same. Mortality was reduced significantly ($P<0.05$) in A (2.93 ± 0.60), B (2.85 ± 0.53), C (2.77 ± 0.49) as compared to control D (3.10 ± 0.82) group. Weight gain (g/bird) was regimen same ($P<0.05$) in A (1972.66 ± 0.33), B (2012.33 ± 35.92), C (1996 ± 14) groups as compared to control group (1985.33 ± 18.58). Feed conversion ratio (FCR) was found significantly same ($P<0.05$) in A (1.55 ± 0.12), B (1.51 ± 0.15), C (1.54 ± 0.13) than D group (1.57 ± 0.11). However, feed intake (g/bird) was not affected ($P>0.05$) by the age wise regimen of eggs group A (3146.92 ± 189.13), B (3138.63 ± 203.4), C (3139.75 ± 201.55) and control group D (3166.72 ± 154.84) were significantly same (Table 4). Water loss was remain significantly different for A (11.93 ± 0.60) then B (12.34 ± 0.76), C (12.24 ± 0.65) as compare to D (10.17 ± 0.55). Whereas, the hatching chicks yield was remain same for A (69.28 ± 0.18), (B 69.51 ± 1.12) and C (69.28 ± 0.88) then D (71.46 ± 1.54) had more chicks yield low water loss, thus chicks had low yolk absorption (Table 4).

Table 2 - Effect of age wise and combine incubation programme

Groups	A (Young)	B (Prime)	C (Old)	D (Combine)
Hatchability %	86.66 ± 0.33^b	89.02 ± 0.41^a	86.80 ± 0.65^b	86.25 ± 1.22^b
Candling %	7.15 ± 0.33^a	4.77 ± 0.17^b	6.05 ± 0.67^c	7.03 ± 0.76^a
DIS %	6.18 ± 0.29^a	6.20 ± 0.37^a	7.13 ± 0.60^b	6.70 ± 0.67^{ab}

^{a-b} denotes difference in columns ($P<0.05$), DIS= dead in shell

Table 3 - Dead in shell (DIS) analysis report

Weeks	A (Young)	B (Prime)	C (Old)	D (Combine)
1 st week %	1.46 ± 0.14^a	1.46 ± 0.17^a	1.76 ± 0.14^b	1.62 ± 0.15^{ab}
2 nd week %	0.54 ± 0.4^a	0.47 ± 0.7^a	0.55 ± 0.8^a	0.58 ± 0.5^a
3 rd week %	2.71 ± 0.73^a	2.79 ± 0.74^a	3.36 ± 0.65^b	3.28 ± 0.74^b
Clear %	0.67 ± 0.41^a	0.69 ± 0.45^a	0.64 ± 0.38^a	0.65 ± 0.32^a
Contamination %	0.56 ± 0.14^a	0.53 ± 0.34^a	0.55 ± 0.26^a	0.45 ± 0.37^a
Crack %	0.24 ± 0.4^a	0.26 ± 0.7^a	0.27 ± 0.8^a	0.23 ± 0.7^a
Total DIS	6.18 ± 0.29^a	6.20 ± 0.37^a	7.13 ± 0.60^b	6.70 ± 0.67^{ab}

^{a-b} denotes difference in columns ($P<0.05$) DIS= dead in shell

Table 4 - Chicks Performance of different groups at farm

Groups	A (Young)	B (Prime)	C (Old)	D (Combine)
Mortality %	2.93 ± 0.60^a	2.77 ± 0.49^a	2.85 ± 0.53^a	3.10 ± 0.82^a
Weight gain (g)	1972.66 ± 0.33^a	2012.33 ± 35.92^a	1996 ± 14^a	1985.33 ± 18.58^a
Feed in take (g)	3146.92 ± 189.13^a	3138.63 ± 203.4^a	3139.75 ± 201.55^a	3166.72 ± 154.84^a
FCR	1.55 ± 0.12^a	1.51 ± 0.15^a	1.54 ± 0.13^a	1.57 ± 0.11^a

^{a-b} denotes difference in columns ($P<0.05$)

Table 5 - Water loss and Chick yield Age wise and Combine Incubation Programme

Groups	A (Young)	B (Prime)	C (Old)	D (Combine)
Water Loss %	11.93 ± 0.60^a	12.34 ± 0.76^b	12.24 ± 0.65^b	10.17 ± 0.55^d
Hatching Yield %	69.28 ± 0.18^a	69.51 ± 1.12^a	69.28 ± 0.88^a	71.46 ± 1.54^d

^{a-b} denotes difference in columns ($P<0.05$)

DISCUSSION

Uniform egg shell temperature is necessary tool to avoid condensation (Marandure, 2012). To achieve better hatchability and good quality of chicks uniform shell temperature and avoid condensation is necessary (Renema et al., 2006). Significant deterioration in the characteristics of the eggshell with the age of the hens was also found (Yilmaz and Bozkurt, 2009). Changes in the quality of eggs, especially of their shells, with the passage of reproductive season can affect the embryonic development during the incubation period and, finally, the hatchability results (Nowaczewski et al., 2016). Hatchability parameters and embryonic mortality are directly affected due to age of female's breeder during incubation period (Al-Bashan and Al-Harbi., 2010, Othman et al., 2014). Age wise incubation stage programme have three different incubation temperature and humidity set point for the eggs given by different age of breeders i.e. prime, young and old which provides uniform temperature for growing embryos, helps to achieve good quality chicks. Uniform temperature of every stage for developing embryos enhances the performance of chicks at farm (Fasenko, 2009) result better FCR and less mortality. It was clearly documented that best egg fertility and hatchability in hen age between 31 and 50 weeks (Islam et al., 2008) as shown in result that group B is better. Better result in term of hatchability and candling, DIS water loss and chick yeild in Ross 308 broiler breeders of Prime aged 32–50 weeks in comparison with older flock age of 51 +weeks (Elibol and Brake, 2006). Higher DIS was recorded in older flock (Nowaczewski et al., 2016). The finding of current study tended to show that age wise incubation stage programme to broiler breeder eggs enhances the hatchability. The high level of DIS and maximum infertility was recorded in older flock age (Nowaczewski et al., 2016) as shown in (Table 2). Hatchability, candling and DIS was found better for group A, B, and C then D but overall group B was found better due to prime age production (Tables 3 and 4). Greater water loss is a result of better chick yield as result good quality of chicks Table 4. So, incubation of eggs with their age wise temperature and humidity set points provides better results as compare to incubation of different age's eggs at same temperature and humidity set points. The age wise and combine incubation profile will not affect the FCR, Feed intake, weight gain of chicks at farms.

CONCLUSION

Age wise incubation stage programme is a good factor, which may be used to enhance the hatchability.

Authors' contribution

Both authors carry equal contribute in this study.

Acknowledgments

The author is thankful to Director of Sadiq Poultry (Pvt) limited Mr. Salman Sadiq and Project Coordinator Mr. Jawad Qazi for their full support and encouragement during the whole period of research work. Authors are also great full hatchery supervisor Mr. Muhammad Ashfaq and Plant Supervisor Mr. Muhammad Akhtar for cooperation.

Conflict of Interests

The authors declare that they have no conflict of interest with respect to the research, authorship, and/or publications of this article.

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EFFECT OF DIETARY FEED ADDITIVES ON HAEMATOLOGICAL AND SERUM BIOCHEMICAL PARAMETERS OF BROILER CHICKENS

Emmanuel Olakunle ALONGE¹, Daisy ERUVBETINE¹, Olusegun Mark Obafemi IDOWU¹, Adewale Olusegun OBADINA³ and Oladapo Oluwaseye OLUKOMAIYA²✉

¹Department of Animal Nutrition, Federal University of Agriculture, P.M.B. 2240, Abeokuta, Nigeria

²Department of Animal Production and Health, Federal University of Agriculture, P.M.B. 2240, Abeokuta, Nigeria

³Department of Food Science and Technology, Federal University of Agriculture, P.M.B. 2240, Abeokuta, Nigeria

✉Email: oladapooluwaseye@gmail.com

ABSTRACT: The effect of dietary feed additives on haematological and serum biochemical parameters of broiler chickens was evaluated. 180 day-old Arbor acre broiler chicks were weighed and randomly allotted to five dietary treatments with 3 replicates of 12 birds each. Broiler starter diet (2855.7 kcal/kg ME; 23.01%) and finisher diet (2911 kcal/kg; 20.71% CP) were formulated. Dietary treatments were control diet (basal diet without additives), OXYT diet (basal diet with oxytetracycline at 600 ppm as antibiotic, GRO-UP diet (basal diet with probiotic at 500 ppm), MOS-500 diet (basal diet with mannan oligosaccharide at 500 ppm) and MOS-1000 diet (basal diet with mannan oligosaccharide at 1000 ppm). Feed and water were supplied ad libitum. At the end of weeks 4 and 8, blood samples were collected and analyzed. The haematological and serum biochemical parameters of broiler chickens fed diets containing feed additives at the starter phase were not statistically significant ($P>0.05$). At the finisher phase, there were no significant ($P>0.05$) differences in all the parameters measured except in the heterophils and eosinophils where birds fed the control diets had the lowest value among all treatments. Serum globulin values were significantly ($P<0.05$) different as birds fed diets containing OXYT (antibiotics) recorded the lowest value among all treatments. The inclusion of prebiotics and probiotics in the diets of broiler chickens elicited no adverse effect on haematological and serum biochemical parameters, thus, they can be used as replacement for antibiotics.

Keywords: Haematology, Serum Biochemistry, Broiler Chickens, Prebiotics, Probiotics

ORIGINAL ARTICLE
 pii: S222877011700004-7
 Received 01 Oct. 2016
 Accepted 10 Jan. 2017

INTRODUCTION

The benefits of broiler chicken production cannot be over emphasized in the face of increasing demand for animal protein in many developing countries of the world. To improve production, poultry birds must be free from infections as well as being fed with suitable diets needed for optimal production (Tannock, 1988). Antibiotics are widely being used in animal feed to enhance animal performance and production. Tetracyclines are arguably the most commonly-used therapeutic antibiotics in food animal production (Fairchild et al., 2005). Oxytetracycline (OTC) is a broad spectrum antibiotic developed to enhance the control of bacterial infections (Alam, 2000). The dietary inclusion of 50 mg/kg OTC during 21-42 days of age increased body weight gain of broiler chicks (Zulkifli et al., 2000). Talabi et al. (2013) also stated that the use of dietary oxytetracycline antibiotic powder at 0.05 g/kg promoted the growth of broiler chicks under different feeding regimes.

However, at low levels of antibiotic administration, resistant microbial cells survive and produce resistance. Thus, the use of antibiotics in animal feeds has been limited in the European Union since January 2006 (Toghyani et al., 2011). As a result, other feed additives such as prebiotics and probiotics have been proposed to livestock producers. They are being increasingly adopted as replacements for antibiotics in improving growth and gut health in poultry and swine (Higgins et al. 2008; Markovic et al. 2009; Zhang and Kim, 2013). Probiotics are live microbial feed supplements that beneficially affect the host animal by improving its intestinal health (Fuller, 1989). Prebiotics are non-digestible food ingredients that beneficially affect a host animal by selectively stimulating the growth and/or activity of one or more bacterial population in the colon (Gibson and Roberfroid, 1995). Due to the fact that more information is still needed on the haematology and serum biochemistry of broiler chickens as far as animal health is concerned, considerable attention is still being drawn towards testing the potency of dietary prebiotics and probiotics as antibiotic substitutes.

Therefore, the present study was carried out to investigate the effect of dietary feed additives on haematological and serum biochemical parameters of broiler chickens.

MATERIALS AND METHODS

The experiment was conducted at the Poultry Unit of Directorate of University Farms, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. The area lies within the rainforest zone of South-Western Nigeria at longitude 7°10', 37'N, latitude 3° 26' 58'E and altitude 173 m above sea level. The climate is humid with a mean annual rainfall of 1037 mm. The mean annual temperature and humidity are 34.7°C and 82%, respectively (Google Earth, 2013). 180 day-old Arbor acre broiler chicks raised on formulated broiler starter and finisher diets were used. They were weighed and randomly divided into 5 groups with 3 replicates of 12 birds each in a completely randomized design. They were brooded for two weeks in a deep litter system. Water and feed were supplied *ad libitum*. Routine medications were administered to the birds accordingly. The test ingredients used were Kepro® Oxytetracycline (antibiotics) - feed grade oxytetracycline (OXYT) was obtained from a reputable veterinary pharmacy shop in Abeokuta having the composition (per g) consists of oxytetracycline hydrochloride 1000 mg. GRO-UP™ (probiotics) was supplied by Bio Ingredients Ltd., Lagos, Nigeria with the composition (per kg) of *Saccharomyces cerevisiae*: 1.5x10¹¹ cfu, *Lactobacillus sporogenes*: 30 million cfu and fortified with phytase phosphorus, proteins, calcium, carbohydrates and vitamins. Oligomanno® (Mannan Oligosaccharides) (prebiotics) was supplied by Yonichi Chemical Institute Co., Ltd. Machikita 9-25, Moriyama-Ku, Nagoya, Japan. Composition: hydrolyzed Guar gum fiber (Mannan Oligosaccharide). Diet 1 was the control without feed additive, diet 2 had the inclusion of OXYT at 600 ppm, diet 3 had the inclusion of GRO-UP at 500 ppm while diets 4 and 5 had the inclusion of MOS at 500 ppm and 1000 ppm respectively as presented in Tables 1 and 2.

Table 1 - Ingredient composition of starter experimental diet (0-4 weeks)

Ingredients (%)	Diets				
	1	2	3	4	5
Maize	50.66	50.66	50.66	50.66	50.66
Wheat offal	5.00	5.00	5.00	5.00	5.00
Fish meal (72%)	3.00	3.00	3.00	3.00	3.00
Soybean meal	24.24	24.24	24.24	24.24	24.24
Groundnut cake	10.00	10.00	10.00	10.00	10.00
Palm kernel cake	3.00	3.00	3.00	3.00	3.00
Bone meal	2.00	2.00	2.00	2.00	2.00
Oyster shell	1.00	1.00	1.00	1.00	1.00
Lysine	0.10	0.10	0.10	0.10	0.10
Methionine	0.25	0.25	0.25	0.25	0.25
*Premix	0.50	0.50	0.50	0.50	0.50
Salt (NaCl)	0.25	0.25	0.25	0.25	0.25
¹ OXYT®	-	+	-	-	-
² GRO-UP®	-	-	+	-	-
³ MOS®	-	-	-	+	++
Total	100.00	100.00	100.00	100.00	100.00
Calculated analysis					
Crude Protein (%)	23.01	23.01	23.01	23.01	23.01
ME (Kcal/kg)	2855.7	2855.7	2855.7	2855.7	2855.7

*A kilogramme premix contains Vit. A: 10000000 IU, Vit. D₃: 2500000 IU, Vit. E: 20000 mg, Vit. K₃: 3000 mg, Vit. B₁: 30000 mg, Vit. B₂: 7000 mg, Vit. B₆: 5000 mg, Vit. B₁₂: 25mg, Panthothenic acid: 10000mg, Folic acid: 800 mg, Biotin: 50mg, Manganese: 80000 mg, Iron: 40000 mg, Zinc: 60000 mg, Copper: 8000 mg, Cobalt: 250 mg, Iodine: 1000 mg, Selenium (1%), 150 mg, Choline: 200000 mg and Antioxidant: 100000 mg.

-: no additive; +: 500 ppm of MOS; ++: 1000 ppm of MOS

¹OXYT® (antibiotics) at 600 mg/kg inclusion; ²GRO-UP® (probiotics) at 500 mg/kg inclusion and ³Mannan oligosaccharides® (prebiotics) at levels of 500 mg/kg and 1000 mg/kg respectively.

At end of weeks 4 and 8, blood samples were drawn from the wing (brachial vein) of two birds per replicate. 2 ml of blood was collected from two birds per replicate into the tube containing Ethylene Diamine Tetra Acetate (EDTA) as anti-coagulant and another 2 ml was collected for serum analysis. The haematological analysis of RBC, WBC and its differentials, MCH, MCHC, MCV, PCV and Haemoglobin concentration (Hb) were done according to standard methods (Schalm, 1986). The second set of bottles without EDTA was centrifuged in a macro centrifuge to obtain serum for biochemical analysis. Serum glucose was determined colorimetrically using GOD/PAD reagent method (Trinder, 1969). Serum uric acid was determined using enzymatic colorimetric method as described by Fossati and Prencipe (1982). Serum cholesterol was determined using enzymatic end point method (Roschlau et al., 1974). Serum total protein was analyzed using bromo cresol purple method of Varley et al. (1980). Serum albumin was determined by the bromo cresol green (BCG) method as described by Doumas et al. (1971). Serum globulin was calculated as the difference between total serum protein and serum albumin. Serum creatinine was analyzed using colorimetric method (Bowers and Wong, 1980).

Table 2 - Ingredient composition of finisher experimental diet (4-8 weeks)

Diets	1	2	3	4	5
Ingredients (%)					
Maize	55.00	55.00	55.00	55.00	55.00
Wheat offal	6.00	6.00	6.00	6.00	6.00
Fish meal (72%)	2.00	2.00	2.00	2.00	2.00
Soybean meal	18.00	18.00	18.00	18.00	18.00
Groundnut cake	13.00	13.00	13.00	13.00	13.00
Bone meal	2.00	2.00	2.00	2.00	2.00
Oyster shell	3.00	3.00	3.00	3.00	3.00
Lysine	0.20	0.20	0.20	0.20	0.20
Methionine	0.30	0.30	0.30	0.30	0.30
*Premix	0.25	0.25	0.25	0.25	0.25
Salt (NaCl)	0.25	0.25	0.25	0.25	0.25
¹ OXYT®	-	+	-	-	-
² GRO-UP®	-	-	+	-	-
³ MOS®	-	-	-	+	++
Total	100.00	100.00	100.00	100.00	100.00
Calculated analysis					
Crude Protein (%)	20.71	20.71	20.71	20.71	20.71
ME (Kcal/kg)	2911	2911	2911	2911	2911

*A kilogramme premix contains Vit. A: 10000000 IU, Vit. D₃: 2500000 IU, Vit. E: 20000 mg, Vit. K₃: 3000 mg, Vit. B: 30000 mg, Vit. B₃: 3000 mg, Vit. B₂: 7000 mg, Vit. B₆: 5000 mg, Vit. B₁₂: 25mg, Panthotenic acid: 10000mg, Folic acid: 800 mg, Biotin: 50mg, Manganese: 80000 mg, Iron: 40000 mg, Zinc: 60000 mg, Copper: 8000 mg, Cobalt: 250 mg, Iodine: 1000 mg, Selenium (1%), 150 mg, Choline: 200000 mg and Antioxidant: 100000 mg.
 -: no additive; +: 500 ppm of MOS; ++: 1000 ppm of MOS
¹OXYT® (antibiotics) at 600 mg/kg inclusion; ²GRO-UP® (probiotics) at 500 mg/kg inclusion and ³Mannan oligosaccharides® (prebiotics) at levels of 500 mg/kg and 1000 mg/kg respectively.

Statistical analysis

Data obtained were subjected to Analysis of Variance (ANOVA) in a Completely Randomized Design (CRD) using SAS (2003). Significant means among variables were separated using Duncan's Multiple Range Test.

RESULTS

The haematological and serum biochemical parameters of broiler chickens fed diets containing feed additives at Week 4 are presented in Tables 3 and 4 respectively. Variations in the parameters measured were not statistically significant (P>0.05).

The haematological parameters of broiler chickens fed diets containing feed additives at Week 8 are shown in Table 5. There were no significant (P>0.05) differences in all the parameters measured except in the heterophils and eosinophils where birds fed the control diets had the lowest value among all the treatments. The serum biochemical parameters of broiler chickens fed diets containing feed additives at Week 8 are shown in Table 6. There were no significant (P>0.05) differences in all the parameters measured except in serum globulin where birds fed diets containing OXYT (antibiotics) recorded the lowest value among all the treatments.

Table 3 - Haematological parameters of broiler chickens fed diets containing feed additives at Week 4

Parameters	Control	OXYT	GRO-UP	MOS (500ppm)	MOS (1000ppm)	SEM
PCV (%)	30.00	26.33	28.33	28.00	26.33	0.56
Hb (g/dl)	9.77	8.57	9.30	8.97	8.57	0.18
RBC (×10 ¹² /l)	1.67	1.44	1.55	1.52	1.43	0.04
MCH (pg)	58.44	59.69	60.15	59.22	61.26	0.71
MCHC (g/dl)	32.54	32.54	31.67	32.06	33.26	0.28
MCV(fl)	17.95	18.34	18.30	18.46	18.44	0.17
WBC (×10 ³ /l)	8.03	6.27	7.13	6.90	7.30	0.35
Heterophils (%)	37.00	37.33	42.00	35.67	42.00	1.33
Lymphocytes (%)	56.67	56.00	52.33	55.00	53.33	1.43
Monocytes (%)	0.33	1.00	0.67	1.00	1.33	0.17
Eosinophils (%)	3.00	3.33	3.00	3.00	3.33	0.09
Basophils (%)	0.00	0.00	0.00	0.00	0.00	0.00

Means on the same row having different superscripts are significantly different (P<0.05). Control (no additive); OXYT: Oxytetracycline (antibiotics); GRO-UP (probiotics); MOS: Mannan Oligosaccharide (prebiotics)

Table 4 - Serum biochemical parameters of broiler chickens fed diets containing feed additives at Week 4

Parameters	Control	OXYT	GRO-UP	MOS (500ppm)	MOS (1000ppm)	SEM
Glucose (mg/dl)	146.67	149.33	151.33	147.67	152.33	1.14
Total protein (g/dl)	26.34	27.33	27.67	27.00	27.00	0.29
Serum albumin (g/dl)	12.67	13.33	13.00	12.67	13.00	0.18
Serum globulin (g/dl)	13.67	14.00	14.67	14.33	14.00	0.19
Uric acid (mg/dl)	5.07	5.10	5.47	5.03	5.49	0.10
Cholesterol (mg/dl)	97.67	90.00	114.00	99.00	104.33	4.40
Creatinine (mg/dl)	0.97	0.93	0.87	0.97	0.97	0.34

Means on the same row having different superscripts are significantly different (P<0.05). Control (no additive); OXYT: Oxytetracycline (antibiotics); GRO-UP (probiotics); MOS: Mannan Oligosaccharide (prebiotics)

Table 5 - Haematological parameters of broiler chickens fed diets containing feed additives at Week 8

Parameters	Control	OXYT	GRO-UP	MOS (500ppm)	MOS (1000ppm)	SEM
PCV (%)	29.67	29.33	30.00	29.33	30.33	0.32
Hb (g/dl)	9.70	9.87	9.70	9.77	10.00	0.10
RBC ($\times 10^{12}/l$)	2.48	2.45	2.51	2.46	2.54	0.32
MCH (pg)	29.19	40.28	38.73	39.76	39.44	2.24
MCHC (g/dl)	32.73	33.66	32.01	33.31	32.98	0.31
MCV(fl)	9.15	12.17	11.97	11.93	11.97	0.55
WBC ($\times 10^3/l$)	6.90	6.40	5.67	6.50	6.77	0.40
Heterophils (%)	46.33 ^c	50.67 ^a	50.00 ^{ab}	47.00 ^{bc}	50.00 ^{ab}	0.59
Lymphocytes (%)	50.33	49.33	47.00	48.00	46.00	0.72
Monocytes (%)	0.00	0.00	0.33	0.00	0.33	0.09
Eosinophils (%)	3.00 ^b	4.33 ^{ab}	5.33 ^a	4.33 ^{ab}	4.67 ^{ab}	0.30
Basophils (%)	0.00	0.00	0.00	0.00	0.00	0.00

Means on the same row having different superscripts are significantly different (P<0.05). Control (no additive); OXYT: Oxytetracycline (antibiotics); GRO-UP (probiotics); MOS: Mannan Oligosaccharide (prebiotics)

Table 6 - Serum biochemical parameters of broiler chickens fed diets containing feed additives at Week 8

Parameters	Control	OXYT	GRO-UP	MOS (500ppm)	MOS (1000ppm)	SEM
Glucose (mg/dl)	147.00	149.00	148.33	154.67	147.33	1.49
Total protein (g/dl)	26.67	25.33	27.00	27.33	27.67	0.35
Serum albumin (g/dl)	12.00	12.33	13.00	13.00	13.00	0.19
Serum globulin (g/dl)	14.67 ^a	13.00 ^b	14.00 ^{ab}	14.33 ^{ab}	14.67 ^a	0.24
Uric acid (mg/dl)	5.00	5.07	5.17	5.27	5.17	0.63

Means on the same row having different superscripts are significantly different (P<0.05). Control (no additive); OXYT: Oxytetracycline (antibiotics); GRO-UP (probiotics); MOS: Mannan Oligosaccharide (prebiotics)

DISCUSSION

The PCV values obtained in the present study were within the normal range (Ridell, 2011) but lower than 35.9 % reported for chickens in Nigeria (Oyewale and Ajibade, 1990). Under normal conditions, blood composition is reasonably constant for any particular species with changes falling within fairly narrow limits (Banerjee et al., 2002). The RBC counts and PCV are known to be mostly affected by dietary treatment (Banerjee et al., 2002). The results obtained for Hb follow the same pattern with that of PCV with values for birds fed control diets reducing while others increased at the finishing phase. The Hb values obtained were close to the average values of 10.27 g/dl. This showed that probiotic based diets are nutritionally adequate to meet the protein needs of the birds since the haemoglobin concentrations decreased in animals on low protein intake and in parasitic infection of liver damage (Lindsay, 1997). The MCH reduced at the 8th week of the experiment for all groups. Birds fed diets supplemented with feed additives had MCH values within the normal range of 33.00-47.00 pg (Bounous and Stedman, 2000) while birds on control diet had MCH value below the normal range. This result suggests that the blood of the birds had an appreciable oxygen-carrying capacity which showed that nutrient transport by the blood was not impaired by feeding diets containing prebiotics. WBC plays a prominent role in disease resistance especially with respect to generation of antibodies and the process of phagocytosis. WBC values for birds fed OXYT supplemented diets increased while others reduced at finishing phase with birds fed on GRO-UP supplemented diets having the lowest value. An elevated value of WBC could be an indication of birds reacting to one or more

factors in the feed (Oduguwa, 2006). At the finisher phase, values of 46.33-50.67% for heterophils and 3.00-5.33% for eosinophils aligns with the normal range (Ridell, 2011). This may be an indication of positive effect of additives exerted in birds to suppress any effect of antibodies at the finisher phase. Heterophils and eosinophils are granulocytes of the WBC. Heterophils plays critical role in immune response. Eosinophils destroy parasites and also help to modulate inflammatory responses (Britannica, 2013). Lymphocytes and monocytes did not follow any trend even though the values were not significant. Glucose values in this study were lower than the values obtained by Priya and Gomathy (2008) who observed a mean value of 160.92 mg/dl. Serum total protein consists of albumin and globulin; a change in nutritional status and malnutrition is often revealed in total protein values (Allison, 1995). The increased serum protein, albumin and glucose and decreased serum activity observed in broilers fed probiotics and prebiotics were due to improvement in protein synthesis, carbohydrate and lipid metabolism (Rosa et al., 2001). Serum globulin was lowest in birds fed OXYT supplemented diets at the finisher phase. This may suggest poor immune response and insufficient antibody production in the birds. The serum cholesterol in this study did not agree with the report of Mohan et al. (1996). Uric acid recorded high values at starter phase and lower values at finisher phase which is in line with the finding of Szabo et al. (2005) who reported a direct relationship between the amounts of ingested protein and serum uric acid. Uric acid gives an indication of the quality of protein fed and high levels in the serum utilization of protein.

CONCLUSION

The inclusion of prebiotics and probiotics in the diets of broiler chickens elicited no adverse effects on the haematological and serum biochemical parameters, thus, they can be used as replacement for antibiotics.

Competing interests

The authors have declared that no competing interest exists.

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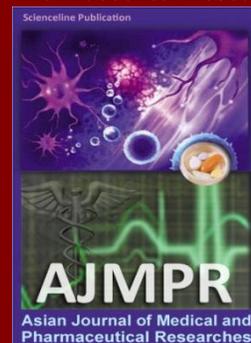
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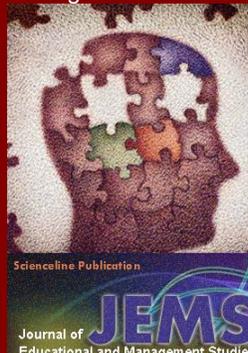
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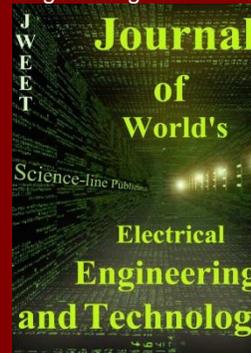
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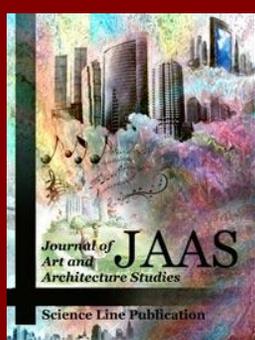
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