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<p>pii: S222877011400010-4</p> <p>Retrospective study of antimicrobial residues and resistance in swine in Aba Abia state Nigeria</p> 	<p style="text-align: center;">Original Research, D10 Nwiyi, P. <i>Online J. Anim. Feed Res.</i>, 4(3): 42-45, 2014.</p> <p>ABSTRACT: Antimicrobials are used by livestock farmers to prevent and control infection. Antimicrobials are also included at sub-therapeutic doses in animal feed as growth promoters and to improve feed efficiency in intensive farming. The aim of this study was to evaluate the antimicrobial residues and resistance that could arise due to antimicrobial use in swine. The study was carried out between September 10th and December 10th 2013 in some selected swine farms in Ogbor Hill water side in Aba, Abia state. The study involved visiting the various farms, evaluating the records of previous treatment. Also the state zonal veterinary clinics visited and record of farms was collected for analysis. From the result obtained, in raining season in a given year, the frequency of tetracycline usage recorded 83.3%, penicillin recorded 75.0%, while sulfonamide recorded 25.0%. Tylosin and ivermox were the least and recorded 8.4% usage each. The swine treatment was done by the farmers hence there was consistent over-dosage of antimicrobials to the pigs as the manufacture's guide was not complied with. The report from the records showed that some of the pigs were slaughtered and sold in the market at any time without recourse to drug with-draw. This result could be one of the responsible reasons for antimicrobial residues and resistance in swine and indeed livestock.</p> <p>Keywords: Antimicrobial residues, Swine, Resistance, Abia state.</p>	<div style="text-align: center;">   </div>
<p>pii: S222877011400011-4</p> <p>The structures of breeding dairy cattle in the Sahara: The status in the M'zab valley (Southern Algerian Shara)</p> 	<p style="text-align: center;">Original Research, D11 Bensaha H., Arbouche F. <i>Online J. Anim. Feed Res.</i>, 4(3): 46-50, 2014.</p> <p>ABSTRACT: The dairy milk sector is a strategic sector in the Algerian agriculture with 2.39 billion liters in 2009, 2.7 billion in 2010 and 2.93 billion in 2011. The milk collection reached 572 million liters in 2011. The enthusiasm for milk production in the M'zab valley led to the creation of many cattle farms. The objective of this study is to investigate the dairy cattle infrastructures and its impacts on milk production. The conducted through a questionnaire over 53 farms representing 57.7% of the exploitations in the M'zab valley. The results show that 25% of farms have mechanical means for milking (milking trolley) and that 92% of buildings are equipped with troughs in a poor condition and do not meet accepted standards. All farms surveyed practice stalls. 32.92% of the buildings are designed with unsuited materials (tin, zinc, shrubs trunks, etc.), 49.88% are designed with a blend of modern (blocks) and traditional (Reeds, Diss) materials, and 17.20% are built with hard poles, walls in cement mortar and asbestos cement roof. The sanitary and hygienic appearance is a function of investments, which are supported by state subsidies in the context of the promotion of the dairy industry. This investigation gave rise to a set of proposals for the upgrading and modernization of the dairy cattle infrastructures, which meet specific climatic conditions, in order to improve the quality and the quantity of milk production in the region of the M'zab Valley with specific climatic conditions.</p> <p>Keywords: Dairy cattle exploitation, Equipment, Livestock building, Hygiene, Saharan region, Valley of M'zab</p>	<div style="text-align: center;">   </div>
<p>pii: S222877011400012-4</p> <p>The growth and reproductive performance of different breeds of rabbits kept under warm and humid environments in Ghana</p>	<p style="text-align: center;">Original Research, D12 Apori SO, Hagan JK and Osei D. <i>Online J. Anim. Feed Res.</i>, 4(3): 51-59, 2014.</p> <p>ABSTRACT: The potential of rabbit production in solving the problem of inadequate animal protein supply cannot be overemphasized. A study was therefore conducted to assess the growth and reproductive performances on 488 bunnies and 87 does of Blue Vienna and 474 bunnies and 80 does of Chinchilla rabbits reared under hot and humid environment in Ghana. The reproductive performance of the two breeds, in terms of litter size at birth and weaning, litter weight at birth and weaning, kindling interval, age at first kindling and gestation length as influenced by season of kindling (rainy and dry), year of kindling (2005-2012) and parity (first to sixth and over) were determined. The overall least square means for litter size at birth, bunny weight at birth, litter size at weaning, bunny weight at weaning, gestation length, kindling</p>	<div style="text-align: center;">   </div>



interval, age at first kindling, pre-weaning growth rate and post-weaning growth rate of Blue Vienna were 5.6±0.1, 51.3±0.3 g, 4.4±0.1, 601.5±0.9 g, 30.0±0.1 days, 94.7±0.4 days, 159.7±0.4 days, 13.1±0.1 g/day and 15.4±0.1 g/day respectively. Those of Chinchilla were 5.9±0.1, 54.2±0.3 g, 4.8±0.1, 601.9±0.9 g, 30.1±0.1 days, 94.6±0.3 days, 159.8±0.2 days, 13.0±0.1 g/day and 15.3±0.2 g/day respectively. Year of kindling had significant (P<0.05) effect on litter size at weaning, litter weight at kindling, gestation length, age at first kindling, post weaning growth rate, body weight at 14 weeks and mortality in both breeds. Season of birth on the other hand had significant effect (P<0.05) on litter weight at birth, gestation length and mortality in both breeds. The number of times the doe has kindled (parity) significantly (P<0.05) influenced all the parameters apart from kindling interval, gestation length, age at first kindling and pre-weaning growth rate performance of both breeds. The growth performances of the breeds studied were indications of their potential of being developed in future into meat types.

Keywords: Chinchilla, Blue Vienna, Growth, Reproduction, Litter Size, Bunnies.

pii: S222877011400013-4

Effective methods for appropriate diagnosis of brucellosis in humans and animals (review article)



Review Article, D13
Molavi MA., Sajjadi HS., Nejatizade AA.
Online J. Anim. Feed Res., 4(3): 60-66, 2014.

ABSTRACT: Brucellosis is one of the most common diseases among human that identification and control of disease transmission methods can promote public health. Clinical signs alone are not sufficient for brucellosis diagnosis. Hence, a sensitive, specific, rapid and inexpensive method is required. Early and appropriate diagnosis of this disease is effective in improving public health as well as disease control and eradication. Several serological tests for probable diagnosis of Brucella infection were used in evaluation of antibodies against Brucella. Using new methods such as Elisa has higher sensitivity and specificity than standard SAT test and complement fixation which can show both G and M immunoglobulins. It is also suitable for examining certain class of immunoglobulin. Research and studies have shown that ELISA is a complete method for in vitro detection of chronic disease, especially when other tests results are negative. In addition to this method, all unique and specific immunoglobulin in tested serum appear with high speed and accuracy. Another diagnostic method is PCR, which has higher sensitivity and specificity in comparison with serologic methods for diagnosis of human brucellosis. PCR shows similar sensitivity as 16srRNA using L7/L12 gene. It can be used in diagnosis of human brucellosis. Another diagnostic method is identification of different forms of IL-10 gene, which is a cytokine. It inactivates macrophages and infects the susceptible subject with brucellosis. Therefore, identification of different forms of IL-10 gene is considered as effective method for diagnosis of the disease. It's recommended to use this new and effective method because many of these methods can overcome limitations of traditional methods.

Keywords: Brucellosis, Diagnosis, Human, Animal



pii: S222877011400014-4

Influence of Weaning Age on the Organoleptic Properties of Pork



Original Research, D14
Moreki J.C., Perfect L., Machete J.B., Montsho T., Ganelang B. and Sello N.
Online J. Anim. Feed Res., 4(3): 67-73, 2014.

ABSTRACT: A trial was conducted to compare organoleptic properties of piglets weaned at 21, 28 and 35 days of age and slaughtered at 70 kg target body weight. A total of 24 pigs (Landrace x Large white x Topigs x Topigs cross) which were weaned at three weaning ages were randomly selected and slaughtered upon reaching target body weight. Each treatment comprised three replicates of two animals each. Out of this number, 18 carcasses were selected for organoleptic evaluation. Piglets were provided with creep diet from 10 to 35 days, a weaner diet from 36 to 70 days of age and pig grower diet from 71 days to target slaughter weight. Thereafter, pigs were sacrificed and their carcasses chilled for 20 hours at 5 0C before cuts were removed. The four meat cuts (the pork chop, chuck, Top sirloin and pork leg) were removed, cooked and tested for organoleptic evaluation, i.e., texture, tenderness, juiciness, appearance and flavour. Data on organoleptic properties were analysed using frequencies and percentages in IBM SPSS statistics for Windows, version 20.0. Results showed that the panelists preferred pork from pigs weaned at 28 days followed by 21 days and 35 days. The appearance of pork was the most preferred characteristic across all weaning periods as it showed high rankings by panelists while juiciness appeared to be the less preferred attribute. These results suggest that weaning age influenced organoleptic properties of pork with 28 days weaning age giving better results.

Keywords: Ad libitum, Organoleptic Properties, Piglets, Pork, Weaning Age



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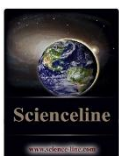
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CONCENTRATION OF NITRATE AND NITRITE IN SOME SELECTED CEREALS SOURCED WITHIN KADUNA STATE, NIGERIA

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ABSTRACT: Concentrations of nitrate and nitrite ions were assessed in maize, millet, guinea corn and wheat obtained in Kaduna State, Nigeria. The results showed that nitrate ion concentration for white maize, red maize, agric millet, normal millet, white guinea corn, red guinea corn, and wheat were 4.5 mg/g, 4.2 mg/g, 15.5 mg/g, 21.3 mg/g, 6.3 mg/g, 4.0 mg/g, and 3.0 mg/g respectively. While the nitrite ion concentration for white maize, red maize, agric millet, normal millet, white guinea corn, red guinea corn, and wheat were 0.035mg/g, 0.030mg/g, 0.074 mg/g, 0.087 mg/g, 0.050 mg/g, 0.050 mg/g and 0.0154 mg/g respectively. The results obtained fall below the WHO's Acceptable Daily Intake (ADI) which is 40-100 mg/g for nitrates, and fall within range for nitrites which is 0.03 to 2.6 mg/g. This means that the nitrate and nitrite contents of maize, guinea corn, millet and wheat grown in Kaduna State may not presently pose a health hazard in the human population.

Keywords: Nitrate, Nitrite, Kaduna State, Cereals, Soils

ORIGINAL ARTICLE
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INTRODUCTION

Nitrates are present naturally in soils, waters, all plant materials and in meats. They are also found in small concentrations (1-40/ $\mu\text{g}/\text{m}^3$) in air as a result of air pollution. Levels in water and cultivated soils, (which normally do not exceed 10mg/L), may be increased by the use of commercial nitrogenous fertilizers and by the return of waters derived from animal husbandry to the soil. Nitrate contents of crops are influenced by the plant species, and environmental factors. In certain crops the level may be very high (1000/mg/Kg or more) (Oztekin, 2002). Nitrate and nitrite are essential nutrients for plants protein synthesis and play a critical role in nitrogen cycle (Parkash et al., 1963). Nitrate is a naturally occurring form of nitrogen. Nitrate is formed from fertilizers, decaying plants, manure and other organic residues. It is also used as a food additive, mainly as a preservative and antimicrobial agent (Walker, 1990).

Due to the increased use of synthetic nitrogen fertilizers and livestock manure in intensive agriculture, vegetables and drinking water may contain higher concentrations of nitrate now than in the past (Santamaria, 2006) Nitrites are formed in nature by the action of nitrifying bacteria as an intermediate stage in the formation of nitrates, but concentrations in plant and water are usually very low. However, microbiological conversion of nitrate to nitrite may occur during the storage of fresh vegetables, particularly at room temperature, when nitrite concentrations may rise to exceptionally high levels (about 3600mg/Kg dry weight) (Oztekin, 2002). Both nitrates and nitrites are widely used in the production and preservation of cured meat products and of some fish. Such uses, which are controlled by law in many countries, are considered vital for the prevention of botulism caused by the growth of the toxin-producing strains of *Clostridium botulinum* that are sometimes present in raw meats and that may persist in cooked meats (Binstok, 1996).

Nitrates occur naturally in food crops and plants. The Nitrate levels commonly present in food is not toxic to humans but serve as the reservoir for conversion to nitrite by the intestinal flora. Nitrate per se has a relatively low toxicity. Nitrite is the compound that is of more health concern (Cassens, 1996).

The factors responsible for nitrate accumulation in plants are mainly nutritional, environmental and physiological. Nitrogen fertilization and light intensity have been identified as the major factors that influence the nitrate content in cereals (Cantliffe, 1973b). Diurnal changes in light intensity lead to a diurnal pattern of nitrate accumulation in plants. Similarly, nutrients such as chloride, calcium, potassium, sulphate and phosphorus are also involved in nitrate accumulation process in plants. Nitrate content varies in various parts of a plant (Santamaria et al., 1999, Anjana et al., 2006). Both nitrates and nitrites are widely used in the production and preservation of cured meat products and of some fish (Uwah et al., 2009). Human exposure to nitrate and nitrite result primarily from dietary ingestion particularly from food crops, vegetables, meat and water. The average adult intake from food

has been estimated to be 40-100/mg for nitrate and 0.3-2.6mg for nitrite. Exposure estimates indicate that for more 99% of the adult populace, only 1-3% of nitrate and nitrite intake comes from drinking water, mainly in areas of notable contamination (Fann and Steinberg, 1996).

The high demand for cereals by the increasing human population and their use for compounding livestock feed has necessitated the assessment of nitrate and nitrite in cereals. The levels of these compounds in food should be of much concern considering their toxicological health implications on humans and animals.

The aim of this study is to obtain information as to whether the level of nitrate and nitrite in the selected cereals is within the laid down standards by the World Health Organization (WHO).

MATERIAL AND METHODS

Sample Collection

A total of 8 samples, 2 samples per cereal, (Maize, Guinea corn, Millet and Wheat) were randomly collected from farm lands in Kaduna State. The cereals were crushed by a mortar and pestle and cereals flours were put in nylon bags with shackles to prevent contamination with air. Therefore the effect of humidity on nitrate/nitrite content of the samples was reduced.

Extraction Procedure

For each sample, 1g of grounded cereal was used for the analysis; 4ml of hot water was added on the sample and blended for 5 min in a blender. The mixture was heated to 75°C for the prevention of ascorbic acid interference and solution was transferred to a volumetric flask and 5ml hot water and 1.2 ml Sodium hydroxide (2% w/v in water) was added and blended again for another 10 mins and 1ml of Zinc hydroxide (7.2% w/v in water) was added and the mixture was agitated for 5min. Thereafter 0.5 ml Sodium hydroxide was then added and the mixture was blended for 5mins. Distilled water (8.3 ml) was added and mixed for 5min to obtain a final volume of 20 ml after which the mixture was then filtered using filter paper (Whatman No. 1) until the filtrate is completely clear (Pinar et al., 2009).

Determination of Nitrate (NO₃⁻) and Nitrite (NO₂⁻) concentrations in the cereals samples

The determination of Nitrate in each of the cereal sample solutions was performed using Spectrophotometer at a wavelength of 543nm. The result will be obtained as Nitrate-Nitrogen (NO₃⁻-N) and converted to ppm Nitrate (NO₃⁻) by multiplying by 4.4 (conversion factor). The concentration levels of Nitrate (µg/g) was calculated from NO₃⁻ (µg/g) = C x V/M, where; C is the concentration of NO₃⁻ in the sample (ppm), V is the total volume of the sample solution (100ml), and M is the weight of the sample (1g). Nitrite levels in the sample solutions were similarly determined except that in this case, different reagents were used. The programmer number for Nitrite was 67 Nitrite-N and the reaction period was five minutes as against ten minutes in the case of Nitrate. Nitrite-Nitrogen (NO₂⁻-N) was converted to ppm Nitrite (NO₂⁻) by multiplying by 3.3 (conversion factor). The concentration levels of Nitrite (µg/g) in the sample were calculated from:

$$\text{NO}_2(\mu\text{g/g}) = C \times V/M$$

Where C is the concentration of NO₂⁻ in the sample (ppm), V is the total volume of the sample solution (100ml) and M is the weight of the sample (1g). Both nitrate and nitrite levels in all the samples were determined as described by (Uwah et al., 2009).

RESULTS AND DISCUSSION

Table 1 shows the concentration of nitrate and nitrite in some selected cereals obtained in Kaduna state, Nigeria. The results from this study show that all the cereals analyzed contain detectable amount of nitrate/nitrite ion. The result indicated higher levels of nitrate in the range of 3.0 mg/g in wheat to 21.3 mg/g normal millet. The trend of nitrate variation was in the order: Normal millet > Agric millet > White guinea corn > White maize > Red maize > Red guinea corn > Wheat. The Joint FAO/WHO Expert Committee on Food Additives (JECFA, 2003) and Scientific Committee on Food (SCF) have proposed an acceptable daily intake (ADI) for nitrite of 0.3-2.6/mg/g in humans. Ezeagu (2006), conducted a similar work on rice, maize, and cowpea with maize varieties having a higher value (1000/mg/kg) which is above 500/mg/kg nitrate limit recommended by WHO/FAO (WHO, 1973). Similarly, Gilbert et al. (1946) reported high concentrations of nitrates in air-dried samples, ranging from 12200.0/mg/kg in millet to 30000.0/mg/kg in oats. However, they speculated that these high figures are artifact and probably reflect contamination during drying rather than actual levels.

The concentrations of nitrite in the cereals were generally low compared to the nitrate contents. The concentrations of nitrite in the cereals analyzed were in the range of 0.030 mg/g in red maize, to 0.154 mg/g in wheat. The trend of nitrite variation in the cereals was also in the order: wheat > normal millet > agric millet > red and white guinea corn > white maize > red maize. These levels are low and within the limits of the recommended normal acceptable daily intake (ADI) level (0.1/mg/kg body weight). Dietary exposure to nitrite is normally very low. Exceptionally, higher levels may result from microbial reduction of nitrate in hygienically poor quality well water or in foods rich in nitrate stored under inappropriate conditions (Heisler et al., 1974; Ezeagu, 1996).



Table 1 - The concentration (mg/g) of NO₃⁻ and NO₂⁻ ions from cereal varieties

Cereals	NO ₃ ⁻	NO ₂ ⁻
White maize	4.5	0.035
Red maize	4.2	0.030
Agric Millet	15.5	0.074
Normal Millet	21.3	0.087
Guinea Corn White	6.3	0.050
Guinea Corn Red	4.0	0.050
Wheat	3.0	0.154

Generally, low levels of nitrate/nitrite are reported for grains and seeds (McNamara et al., 1971). Differences in species, strain and agro-technical operations as well as environmental pollution could account for the wide differences of nitrate and nitrite concentrations in this study vis-à-vis other related studies. It could also be as a result of soil type of the farmland and agricultural management practices.

Nitrates are soluble in water; they are easily washed off fields into rivers where they cause water pollution problems. The rate of absorption of dissolved nitrates from underground and surface water differs from plant to plant, and specie to specie, and this could be responsible for the difference in concentration of nitrate/nitrite as obtained from the current study. Also, the application of artificial fertilizers and animal waste is also seen as a major source of nitrates to plant crops. Furthermore, in farmlands where these practices are adopted, there is a possibility of having high levels of nitrate and nitrite ion contents in the plant crops.

The low concentration of nitrate and nitrite reported in this study compared to other related studies could be as a result of differences in geographical location of the cereals, geographical location of farmlands also determines the levels of nitrate in plant. Farmlands situated in industrialized areas are prone to pollution by the release of chemicals into the farmlands leading to contamination of plant crops and the wash off of nitrate contents in the sub-soils, whereas those farmlands situated in non-industrialized areas are free from contamination and may have higher nitrate contents. In natural system, nitrates in the soil are lost by denitrification, erosion, leaching and replaced by fixation and the application of either inorganic or organic manure. Human intrusion in the nitrogen cycle can result in fewer nitrates being cycled, or in an overload of the system. For example, the cultivation of croplands, harvesting of crops and cutting of forests are known cause of steady decline in nitrate content of the soil. The consumption of supplement sources of nitrate in diet (e.g. legumes, vegetables, and some water sources) and other classes of food along with cereals could allow the Acceptable Daily Intake (ADI) to increase above the set values recommended by world health organisation (WHO).

CONCLUSION

Nitrate levels present in the cereals analysed falls below the acceptable standards of 40-100/mg/g, while nitrite levels fall within the acceptable standard of 0.03-2.6/mg/g, which is below the toxicity level. This means that the nitrate and nitrite content of maize, millet, guinea corn and wheat grown within Kaduna State may not presently, pose a health hazard in the humans' population.

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RETROSPECTIVE STUDY OF ANTIMICROBIAL RESIDUES AND RESISTANCE IN SWINE IN ABA ABIA STATE, NIGERIA

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ABSTRACT: Antimicrobials are used by livestock farmers to prevent and control infection. Antimicrobials are also included at sub-therapeutic doses in animal feed as growth promoters and to improve feed efficiency in intensive farming. The aim of this study was to evaluate the antimicrobial residues and resistance that could arise due to antimicrobial use in swine. The study was carried out between September 10th and December 10th 2013 in some selected swine farms in Ogbor Hill water side in Aba, Abia state. The study involved visiting the various farms, evaluating the records of previous treatment. Also the state zonal veterinary clinics visited and record of farms was collected for analysis. From the result obtained, in raining season in a given year, the frequency of tetracycline usage recorded 83.3%, penicillin recorded 75.0%, while sulfonamide recorded 25.0%. Tylosin and ivermox were the least and recorded 8.4% usage each. The swine treatment was done by the farmers hence there was consistent over-dosage of antimicrobials to the pigs as the manufacture's guide was not complied with. The report from the records showed that some of the pigs were slaughtered and sold in the market at any time without recourse to drug with-draw. This result could be one of the responsible reasons for antimicrobial residues and resistance in swine and indeed livestock.

Keywords: Antimicrobial residues, Swine, Resistance, Abia state.

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INTRODUCTION

Antimicrobials are any substances including synthetic compounds which destroys microbes (Guardabasse and Courvalin, 2006). In piggery production in most countries of the world, antimicrobials are administered both for prophylaxis and therapeutic purposes. Some antimicrobials are used to prevent disease in fairly healthy animals more especially during perceived increase susceptibility period. Mastitis is a common disease suffered by lactating pigs and treatment is by the use of antimicrobial. Besides, lactating pigs, antimicrobial are also use for treating other infection disease. The use of antimicrobials to treat food animals has the potentials to affect human health via two mechanisms: increasing the risk of antimicrobial residues influencing the selection of antimicrobial resistant food borne pathogens (Yan and Gilbert 2004). The risk of antimicrobial residues is well known and there is increasing concern about the impact of antimicrobial usage in food animals on the development of antimicrobial resistance. Antimicrobial agents disrupt bacterial processes needed for growth. Compounds that inhibit bacterial growth are described as bacteriostatic while those that kill the bacteria are termed bactericidal. Antimicrobial agents can be bacteriostatic when they reach the minimum inhibitory concentration (MIC) but become bactericidal when they reach a higher concentration called the minimum bactericidal concentration (MBC). If the MIC and the MBC are distinctly separated, the agent is considered bacteriostatic. If the MIC is close to the MBC, the compound is said to be bactericidal (Prescott, 2000).

Bacterial resistance can be intrinsic or acquired. Acquired resistance occurs when a previously susceptible bacterium becomes resistant via mutation (Hall and Collis, 1995). There are several mechanisms of resistance that had been described. Some bacteria contain enzymes that inactivate antibiotics. The most well-known example is β -lactamase. These enzymes inactivate β -lactam antibiotics by clearing the β -lactam rings. Some bacterial develop resistance by preventing the antibiotic from entering the bacterial cell or by increasing the removal of the drug out of the cell.

Recently, Pol and Ruegg (2006) developed a method to quantify antimicrobials usage and treatment practices. The need for antimicrobial withdrawal from animals before slaughter or use as milk is very important since scientific experiment provide data that shows how long a drug is present in the body of animal and what the animal body does to the drug (Graham et al., 2009). The Food and Drug Administration (FDA) of United States Department of Agriculture and Centers for Disease Control and Prevention affirms that there is a definitive link



between the routine, non therapeutic use of antibiotics in food animal production and the challenge of antimicrobial resistance in humans (Gilchrist et al., 2006). Some researcher reported use in livestock production was a factor in the high prevalence of antibiotic resistant bacteria in Korea (Pereira and Siqueira–Junior, 1995). The relatively high usage of antimicrobials in livestock production had lead to the banned use of antibiotics as growth promoters in livestock (Woo-Joo and Seung, 1998). The objective of the study is to evaluate antimicrobial usage in swine by pig farmers and possible resistance.

Concern for Antibiotic Resistance:

Of late, there has been increased concern about the use of anti – microbial in animals contributing to the rise in antibiotic resistant infections in humans. The use of antimicrobials has been linked to the rise of resistance in every drug and species where it has been studied, including humans and livestock. The role of antimicrobial use in food animals and resistant infection is gradually on the rise. The use of antimicrobials in various forms is widespread throughout the animal industry, and is presented as key to preventing animal suffering and economic loss.

Antimicrobial Susceptibility Tests;

Antimicrobial susceptibility tests measure the ability of an antimicrobial agent to inhibit bacterial growth invitro and are performed using methods that are based on diffusion (Walker, 2000). The agar disc diffusion is one of the most common methods and is referred to as the Kirby – Bauer method. A standardized suspension of bacteria is streaked over a Muller – Hinton ager plate and antimicrobial impregnated discs are applied. During overnight incubation, a gradient of antimicrobial concentration is closest to the disc and progressively lower concentrations occur as distance from the disc increase. If the bacteria are susceptible to the antimicrobial tested, a distinct inhibition zone will be observed. If the bacteria are resistant to the antimicrobial, bacterial growth will be observed close to the antimicrobial disc. The diameter of each inhibition zone is recorded and the outcome is interpreted for each antimicrobial using standards based on the size of the zone of inhibition (Walker, 2000).

MATERIALS AND METHODS

Five pig farms were randomly selected out of over seventeen different pig farm all located within and around Ogbor Hill River at Aba Abia State in Nigeria. The choice of the farm was due to the good record keeping Obtained from the State Zonal veterinary clinics Aba. To confirm the report in the document, visit was paid to the five farms and the total number of pigs in each farm was recorded. The records of the antimicrobial used, the various dosages use, the season of usage and frequency of usage were all taken for analysis. From the available documented case file both by the veterinarian and more especially by the individual pig farmers.

RESULTS AND DISCUSSION

Antimicrobial are used by the pig farmers for therapeutic and prophylaxis control of the infection. Table 1 shows a class representation of a pig farm. The ratio of the sow to the boar was best presented by ventures farm and all the farms except new-hope which has no piglet as at the time of visit

Antimicrobial were used throughout the year, however the number of antimicrobial used during the rain season as represented in Table 2. Global farms and Orcharkk farms recorded the highest antibiotic usage per year than all others. Tetracycline recorded 83.3% usage annually penicillin 75.0%, while ivermox and tylosin recorded 8.4% each which was the lowest as shown in Table 3. The relative humidity and water logged site of the pen could contribute to the multiplication of microorganism.

The use of antimicrobial by the farmers was regardless of the manufactures recommendation similar result was reported (Erskine et al., 2002). The treatment was carried out by the farmers. The veterinarians are consulted when the situation of the animal is in deplorable condition. However, there were farmers who adhere strictly to the manufactures guide. In Table 4, dosages of sulfonamide and Tetracycline given were 2-2.5ml/10kg and 2.5-3ml/10kg compared to the recommendation dosage of 1ml/10kg and 1-1.5mi/10kg which is of higher range. Similar result was reported (Kirk et al., 2005). The prescription close to the manufactures guide was ivermox and ampicillin which was 1ml/10kg and recorded usage was 1-2ml/10kg. The rampant use of antimicrobials in high doses lead to high quantities of residues released when the animals are slaughtered without any withdraw thereby posing health hazard to humans on consumption of the meat (Makovec and Ruegg, 2003) reported similar conclusion in a larger study.

Table 1 - Average Farm Size of Five Selected Pig Farms Located In Ogbor Hill, Aba

Name of Farms	Total Number of Swine	Sow	Boar	Grower	Piglets
Ventures	210	160	20	11	09
Global	124	54	23	25	22
El-shaddi	67	31	12	13	11
Orchakk	42	18	07	11	06
New-hope	28	14	06	08	00



Table 2 - Average number of different antimicrobial used per year in five farms

Name of Farms	Dry Season	Raining Season	Total/year
Ventures	1	4	5
Global	1	7	8
El-shaddi	1	3	4
Orchakk	2	6	8
New-hope	1	5	6

Table 3 – Frequency of antimicrobial usage per year

Types of Antimicrobials	Frequency of Occurrence	Percentage usage/year
Ampicillin	3	25.0% (3/12x100/1)
Penicillin	9	75.0% (9/12x100/1)
Streptomycin	2	16.6% (2/12x100/1)
Tetracycline	10	83.3% (10/12x100/1)
Tylosin	1	8.40% (1/12x100/1)
Sulfonamide	3	25.0% (3/12x100/1)
Ivermox	1	8.40% (1/12x100/1)

Table 4 – Recommended Dosage for Antimicrobial and Dosage used by Swine Farmers

Types of Antimicrobials	Recommended dosage ml/kg	Average Dosage used ml/kg
Ampicillin	1ml/10kg	1-2ml/10kg
Penicillin	1ml/10kg	2.5-3ml/10kg
Streptomycin	1ml/10kg	1-2ml/10kg
Tetracycline	1ml/10kg	2.5-3ml/10kg
Tylosin	1.5ml/20kg	1-2ml/10kg
Sulfonamide	1ml/10kg	2.0-2.5ml/10kg
Ivermox	1ml/10kg	1.5-2ml/10kg

CONCLUSION

The populace will continue to be concerned about the development and transfer of antimicrobial resistance in livestock and veterinarians will need to be responsive to the occasion. The amount of exposure to some antimicrobial has been linked to increase resistance. There is the need to educate swine farmers on the importance of consulting the veterinarians on time for expatriate advice and treatment in the event of outbreak of disease can not be over-emphasis. Antimicrobial sensitivity test should be recommended before antimicrobial could be administered to animals.

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THE STRUCTURES OF BREEDING DAIRY CATTLE IN THE SAHARA: THE STATUS IN THE M'ZAB VALLEY (SOUTHERN ALGERIAN SHARA)

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ABSTRACT: The dairy milk sector is a strategic sector in the Algerian agriculture with 2.39 billion liters in 2009, 2.7 billion in 2010 and 2.93 billion in 2011. The milk collection reached 572 million liters in 2011. The enthusiasm for milk production in the M'zab valley led to the creation of many cattle farms. The objective of this study is to investigate the dairy cattle infrastructures and its impacts on milk production. The conducted through a questionnaire over 53 farms representing 57.7% of the exploitations in the M'zab valley. The results show that 25% of farms have mechanical means for milking (milking trolley) and that 92% of buildings are equipped with troughs in a poor condition and do not meet accepted standards. All farms surveyed practice stalls. 32.92% of the buildings are designed with unsuited materials (tin, zinc, shrubs trunks, etc.), 49.88% are designed with a blend of modern (blocks) and traditional (Reeds, Diss) materials, and 17.20% are built with hard poles, walls in cement mortar and asbestos cement roof. The sanitary and hygienic appearance is a function of investments, which are supported by state subsidies in the context of the promotion of the dairy industry. This investigation gave rise to a set of proposals for the upgrading and modernization of the dairy cattle infrastructures, which meet specific climatic conditions, in order to improve the quality and the quantity of milk production in the region of the M'zab Valley with specific climatic conditions.

Keywords: Dairy cattle exploitation, Equipment, Livestock building, Hygiene, Saharan region, Valley of M'zab

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INTRODUCTION

The dairy industry is a strategic sector of the Algerian agriculture, with 2.39 billion liters in 2009, 2.7 billion in 2010 and 2.93 billion in 2011 and a collection of milk from 572 million liters in 2011 (MADR, 2012). The craze for milk production in the valley of Mzab led to the creation of many cattle farms.

In dairy cattle farms, worldwide, it was found that the architecture of the barn plays a major functional role in dairy production, including providing comfort in the work of the farmer and avoiding stress livestock. Indeed, during the construction of a building, the question of its size is related to the size of the herd and the requirements of the race in terms of housing. Building a barn is a big investment for the farmer, but in view of its life, the constraints of the operating cash will fade as and when capital depreciation and due to the rapid return on investment.

In general, it is recognized that the success of breeding dairy cattle is based on the mastery of his technique driving through the components that are: feeding, reproduction, health, and modernized building. This allows the emergence of modern farming incorporating the principle rationality and sustainability which, logically, should result in better technical and economic performance because of the increased milk production potential.

Indeed, infrastructure (technical buildings) must conform to accepted standards to ensure the welfare of animals and improving working conditions (atmosphere and organization of work). The low level of technology and lack of knowledge of farmers, highlight management issues (Bouaboub, 2008).

Also, it should be noted that to ensure food quality and animal welfare, the hygiene is particularly important in environmental surfaces of barns (Hanna et al., 2010). All these environmental and organizational constraints are imposed on the cattle industry Maghreb and induce some challenges in terms of meeting the demand for all the operators concerned: government in the foreground, farmers and industrial milk (Srairi, 2008).

The region of the Valley Mzab has received significant funding for the program of development of the dairy cattle industry. However, these effects are not evaluated. Indeed, no study has been conducted to determine the status of dairy cattle in relation to the structures of the livestock in this area.

This study subjected to identify the current state of infrastructure of dairy cattle and analyze different practices and strategies in place to manage these cattle farms, in order to recommend actions for their upgrade to improve the quality and quantity of milk production in the region of the valley of Mzab.



MATERIAL AND METHODS

In the current environment, the available data on cattle and their owners in the Saharan regions are elementary and fragmentary. Given the lack of data on the dairy cattle industry, the livestock and socio-economic surveys are now proving an essential tool to generate a significant amount of information about the industry to better appreciate its performance.

The approach taken in this study is based on survey of producers farmers, agricultural institutions and local authorities. For investigations us a sample of 53 cattle sheds was selected on a population of 101 mother stables (or 57.74% of the statistical community) divided the territories of seven (07) municipalities of the province of Ghardaia. The selection criteria for the basis of this selection are related to milk production potential:

- The size of the farm (heads ≥ 12);
- Speculation owner;
- The high production potential.

Summary data on the geographical distribution of the sample are presented in table 1.

Table 1 - The different farms selected for investigation

Towns	Total number of cattle farms	Number of cattle farms surveyed	Percentage
Guerra	45	20	44.44
Berriane	10	6	60
Zelfana	13	7	53.84
El Atteuf	10	6	60
Bounoura	5	3	60
Daya BenDahoa	11	6	54.54
Ghardaia	7	5	71.42
Total	101	53	57.74

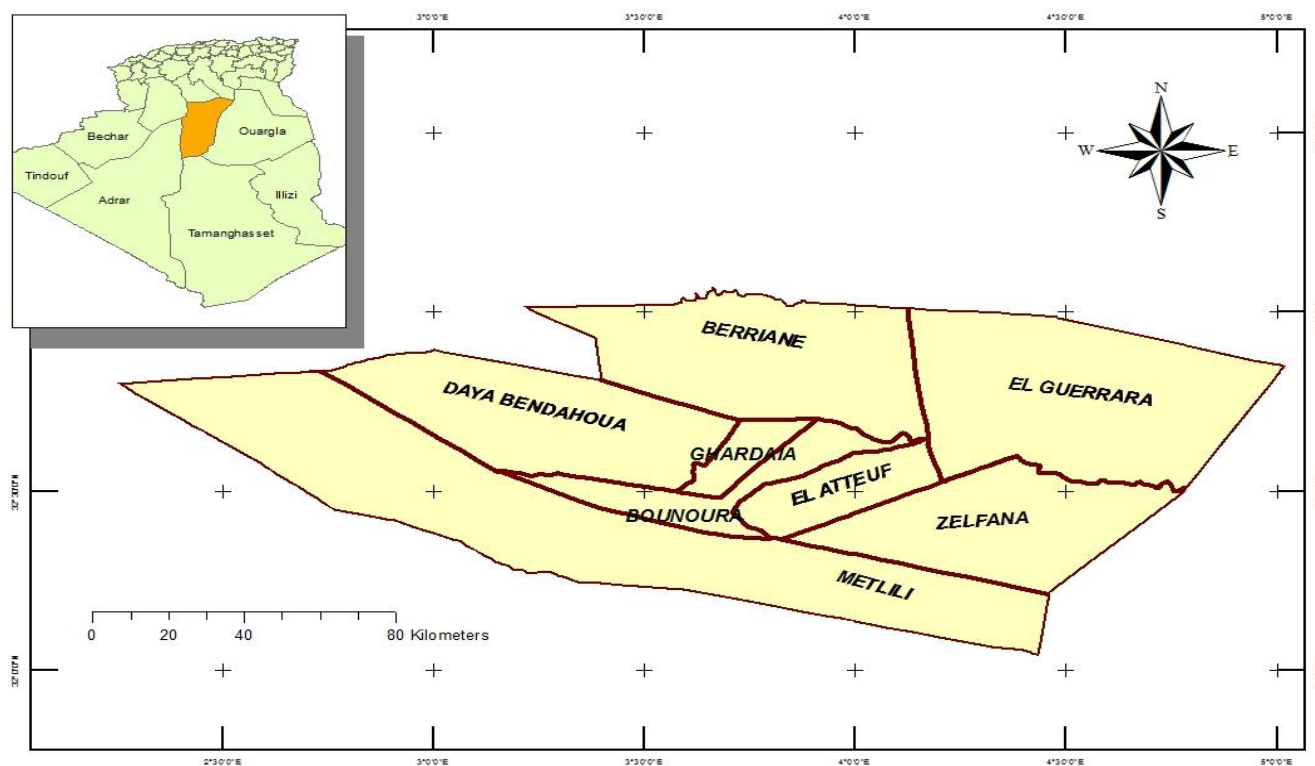


Figure 1. Geolocation of the study area

RESULTS AND DISCUSSION

During the investigation, the following items related to the quality of the buildings have been identified: the model of stable and age of infrastructure, building materials and flooring, the design of the building / rooms, furniture (feeders, drinkers, ..), mechanical means (trolley milking ..) and hygiene aspects.

Stable model and age of infrastructure

The barn is the most important parameter in the system of intensive dairy production. It appears from the survey that all stables visited are free stall. This model seems to be the benchmark in the area of the Valley Mzab.



The most popular is the stables with floor covered with straw. In this hot and dry region (with low humidity), it is possible to keep the animals in a barn sleeping areas composted - with a litter consisting of dried manure.

As noted Charron (1986), this mode provides little comfort barn animals, it causes difficulty calving and heat detection, and has repercussions on the health of the animals, which are generally dirtier.

Regarding the condition of buildings: 32.92% are in poor condition, 49.88% are in average condition and 17.20% are in good conditions (see Figure 02).

Animal load in the building is respected by all farmers. However, a living building for a possible adaptation of the production tool is not intended for farmers.

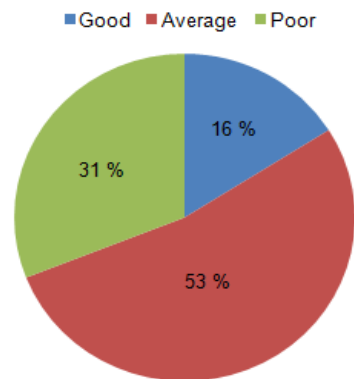


Figure 2. State farms surveyed buildings

Building Materials

The stables have surfaces ranging from 150 to 400 sqm. Most barns are built of granite stone with low ventilation conditions and without the possibility of mechanization to remove manure. They have a metal roof or dry palms to avoid sun exposure to rain. They are surrounded by wire fences and wood, or sometimes galvanized iron pipe.

Almost all the buildings (95%) have a surface composed of sand and are without litter. The few litters found consist of gorse and bracken. Stables have some part of their surface area paved. In the case of the presence of concrete slabs, cattle should not be on the bare concrete floors. Too hard floors can increase the incidence of injuries tail, swollen knees and hock abrasions.

But without a well built with good materials, essential parameter for efficient power management, cleaning and care of cows and calves building, the dairy industry can be a profitable business.

Building Design

Milking: A milking parlor should be comfortable for the milking and animal traits. Trafficking represents 50% of the time devoted to farming (DSA, 2012). The design of the milking parlor must enable efficient work and as comfortable as possible because it directly affects herd health, milk quality and farm profitability. In all farms visited the parlor is present. Its surface varies with the number of cows. On an average area of 3.5 m² it is isolated from the stable. The number of troughs is 5 to 10, constructed based cinderblock. It should be noted, in some stables without a waiting area for cows. It may be a temporary area defined not movable barriers.

Place food storage: All farms have a food premise (mainly concentrate). Most are in good condition (83%) and other (17%) in average condition. The surface of the deposit of food varies from 9 to 20 m². The building material is, in most cases block, but sometimes stone or clay are used.

Stall the bull: A stall consists of 6 m² useful area of straw per cow. The increase in animal density causes a decrease in the use of stalls and crowded hallways. Cubicles bulls differ from one farm to another. In 58% of farms, the bull is in a detached barn on an area of 9 to 16 m². In 37% of farms, it is attached to a corner of the barn. For the remaining operations (5%), the bull is always with cows. The housing conditions of the cows and the lack of space inside the barn, have a negative impact on economic performance, as well as the time and working conditions of farmers. Decrease of the surface, or the number of seats, also increases the risk of infections including breast lesions teat, microbisme.

Location projections: In 87% of the project takes place in the barn bull rarely in the building 13% of the cows. The cow in heat will be placed with the bull in his stall.

Room calving: In 88% of farmers, there being held in isolated rooms. In the rest of the farmers (12%), living calving is absent and there are places put in the stable, then used as a box for calves.

Room newborns: Newborns are collective boxes in all farms (100%). In 75% of farms are small rooms with surfaces ranging from 4 to 6 m² and 25% of cases, there are small semi-paved area surrounded by wooden fences or sheet. These boxes are used as stables for the calves in all farms (100%). This means that newborns are in their stables until the age of eight months on average for the bulls, or 14 months for heifers.

Desk: He noted the lack of an office in the barn to facilitate administrative tasks of the farmer, but also to optimize the management.

Furnisher

Furniture building is represented mainly by feeders and waterers small group.

Feeders

Drinkers are two types: A) Group: They are built either block, with a volume that differs from one farm to another, either based on galvanized sheets in the form of a half-cylinder with an opening of 40 to 60 cm and a length of 1, about 5-2 m. Are also found troughs made half a tube with a diameter / opening of 20 to 30 cm. The water in this type of drinkers tend to be quickly soiled (Kaouche et al., 2012). B) Automatic: modern metal, based



on aluminum, they have the shape of a hemisphere adhered to a tube where the water passes. Found, within the half-spheres, a movable plate which, driven by the stock of the animal, allows the water outlet.

Hygiene practices and prevention

The hygienic behavior includes hygiene of the building, healthy food, healthy watering the animals and maintains the treatment of major diseases encountered in the study area. Poor building design can generate health problems with significant negative economic impact, as well as the work of the farmer.

Health Building: The hygiene of the building is often poorly controlled. Only 12.82% of them have a good state of health, an average 38.46% and 48.71% hygienic condition ill hygiene. As mentioned Kaouche (2012), housing animals are cleaned to a variable operating frequency to another and depending on the season once a day in winter twice in summer, with detergents broad spectrum or only with water. At all farms, with the exception of four veterinary farmers who practice good hygiene, housing never disinfection, milking machines lying around on the ground, the animals receive brushing once or twice during summer and litter is renewed once every 30 to 80 days. In 90% of farms, cleaning is done manually, the organic material is stored a few meters from the building. Only 10% of farmers do mechanically. The manure is stored for sale or used for soil fertilization (organic fertilizer) Only 10% of farmers do mechanically. In 85% of farmers, cleaning is done irregularly during the year, when the thickness of the layer of manure reaches 15 cm. Only in 15% of operators it is frequently every month. The least popular tasks are those cleaning the milking parlor and barns, as well as trimming feet. As noted by Vallet (1981), under such conditions, the role of favorable thermal comfort straws disappears and they can instead become harmful by their microbial pollution that can cause diseases and mastitis podal.

Moreover, in most cases, the notion of building is only theoretical; stables are made of stone walls, no roof.

Healthy food: The concentrated power is distributed in troughs cinderblock. A large food waste is stored in 92% of farmers whose troughs are generally in average condition, especially for a lack of green fodder racks

Hygiene watering: Drinkers are basins block, half-tubes or modern metal troughs, which are not disinfected. In 92% of farms, the water is not renewed beyond the basins and creating a favorable environment for diseases. Among farmers who have automatic waterers (8%), the state of health of watering is good.

Animal Care: The state animal care differs from one farm to another. Herds of group Kharfi and BSIS, have relatively better than animals of other holdings which are often covered with mud and manure state.

Grazing animals can maintain a relatively good hooves, especially among farmers who feed crops under pivot. Washing the udder before milking is done at all breeders.

Prophylactic measures: It is necessary to emphasize the positive aspects of regular veterinary health action (semi-) associated with epidemiological investigations initiated locally to keep all epidemiological evidence to a satisfactory level. The results in visits to breeding establishments performed periodically to sensitize farmers on various aspects of their business: health, herd management, artificial insemination (Bensaha et al., 2012).

98% of farmers bring their animals to the vet in case of illness, others use traditional methods. All farmers (100%) reported having received one to two times a year, visiting the vet agricultural subdivision and have been vaccinated their cows. Vaccination of animals is carried out by the veterinary inspection management of agricultural services. Every six months, the agency made a blood sample and an injection of tuberculin for those who adhere to the program of the NADP and an annual vaccination against rabies and against FMD (Guerra, 2008).

Encountered major diseases and their treatments: The most common in the farms studied diseases are respiratory diseases and mastitis. There are also other such diseases, metritis, foot and mouth disease and lameness but at low intensities. In case of disease, breeders call the vet for treatment. According to our respondents and veterinary doctors, a certain number of diseases have been recorded (Table 2).

Table 2 - Major diseases and their treatments

Diseases	Symptoms	Treatments	Drugs
Mastitis	Inflammation of the udder	Antiinflammatory	MASTIJET
Météorisations	Swelling of the rumen	Esophageal probe	-
Moths	Mushrooms	fungicides	Antibiotiques
Diarrhea	Acute diarrhea	Antibiotics	-
Infertility	Repeated abortions	-	-
Brucellosis	fever	Slaughter	-

CONCLUSION

Most studies on dairy cattle production focus on the production, nutrition, reproduction and welfare of animals because of the economic importance of these aspects both in regard to the dairy industry and breeding beef breeds. By cons, very few studies have examined the impact of buildings, facilities and their hygiene. However, effective management of livestock is incomplete without a well-designed building, well arranged and held. Failure to do so may affect the production level; additional charges apply for labor and limit the benefits of owner.

The maintenance and hygiene of cattle farms is complex, both for management and development actors and producers breeders. This is due to the complexity of strategies facing those responsible to implement the decrees raising and organization of facilities. This stable management should be based on consultation between



management and ranchers to remove all constraints on the improvement of milk production. It is only then that we can put these government interventions in sustainability.

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THE GROWTH AND REPRODUCTIVE PERFORMANCE OF DIFFERENT BREEDS OF RABBITS KEPT UNDER WARM AND HUMID ENVIRONMENTS IN GHANA

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ABSTRACT: The potential of rabbit production in solving the problem of inadequate animal protein supply cannot be overemphasized. A study was therefore conducted to assess the growth and reproductive performances on 488 bunnies and 87 does of Blue Vienna and 474 bunnies and 80 does of Chinchilla rabbits reared under hot and humid environment in Ghana. The reproductive performance of the two breeds, in terms of litter size at birth and weaning, litter weight at birth and weaning, kindling interval, age at first kindling and gestation length as influenced by season of kindling (rainy and dry), year of kindling (2005-2012) and parity (first to sixth and over) were determined. The overall least square means for litter size at birth, bunny weight at birth, litter size at weaning, bunny weight at weaning, gestation length, kindling interval, age at first kindling, pre-weaning growth rate and post-weaning growth rate of Blue Vienna were 5.6±0.1, 51.3±0.3 g, 4.4±0.1, 601.5±0.9 g, 30.0±0.1 days, 94.7±0.4 days, 159.7±0.4 days, 13.1±0.1 g/day and 15.4±0.1 g/day respectively. Those of Chinchilla were 5.9±0.1, 54.2±0.3 g, 4.8±0.1, 601.9±0.9 g, 30.1±0.1 days, 94.6±0.3 days, 159.8±0.2 days, 13.0±0.1 g/day and 15.3±0.2 g/day respectively. Year of kindling had significant ($P<0.05$) effect on litter size at weaning, litter weight at kindling, gestation length, age at first kindling, post weaning growth rate, body weight at 14 weeks and mortality in both breeds. Season of birth on the other hand had significant effect ($P<0.05$) on litter weight at birth, gestation length and mortality in both breeds. The number of times the doe has kindled (parity) significantly ($P<0.05$) influenced all the parameters apart from kindling interval, gestation length, age at first kindling and pre-weaning growth rate performance of both breeds. The growth performances of the breeds studied were indications of their potential of being developed in future into meat types.

Keywords: Chinchilla, Blue Vienna, Growth, Reproduction, Litter Size, Bunnies.

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INTRODUCTION

According to Biobaku and Dosunmu (2003) and Fayeye and Ayorinde (2003), the increasing human population especially in developing countries coupled with inadequate supply of animal source of protein from the principal livestock species (cattle, sheep, goats, pigs and poultry) has made it imperative that attention be shifted to other micro-livestock such as rabbit. This is because rabbit production has the potential in alleviating the problem of inadequate animal protein supply in developing economies. This, according to Ghosh et al. (2008), is attributed largely to the rabbit's high rate of reproduction, early maturity, rapid growth rate, efficient food utilization and meat of high nutritional value. Its meat is highly digestible, wholesome, tasty, low in cholesterol, sodium and fat with high protein content (Herbert, 2011). Rabbits, because of the enormous benefits associated with their production, and with the belief that the unconventional livestock will certainly bridge the animal protein gap being experienced by man, it is imperative to give available rabbit breeds the needed attention just like other animal genetic resources so as to have more animal products that could supply the immediate needs of man.

The production efficiency of commercial rabbit farms is largely dependent on the litter size at kindling and the survivability of the bunnies up to weaning (Odeyinka et al., 2008). In addition, the pre-weaning growth is very critical in meat rabbits due to its impact on the meat produced at the finisher stage of production (Gerencser et al., 2011).

As production directly depends on reproduction, the reproductive performance of rabbits becomes an important aspect in determining the profitability of commercial rabbit breeding. Factors such as breed, season, age, and weight of females, according to Lazzaroni et al. (2012), influence the reproductive performance of animals. Among the available adaptable foreign breeds of rabbits reared for meat production in Ghana are Flemish Giant,

Chinchilla, New Zealand White and California White. Over the years, attempts to introduce large scale rabbit production as poverty alleviation strategy in Ghana was limited by non-availability of accessible data for informed decision making. There is also paucity of information on the growth and reproductive efficiency of the available breeds in Ghana. Therefore, the present study was undertaken to assess the growth and reproduction performances of Blue Vienna and Chinchilla breeds as influenced by year, season and parity in order to come out with the best strategies for improving the productivity of these rabbits in Ghana.

MATERIAL AND METHODS

Location of the study

The study was carried out at a private breeding farm situated at Amanfro in the Awutu-Senya district of the Central region of Ghana. Two breeds comprising Blue Vienna and Chinchilla were studied. All the animals were reared at the farm of Farmer Brown's Livestock Farm located in the Awutu-Senya District of Ghana. The climate is generally hot, semi-arid and tropical in nature with an average annual rainfall of between 400 and 500 mm with mean annual minimum and maximum temperatures of 22°C and 28°C respectively.

Management of the animals

A doe and its litters were kept together in cages and fed together. The weaned rabbits were however kept in groups of four and five in standard galvanised iron cages measuring 75 X 45 X 35 cm and provided with similar management. In the mornings, concentrate mixture (16% crude protein and 2400 kcal metabolizable energy) was given at the rate of 75 g/d up to 6th wk of age and 100 g/d from 7th to 14th wk of age. For the lactating does and bunnies, a concentrate mixture of 200 to 250 g/d was given, according to their body weight and litter size. In addition, the does were supplemented with green fodder of guinea grass, *Euphorbia spp* and *Desmanthus virgatus* in the afternoons. They were provided with portable water *ad libitum*. Does and their bunnies were housed together up to weaning (i.e. 42 d). A standard prophylactic endo- and ecto-parasitic control schedule was applied. Bucks started their reproductive lives at 8 months of age and were randomly assigned to the females for natural mating. Mating was planned to avoid close relatives mating in order to reduce the level of inbreeding.

Data collection

Reproductive and growth performance records on 488 bunnies and 87 does of Blue Vienna and 474 bunnies and 80 does of Chinchilla obtained from 2005 to 2012 were used. The reproductive performance of the two breeds, in terms of litter size at birth and weaning, litter weight at birth and weaning, kindling interval, age at first kindling and gestation length as influenced by season of kindling (rainy and dry seasons), year of birth (2005 to 2012) and parity (first to sixth and over) were determined. Also the growth performance of the breeds in terms of weight at weaning, pre-and post-weaning growth rate as influenced by the environmental factors mentioned above were also determined. All bunny weights in each litter were obtained within twenty-four hours following kindling and in groups. To study the effect of season of kindling on both the reproductive and growth performance, the calendar year was divided into two seasons: the rainy season (April-November) and dry season (December-March). Age at first kindling was calculated as the age at which the doe had its first bunny. Kindling interval was also calculated as the time elapse between two successive kindling. Gestation length was estimated as period between conception and kindling. Litter size at birth and weaning was done by counting the number of bunnies per litter. On the day of kindling, gloved hand was used to pick the bunnies from one litter from the kindling box and placed on weighing scale. The litter weight was estimated as the weight of the bunnies from a particular doe. This was done by weighing the bunnies using a top-loading sensitive balance of 500g capacity. Weaning weight was taken when the bunnies were 42 days old and in this case they were weighed individually. Growth rate was calculated as the weight gained over a period. After weaning, the body weights were taken individually with a 2kg capacity sensitive top-loading balance.

Data analysis

A fixed effect model was fitted using the Generalized Linear Model (GLM) procedure of GenStat (Discovery Edition) to investigate the fixed effects of season of kindling (2 classes), year of birth (8 classes) and parity (6 classes) on the growth and reproductive performance of Chinchilla and Blue Vienna breeds of rabbits. Where differences in means were observed, the means were separated using the least significant difference at 5% level of significance. The statistical model for the birth weight and other reproductive traits was as follows:

$$Y_{ijk} = \mu + S_i + P_j + Y_k + e_{ijk}$$

Where Y_{ijkl} = any of the reproductive traits

μ = overall mean of the trait

S_i = fixed effect of j^{th} season of kindling (1, 2)

P_j = fixed effect of k^{th} parity of doe (1, 2, 3, 4, 5, 6+)

Y_k = fixed effect of l^{th} year of birth (1, 2...8)

e_{ijk} = random error associated with each observation



RESULTS AND DISCUSSION

Litter size in rabbits is regarded as one of the most important economic traits in any breed development and improvement programmes for intensive meat production. According to Moce and Santacreu (2010) most maternal lines are selected based on litter size at weaning, since this trait reflects both the prolificacy and mothering ability of the doe. The least-squares means of litter size at kindling and weaning in Blue Vienna and Chinchilla breeds are presented in Tables 1 and 3 respectively. The litter size at kindling values obtained in this present study was higher than the values obtained for similar breeds (2.7 to 4.6) under similar environments by Sivakumar et al. (2013) in India; Fayeye and Ayorinde (2010), Okuige and Okocha (2008) and Akpo et al. (2008) in Nigeria; Hasanat et al. (2006) in Bangladesh and Kumar et al. (2006) in India. They were however similar to values of 4.5 – 6.3 obtained by Oseni and Ajayi (2010) for similar breeds in Nigeria. However, other authors such as Ghosh et al. (2006), Das and Yadav (2007), Ghosh et al. (2008) and Saidj et al. (2012) have found higher values of litter size at birth and weaning in these breeds, under similar climatic conditions.

Since litter size at birth and weaning are very important economic traits, the Blue Vienna and Chinchilla rabbits in Ghana could be said to have the potential of being developed into meat types when conscious selection is applied in the populations available. The mean litter size at birth and weaning observed in the Blue Vienna and Chinchilla breeds was comparable to those found in the tropics (Iyeghe-Erakpotobor et al., 2005; Chineke et al., 2006 and Laxmi et al., 2009) under similar tropical climatic conditions of India. Litter size at weaning is an indication of the mothering ability of the doe. The higher the number of bunnies that survive to weaning, the better the mothering ability of the does since before weaning the bunnies depend solely on the doe for their nutrient requirements.

Year of kindling was found to significantly ($P < 0.05$) influence litter size at weaning, age at first kindling, gestation length, post-weaning growth rate and market weight in both breeds. This agrees with the reports by Sivakumar et al. (2013), Fayeye and Ayorinde (2010), Okuige and Okocha (2008) and Akpo et al. (2008) in Nigeria, Kumar et al. (2006) and Sood et al. (2006) on similar rabbit breeds. The possible reason for the variation would probably be the differences in nutritional and management aspects during the years. There was however no clear trend with respect to the year of kindling on the parameters measured. Litter size at kindling, kindling interval and bunny weight at birth were however not significantly ($P > 0.05$) influenced by year of birth, an observation which disagrees with findings by Sivakumar et al. (2013) and Sood et al. (2006).

The season of kindling had significant effect ($P < 0.05$) on litter weight at birth and weaning and gestation length in both breeds. It however did not significantly ($P > 0.05$) influence the other parameters studied. Authors who worked on similar breeds observed significant ($P < 0.05$) effect of season on litter size at birth (Iyeghe-Erakpotobor et al., 2005). The litter weight at birth and weaning and gestation length were significantly better in the rainy season than in the dry season, an observation which was supported by the findings of Chineke et al. (2006) and Sivakumar et al. (2013). Differences in litter weight and gestation length relating to season of kindling might be due to differences in environmental and nutritional conditions (presence of available feed resources) and mothering ability. The litter weight was lower in the dry season as compared to the rainy season in both breeds, an indication of probable stressful environments during the dry season. This might have resulted in reduced feed intake by the lactating doe leading to low production of milk for the bunnies albeit reduced growth of bunnies. This agrees with the findings of Ayyat et al. (1995). Kumar et al. (2006) however reported a non-significant effect of season on gestation length for foreign rabbit breeds like Chinchilla, White Giant and New Zealand White kept in the high altitude conditions of Tamil Nadu. The influence of season on the growth and reproductive performance observed in this study might be attributed to changes in photoperiod (Hudson and Distel, 1990) during the dry and rainy seasons. The availability of quality fodder during the rainy season might have contributed to the higher performance of rabbits in this period of the year. This is because during the rainy season (April-October) the fodder was lush, having higher nutritive value with more protein and carotene content as compared to other seasons. It has been observed that higher feed intake during lactation increased both litter size and weight at birth and weaning (Pascual et al., 2003). It has been recognised that the environment plays an important role in the regulation of reproductive function and that environmental-stimuli, which acts through the nervous system and the hypothalamo-pituitary axis, can affect the reproductive physiology controlled by hormones (Theau-Clement and Mercier, 2004). These stimuli associated with the natural day-length variations, can modify the reproductive performance and the hormonal balance, consequently, the gestation length and kindling interval.

The parity of doe is the number of times a doe has kindled. In this present study, parity was found to have significantly ($P < 0.05$) influenced litter size at birth and weaning, bunny weight at birth and weaning, litter weight at birth and post-weaning growth rate. According to Aksakal and Bayram (2009) the individual pre-weaning body weights are significantly lower in the 1st parity born animals than in other higher parity.

An explanation of this observation is that earlier-parity animals continue to grow until reaching adult size and compete with the foetuses for available nutrients during pregnancy. Again, increased birth weight with increased parity is an indication of older dams' ability to utilize feed more efficiently to support foetal development than younger ones as reported by Aksakal and Bayram (2009) in cows. Results from the present study indicated that bunnies born in late parities were significantly ($P < 0.05$) heavier than their early parity counterparts. This agrees with observations by Ouyed et al. (2011) who recorded significant effect of parity on pre-weaning bunny weight; Xiccato et al. (2004) on litter size at weaning and Das and Yadav (2007) on litter weight at birth as well as litter size and weight at weaning. In contrast, Sivakumar et al. (2013) found no significant effect ($P > 0.05$) of parity on the growth and reproductive performances of similar rabbit breeds studied.



Table 1 - The least mean square of the reproductive performances of Blue Vienna rabbits as influenced by year, parity and season of kindling

Effects	No	Litter size at kindling/no	Litter size at weaning/no	Litter wt. at birth/g	Bunny weight at birth/g	No	Age at first kindling/days	Kindling interval/days	Gestation length/days
Overall	488	5.8±0.1	4.4±0.1	295.6±2.1	51.3±0.3	87	159.7±0.2	94.7±0.4	30.0±0.1
Year									
2005	45	5.9±0.1	4.0 ±0.2 ^{bc}	300.6±6.0 ^b	52.1±0.8 ^a	8	162.5±0.5 ^a	95.2±1.0	30.4±0.2 ^a
2006	55	5.8±0.1	3.7 ±0.3 ^c	284.9±6.0 ^d	51.9±0.9 ^b	10	158.4±0.6 ^b	95.0±1.0	30.2±0.2 ^a
2007	60	6.0±0.1	4.9 ±0.3 ^a	307.1±6.1 ^a	50.9±0.9 ^c	10	158.6±0.6 ^b	94.5±1.0	28.4±0.3 ^b
2008	60	6.1±0.1	4.5 ±0.2 ^a	305.2±6.0 ^a	51.2±0.9 ^b	10	158.7±0.5 ^b	95.4±1.0	30.0±0.2 ^a
2009	75	5.9±0.1	4.4 ±0.2 ^a	283.5±6.0 ^d	50.7±0.9 ^c	13	163.0±0.5 ^a	93.3±1.0	28.8±0.2 ^b
2010	60	6.0±0.1	4.5 ±0.3 ^a	306.9±6.0 ^a	52.5±0.9 ^a	10	158.5±0.6 ^b	94.7±1.0	30.2±0.2 ^a
2011	67	5.9±0.1	4.1 ±0.3 ^{bc}	291.1±6.1 ^c	52.3±0.9 ^a	11	159.0±0.6 ^b	94.0±1.0	30.2±0.3 ^a
2012	66	5.8±0.1	4.7 ±0.3 ^a	285.2±6.0 ^d	50.5±0.9 ^c	15	159.0±0.6 ^b	94.8±1.0	30.2±0.3 ^a
Season									
Rainy	255	5.9±0.1	4.3±0.1	300.5±3.1 ^a	51.9±0.5 ^a	47	159.8±0.3	94.4±1.0	30.6±0.1 ^a
Dry	233	5.8±0.1	4.5±0.1	294.8±3.0 ^b	50.6±0.4 ^b	40	159.7±0.3	94.9±1.0	28.6±0.1 ^b
Parity									
1 st	65	5.5±0.1 ^b	3.8±0.2 ^c	298.9±5.0 ^c	50.3±0.7 ^c	12		95.2±1.0	30.3±0.2
2 nd	70	5.3±0.1 ^b	4.3±0.2 ^{bc}	280.3±5.0 ^d	50.7±0.7 ^c	16		94.9±1.0	30.1±0.2
3 rd	73	5.6±0.1 ^b	3.9±0.2 ^c	282.5±5.0 ^d	51.2±0.7 ^b	13		94.4±1.0	29.8±0.2
4 th	78	5.5±0.1 ^b	4.9±0.2 ^{ab}	294.0±5.1 ^c	53.1±0.7 ^a	14		94.9±1.0	29.7±0.2
5 th	90	6.4±0.1 ^a	4.4±0.2 ^{bc}	318.1±5.3 ^a	51.0±0.8 ^b	14		94.8±1.0	29.9±0.2
≥6 th	112	6.4±0.1 ^a	5.1±0.2 ^a	303.2±5.3 ^b	52.6±0.8 ^a	18		95.3±1.0	30.5±0.2

Means within the same column with different superscripts (^{a,b,c}) are significantly different (P<0.05).



Table 2 - The least mean square of the growth performances of Blue Vienna rabbits as influenced by year, parity and season of kindling

Effects	No	Bunny weight at weaning/g	Pre-weaning growth wt/g/day	Bunny weight at 12 weeks/g	Post-weaning growth weight/g/day	Bunny weight at 14 weeks/kg	Mortality/%
Overall	488	601.5±0.9	13.1±0.1	1342.0±12.0	15.4±0.1	1420.0±30.0	1.5±0.1
Year							
2005	45	601.2±2.0	13.1±0.1	1467.0±32.0 ^a	16.9±0.4 ^a	1510.0±32.5 ^a	1.9±0.2 ^a
2006	55	598.2±3.0	13.0±0.1	1388.0±33.0 ^c	15.9±0.4 ^a	1445.0±32.6 ^b	1.7±0.2 ^a
2007	60	600.8±3.0	13.1±0.1	1408.0±33.4 ^b	16.2±0.4 ^a	1505.0±32.6 ^a	1.3±0.2 ^b
2008	60	603.3±2.0	13.1±0.1	1276.0±32.0 ^d	14.6±0.4 ^b	1355.0±32.5 ^c	1.6±0.2 ^{ab}
2009	75	601.7±2.0	13.0±0.1	1231.0±32.0 ^e	14.1±0.4 ^b	1350.0±32.6 ^c	1.2±0.2 ^b
2010	60	603.7±3.0	13.3±0.1	1407.0±32.4 ^b	16.1±0.4 ^a	1505.0±32.6 ^a	1.6±0.2 ^{ab}
2011	67	602.3±3.0	13.1±0.1	1269.0±33.4 ^d	14.5±0.4 ^b	1350.0±33.0 ^c	1.5±0.2 ^{ab}
2012	66	600.8±3.0	13.2±0.1	1292.0±33.4 ^d	14.8±0.4 ^b	1355.0±32.5 ^c	1.1±0.2 ^b
Season							
Rainy	255	601.4 ±1.0	13.1±0.0	1345.0±17.2 ^a	15.4±0.2	1445.0±20.3	1.7±0.1 ^a
Dry	233	602.6 ±1.0	13.0±0.0	1300.0±16.0 ^b	15.3±0.2	1440.0±20.2	1.3±0.1 ^b
Parity							
1 st	65	597.4 ±2.0 ^b	13.0±0.1	1311.0±28.0 ^c	15.0±0.3 ^b	1445.0±25.5 ^b	2.1±0.2 ^a
2 nd	70	598.8 ±2.0 ^b	13.0±0.1	1355.0±28.0 ^b	15.5±0.3 ^b	1440.0±25.3 ^b	1.0±0.2 ^c
3 rd	73	600.8 ±2.0 ^b	13.2±0.1	1267.0±28.0 ^d	14.5±0.3 ^b	1385.0±25.3 ^c	1.7±0.2 ^b
4 th	78	611.8 ±2.0 ^a	13.3±0.1	1300.0±28.1 ^c	14.8±0.3 ^b	1440.0±25.4 ^b	0.7±0.2 ^d
5 th	90	611.4 ±2.0 ^a	13.1±0.1	1478.0±29.4 ^a	17.0±0.3 ^a	1520.0±24.5 ^a	2.0±0.2 ^a
≥6 th	112	618.4 ±2.0 ^a	13.0±0.1	1348.0±29.4 ^b	17.5±0.3 ^a	1440.0±24.5 ^b	1.3±0.2 ^{cd}

Means within the same column with different superscripts (^{a,b,c}) are significantly different (P<0.05).



Table 3 - The least mean square \pm of the reproductive performances of Chinchilla rabbits as influenced by year, parity and season of kindling

Effects	No	Litter size at kindling/no	Litter size at weaning/no	Litter wt. at birth/g	Bunny weight at birth/g	No	Age at first kindling/days	Kindling interval/days	Gestation length/days
Overall mean	474	5.9 \pm 0.1	4.8 \pm 0.1	317.3 \pm 2.1	54.2 \pm 0.3	80	159.8 \pm 0.2	94.6 \pm 0.3	30.1 \pm 0.1
Year									
2005	50	5.9 \pm 0.1	4.5 \pm 0.2 ^b	305.4 \pm 5.8 ^b	55.7 \pm 0.9 ^a	9	163.0 \pm 0.5 ^a	95.7 \pm 1.0	30.5 \pm 0.2 ^b
2006	48	6.0 \pm 0.1	4.6 \pm 0.2 ^b	327.8 \pm 5.8 ^a	55.7 \pm 0.9 ^a	8	158.3 \pm 0.5 ^b	94.6 \pm 1.0	30.1 \pm 0.2 ^b
2007	60	6.1 \pm 0.1	5.4 \pm 0.2 ^a	329.2 \pm 5.9 ^a	55.1 \pm 0.9 ^a	10	159.0 \pm 0.6 ^b	95.7 \pm 1.0	29.1 \pm 0.2 ^c
2008	65	5.8 \pm 0.1	4.4 \pm 0.2 ^b	304.4 \pm 5.9 ^b	53.6 \pm 0.9 ^b	11	159.2 \pm 0.6 ^b	95.8 \pm 1.0	30.5 \pm 0.2 ^b
2009	65	6.0 \pm 0.1	4.7 \pm 0.3 ^b	329.1 \pm 6.1 ^a	52.6 \pm 0.9 ^c	11	162.8 \pm 0.6 ^a	94.4 \pm 1.0	29.1 \pm 0.3 ^c
2010	55	5.9 \pm 0.1	5.0 \pm 0.2 ^a	305.0 \pm 5.9 ^b	52.5 \pm 0.9 ^c	9	158.2 \pm 0.6 ^b	94.2 \pm 1.0	31.2 \pm 0.2 ^a
2011	60	5.8 \pm 0.1	4.5 \pm 0.2 ^b	305.4 \pm 5.9 ^b	53.6 \pm 0.9 ^b	10	158.7 \pm 0.6 ^b	93.9 \pm 1.0	31.3 \pm 0.2 ^a
2012	71	6.1 \pm 0.1	5.1 \pm 0.3 ^a	332.0 \pm 6.1 ^a	55.1 \pm 0.9 ^a	12	158.8 \pm 0.6 ^b	93.3 \pm 1.0	31.1 \pm 0.3 ^a
Season									
Rainy	274	6.2 \pm 0.1 ^a	4.6 \pm 0.1	323.5 \pm 3.2 ^a	55.3 \pm 0.5 ^a	46	159.9 \pm 0.3	94.1 \pm 0.5	31.4 \pm 0.1 ^a
Dry	200	5.9 \pm 0.1 ^b	4.9 \pm 0.1	312.4 \pm 2.7 ^b	53.4 \pm 0.4 ^b	34	159.7 \pm 0.3	94.9 \pm 0.5	29.9 \pm 0.1 ^b
Parity									
1 st	60	5.4 \pm 0.1 ^b	4.6 \pm 0.2 ^{bc}	314.6 \pm 5.0 ^b	52.1 \pm 0.8 ^c	11		95.1 \pm 0.8	29.0 \pm 0.2 ^c
2 nd	82	5.6 \pm 0.1 ^b	4.4 \pm 0.2 ^c	304.3 \pm 5.0 ^c	54.6 \pm 0.8 ^b	10		94.6 \pm 0.8	30.1 \pm 0.2 ^b
3 rd	85	6.0 \pm 0.1 ^a	4.8 \pm 0.2 ^b	325.0 \pm 5.0 ^a	54.5 \pm 0.8 ^b	14		94.6 \pm 0.8	30.4 \pm 0.2 ^b
4 th	80	6.2 \pm 0.1 ^a	5.1 \pm 0.2 ^{ab}	312.9 \pm 5.2 ^b	54.0 \pm 0.8 ^b	13		94.9 \pm 0.9	30.4 \pm 0.2 ^b
5 th	87	6.1 \pm 0.1 ^a	5.0 \pm 0.2 ^{ab}	323.9 \pm 5.2 ^a	55.2 \pm 0.8 ^a	15		95.5 \pm 0.9	31.4 \pm 0.2 ^a
6 th	80	6.0 \pm 0.1 ^a	5.7 \pm 0.2 ^a	325.4 \pm 5.3 ^a	55.1 \pm 0.8 ^a	17		95.4 \pm 0.9	31.9 \pm 0.2 ^a

Means within the same column with different superscripts (^{a,b,c}) are significantly different ($P < 0.05$).



Table 4- The least mean square of the growth performances of Chinchilla rabbits as influenced by year, parity and season of kindling

Effects	No	Bunny weight at weaning/g	Pre-weaning growth wt/g/day	Bunny weight at 12 weeks/g	Post-weaning growth weight/g/day	Bunny weight at 14 weeks/kg	Mortality/no
Overall mean	474	601.9±0.9	13.3±0.1	1442.0±31.5	15.8±0.2	1445.0±30.1	1.2±0.1
2005	50	599.5±2.4	13.0±0.1	1473.0±31.9 ^a	14.9±0.4 ^b	1395.0±30.0 ^c	1.1±0.2
2006	48	602.3±2.4	13.0±0.1	1362.0±32.0 ^c	15.6±0.4 ^{ab}	1395.0±30.0 ^c	1.5±0.2
2007	60	602.9±2.5	13.1±0.1	1389.0±32.4 ^c	15.9±0.4 ^a	1445.0±30.2 ^b	0.7±0.2
2008	65	600.9±2.5	13.0±0.1	1309.0±32.4 ^d	14.9±0.4 ^b	1400.0±30.1 ^{bc}	1.4±0.2
2009	65	606.3±2.5	13.2±0.1	1263.0±33.4 ^e	14.4±0.4 ^b	1355.0±30.0 ^c	1.7±0.2
2010	55	600.1±2.5	13.0±0.1	1406.0±32.4 ^b	16.1±0.4 ^a	1495.0±30.2 ^a	0.9±0.2
2011	60	600.5±2.5	13.0±0.1	1251.0±32.4 ^e	14.3±0.4 ^b	1420.0±30.1 ^b	1.3±0.2
2012	71	602.6±2.5	13.0±0.1	1281.0±33.4 ^e	16.6±0.4 ^a	1500.0±30.1 ^a	1.0±0.2
Rainy	274	601.4±1.4	13.0±0.1	1372.0±17.8 ^a	15.7±0.2	1505.0±20.2 ^a	1.4±0.1
Dry	200	602.3±1.1	13.1±0.1	1318.0±15.1 ^b	15.1±0.2	1445.0±20.3 ^b	1.1±0.1
1 st	60	600.2±2.1 ^b	13.3±0.1	1339.0±27.5 ^b	15.0±0.3 ^{bc}	1395.0±30.2 ^c	0.5±0.2
2 nd	82	600.9±2.1 ^b	13.0±0.1	1241.0±27.5 ^c	14.5±0.3 ^c	1495.0±30.1 ^a	1.2±0.2
3 rd	85	594.5±2.1 ^b	12.9±0.1	1348.0±27.5 ^b	15.4±0.3 ^a	1395.0±30.1 ^c	1.3±0.2
4 th	80	596.4±2.2 ^b	12.9±0.1	1247.0±28.7 ^c	15.4±0.3 ^b	1390.0±30.1 ^c	0.7±0.2
5 th	87	598.8±2.2 ^b	13.0±0.1	1458.0±28.7 ^a	15.5±0.3 ^b	1420.0±30.2 ^b	2.1±0.2
6 th	80	612.4±2.2 ^a	13.2±0.1	1441.0±29.4 ^a	16.1±0.3 ^a	1505.0±30.2 ^a	1.4±0.2

Means within the same column with different superscripts (^{a,b,c}) are significantly different (P<0.05).



Year, season of birth and parity had a significant ($P < 0.01$) effect on average weight of bunnies at weaning and post-weaning growth rate in both breeds (Tables 2 and 4). These results were in agreement with the findings of Sivakumar et al. (2013) and Sood et al. (2006). Bunnies kindled during the rainy season seem to show higher average body weight at kindling and weaning and post-weaning growth rate. The bigger bunny weight as the years went by could be that when the does were young they tended to reproduce lighter bunnies and as they grew they kindled bigger bunnies due to increase in body size and organs (the size of the womb) to accommodate bigger foetuses. Also increased body size influences feed nutrient intake and its attendant partitioning for growth and other production activities.

There were however no significant effect of parity on gestation length, kindling interval and age at first kindling, which disagrees with the findings of Sivakumar et al. (2013). The gestation length, kindling interval and age at first kindling values obtained in this study were within the ranges (30-32 days; 92-96 days and 157-162 days for gestation length, kindling interval and age at first kindling respectively) obtained for the same breeds by several authors (Sivakumar et al., 2013; Oke and Iheanochi, 2011; Singh et al., 2007; Kumar et al., 2006; Chineke et al., 2006; and Iyeghe-Erakpotobor et al., 2005) under similar environments elsewhere.

The seasonal effect on the early growth performance of rabbit was also reported by Kumar et al. (2001, 2006) and Sood et al. (2006). The lower litter weight at birth during the dry season could be due to the limited availability of good quality green forage to the females. Parity had a significant effect ($P < 0.01$) on litter weight at weaning in both the breeds. Litter weight at weaning increased with parity order. This is in agreement with increase in milk production as parity order advanced (Maertens et al., 2006). Higher litter weight at birth and weaning were reported for White Giant rabbits at different parities (Singh et al., 2007) reared under sub-temperate conditions of India.

CONCLUSION

Rabbit production in Ghana is generally under the intensive system with the animals zero-grazed. The growth and reproductive performance of Chinchilla and Blue Vienna rabbits obtained under this present study were comparable to values obtained in the tropics. With the values obtained for both breeds, it is possible to develop the breeds especially the Chinchilla (due to its superior growth characteristics) into meat types in future if intense selection is undertaken. The objective of commercializing rabbit production as a means of meeting the animal protein needs of the Ghanaian populace could be realized with the current productive performance of the breeds studied. The significant effects of the non-genetic factors on the growth and reproductive performances of the rabbit breeds are indications that any future breed development strategy must take into consideration the environment so that the full genetic potential can be realized.

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EFFECTIVE METHODS FOR APPROPRIATE DIAGNOSIS OF BRUCELLOSIS IN HUMANS AND ANIMALS (REVIEW ARTICLE)

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ABSTRACT: Brucellosis is one of the most common diseases among human that identification and control of disease transmission methods can promote public health. Clinical signs alone are not sufficient for brucellosis diagnosis. Hence, a sensitive, specific, rapid and inexpensive method is required. Early and appropriate diagnosis of this disease is effective in improving public health as well as disease control and eradication. Several serological tests for probable diagnosis of Brucella infection were used in evaluation of antibodies against Brucella. Using new methods such as Elisa has higher sensitivity and specificity than standard SAT test and complement fixation which can show both G and M immunoglobulins. It is also suitable for examining certain class of immunoglobulin. Research and studies have shown that ELISA is a complete method for in vitro detection of chronic disease, especially when other tests results are negative. In addition to this method, all unique and specific immunoglobulin in tested serum appear with high speed and accuracy. Another diagnostic method is PCR, which has higher sensitivity and specificity in comparison with serologic methods for diagnosis of human brucellosis. PCR shows similar sensitivity as 16srRNA using L7/L12 gene. It can be used in diagnosis of human brucellosis. Another diagnostic method is identification of different forms of IL-10 gene, which is a cytokine. It inactivates macrophages and infects the susceptible subject with brucellosis. Therefore, identification of different forms of IL-10 gene is considered as effective method for diagnosis of the disease. It's recommended to use this new and effective method because many of these methods can overcome limitations of traditional methods.

Keywords: Brucellosis, Diagnosis, Human, Animal

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INTRODUCTION

Malta fever or brucellosis is commonly called contagious abortion in animals. This is one of the infectious diseases transmissible between humans and animals. This is also called undulant fever, frenzy fever and Mediterranean fever. This disease occurs in all seasons; however, it is more common in springs and falls during calving and lactation periods in animals. This disease was first discovered by David Bruce in 1887 from spleen of English soldiers killed in war in Malta Island. Therefore, it is called brucellosis disease (AGHA, 2002). Brucellosis is still common in many countries in Mediterranean domain, Middle East, Arabian Peninsula, Central and South America, Asia and Africa. Only 17 countries such as Norway, Scotland, Switzerland, United Kingdom, Denmark, Romania and Netherlands, Manish Island, Sweden and several other countries were formally declared free of Brucellosis. In some countries, like the United States, this disease is primarily considered as an occupational hazard (Gotuzzo et al., 1998). However, this bacterial infection is not restricted to specific jobs in other countries such as Iran. In addition, it is one of the most important disease common between animals and humans. This is also one of the most important health problems. Prevalence of Malta fever in humans and animals depend directly on prevalence of brucellosis. Therefore, this disease should be inevitably controlled and eradicated in order to avoid economic losses and health risks caused by this disease (Megid et al., 2010). Pathogen of this disease is a gram-negative coccobacillus Brucella, which is a small, aerobic, non- motile and non- sporic and non-capsulic. These bacteria grow slowly. Although Brucella grow properly at 37 °C and PH = 6.7 in Brucella broth medium, Brucella colonies usually grow in solid media as smooth, clear, bluish to white and amber colonies. Brucella Canis and Brucella Ovis grow as rough colonieand sometimes as mucoid colonies. There are a few numbers of strains of bacteria with lipopolysaccharide in their outer membranes, which are less virulent. Brucella Abortus and Brucella Ovis need a medium containing 5% to 10% carbon dioxide in initial isolation.

Brucella's resistance in various conditions: Survival of Brucella species depends on type of nutrients, amount of heat and moisture and PH-level. The pathogen survives in proper environmental conditions with appropriate humidity, in animal feces and urine for weeks and sometimes months. Brucella species can survive in frozen meat



for three weeks, in raw milk for 10 days, in a fresh cheese up to three months and in ice cream and cream for a month. These bacteria are not destroyed with freezing. These bacteria cannot survive in yogurt due to presence of lactic acid. This organism survives for 40 days in dry soil contaminated with urine, feces and discharge and products of infected pregnant animals. This bacterium is much more resistant in moist soil. This survives in animal feces in open air for 100 days while it survives at 8° c more than one year. However, it is destroyed at 60° c in 10 minutes. Nevertheless, the number of these organisms severely decreases within a few days by smoking, salting and freezing the infected meat (AGHA, 2002).

Brucella types: there are four types of Brucella detected as agent of majority of brucellosis infection in humans.

Brucella Melitensis has three serotypes. Most cases are infected by direct or indirect contact with sheep and goats while little number of cases occurs due to contact with camels and cattle. *Brucella melitensis* is a major cause of brucellosis in humans. Serotype 1 is common in Iran.

Brucella Abortus has seven serotypes. Most cases occur due to contact with cattle while few cases occur due to contact with camels and yaks. This type of Brucella is less virulent than *Brucella melitensis* in humans. Serotype 3 is common in Iran.

Brucella Suis has five serotypes. The infected cases occur due to contact with pigs. This type of Brucella can cause abortion in pigs. Serotypes 1 and 3 cause infection in humans.

Brucella Canis occurs due to contact with dogs. It causes asymptomatic infection in humans.

Brucella Ovis, *Brucella Maris*, *Brucella neotome* are less common than above-mentioned four strains (Cecil, 2000; Oxford, 1996).

Brucella melitensis is the most common pathogen of this disease in humans. Any disease caused by *Brucella Canis* and *Brucella Maris* is extremely rare in humans. *Brucella Ovis* causes testicular swelling in ram while it does not cause disease in humans. It is not proved whether *Brucella neotome* and *Brucella microti* (pathogen of brucellosis in rodents) cause disease in humans.

Brucellosis transmission methods in humans and animals

Malta fever is transmitted to animals by mating, consumption of infected milk, respiratory transmission in folds and stalls, contact with uterine secretions of infected cattle or infected and aborted fetuses and placenta. This infection is mostly transmitted to animals through ingestion of food contaminated with fecal contents. Due to acidic vaginal medium of animals, the bacteria that enter into vagina through sex may not be pathogenic.

Brucella bacteria enter human body through several ways. It infects human body orally through consumption of infected unpasteurized milk and dairy products, which is one of the most common methods of transmitting the disease. Other consumers products made from infected animals such as liver, meat, blood consumed raw or undercooked are also considered as source of infection. Transmission of infection through inhalation routes is usually considered as an occupational hazard among shepherds, animal transporters, farm workers, slaughterhouse workers, veterinarians and veterinary technicians. Respiration is the most common way of transmitting the infection among laboratory workers. The bacteria may randomly infect cases during butchering. Veterinarians or animal husbandry workers may be infected through skin contact with infected secretions of livestock. Infection of eyes with infected fecal material during animal care is also common. Accidental injection of live *Brucella* vaccine (Rev1, S19 and RB51) to animals may lead to mild form of the disease. No case of infection transmission from human to human was reported. However, the risk of transmission through intrauterine, breastfeeding, blood transfusions, bone marrow transplants and sexual contact still exist (Megid et al., 2010).

Brucellosis diagnosis in humans and animals

Risk of the disease and observing clinical symptom are evident in case of contact with bacteria. However, the disease can be diagnosed in in vitro observation. Brucellosis disease can be diagnosed early in livestock through blood serum, milk, aborted fetuses in morbidly appropriate samples sent to the laboratory using screening tests such as Rose Bengal RBPT, Milk Ring Test (MRT) and direct ELISA (D-ELISA). Subsequently, the disease is confirmed and diagnosed by complementary tests such as Wright seroagglutination test (SAT), 2ME Mercaptoethanol test, indirect ELISA (I-ELISA) and competitive ELISA (C-ELISA). Cultures of clinical specimens and isolation of infectious pathogens indicate definite diagnosis of the disease.

Brucellosis laboratory tests in humans are often performed in case the patients visit hospitals. Brucellosis is diagnosed based on epidemiologic history, clinical findings and high or increasing *Brucella* antibody titers with or without positive cultures of blood or other fluids and tissues. Many different studies were conducted in order to achieve faster and better diagnostic procedures (Megid et al., 2010; Kokoglu et al., 2006). The Golden Standard for diagnosis of this disease through isolation of bacteria from blood, bone marrow or septic aggregation is not reliable. However, in practice, obtaining a positive blood culture and using brucellosis diagnostic methods are associated with several problems such as time consuming, risk of personnel infection and getting false negative results. Therefore, serological testing is essential (Haddadi et al., 2006; Malik, 1997; Elbeltagy, 2001; Alvarez et al., 2000).

Diagnosis through clinical symptoms

- **Symptoms in humans:** Disease latency (From time of contact with source of infection to occurrence of symptoms) is often between 1 and 3 weeks. However, it is sometimes up to 6 months. Based on severity of the disease, symptoms are manifested in three forms: acute, sub-acute and chronic.



The acute form: the patient suffers from sudden chills, general body aches, especially back pain and intense sweating. He may lose his appetite and suffer from weaknesses and lassitude. In addition, symptoms may occur no more than three months passed from infection with the disease.

Sub-acute form: it begins silently. The patient mainly complains of weakness and fatigue. The symptoms may manifest from 3 to 12 months since beginning of the disease.

Chronic form: chronic form of this disease may occur in case that more than one year has passed from time of diagnosis when the patient is still suffering from the disease. The subjects who may show those symptoms such as fever, lack of appetite, muscle aches, and night sweating or have a history of contact with infected animals, are suspected of having brucellosis. Those who consume infected dairy products should be evaluated in terms of brucellosis. This disease is differentially diagnosed due to variety of clinical symptoms in accordance with many other infectious and non-infectious diseases (Megid et al., 2010).

Symptoms in animals

Brucellosis highly reproduces in milk glands and uterus of pregnant and lactating animals. These organisms reside in chorionic epithelial cells, which cause necrosis in placental cotyledons. This usually leads to abortion of fetuses in pregnant animals due to intrauterine infection. Animals usually recover on their own. However, the infected animals dispose pathogenic organisms through their uterine, urine and milk secretions for a variable period and cause infection in other animals or humans. On the other hand, cattle and goats may remain infected during their entire life. In addition, animals with chronic brucellosis may not suffer from abortion. It is not likely that they produce less milk. Moreover, they can transmit the disease to other animals. They may be an important source of human infection through their milk products. Occurrence of clinical signs of disease in livestock is dependent on level of safety of the herds. In non-vaccinated flocks, abortion is the most important symptom of the disease. Abortion occurs in cattle after the fifth month of pregnancy while it occurs in the last two months of pregnancy in sheep and goats. Although incidence of abortion is observed in the second and ongoing months of pregnancy in a number of infected animals, most animals do not miscarry in the second pregnancy and afterwards. Retained placenta, metritis, arakit, hygroma, decreased milk production, permanent or temporary infertility, delay in reproductive seasons and increased lactation intervals can be cited as other symptoms of this disease. Those symptoms such as fever, respiratory impairment, weight loss, diarrhea and limping may be observed in acute form of the disease (Megid et al., 2010).

Evaluation of clinical signs and symptom in various studies: In a study conducted in Turkey, 78.3% had fever while 77.5% had arthralgia, 72.5% showed sweating and 7.5% had epididymoorchitis. In a study carried out in Saudi Arabia, 79.2% had fever while 70.4% had arthralgia and 3.8% had splenomegaly (Kokoglu et al., 2006; Fallatah et al., 2005).

The most common symptoms were fever (67.22%), sweating (73.25%), malaise and fatigue (65.45%), arthralgia (29.15%), headache and coughing (Haddadi et al., 2006).

In another study conducted on 104 patients in Saudi Arabia, 100% had fever while 96.2% had sweating, 70% had headache, 73.1% had back pain and 76.9% had arthralgia (Malik, 1997). Hasanjani Roushan et al. conducted a study on 404 subjects and showed that the most common symptoms were fever, sweating and arthralgia (Hasanjani Roushan et al., 2004). In a study conducted in Kuwait, the most common symptoms were sweating, fever, headache and arthralgia with respectively 91%, 40%, 49% and 23% incidences (Mousa et al., 1987). In another study carried out in Saudi Arabia, 100% had fever while 46.2% had hepatomegaly, 42.3 had splenomegaly and 26.6% had arthritis (Fallatah et al., 2005).

In another study conducted in Turkey, 66.6% had fever, 63.3% had hepatomegaly, 56.6% had splenomegaly, 23.3% had arthritis and 6.8% had epididymo-orchitis (Namiduru et al., 2003). In a study conducted in Kuwait, 27% had hepatomegaly while 37% had arthritis (Mousa et al., 1988). Rasoulinejad et al. conducted a study on 505 patients and showed that 42% had hepatomegaly while 34% had splenomegaly and few cases had arthritis. The most involved joints were knees, hips and sacroiliac joints (Rasoulinejad et al., 2002). In another study conducted in Iran, the most involved joints were knee, sacroiliac and spondylitis joints (Hasanjani Roushan et al., 2004). In a survey conducted on 238 patients in Turkey over 6 years, 36.5 % patients had osteoarticular involvement. The most involved joints were respectively sacroiliac, peripheral arthritis, spondylitis and bursitis (Tasova et al., 1999) in two studies conducted in Spain; the most involved joints were sacroiliac and spondylitis (Ariza et al., 1993; Gonzalez et al., 1999).

Laboratory

Cell blood count: monocytosis, lymphocytosis and anemia were observed in a study conducted in Turkey (Tasova et al., 1999). In the study conducted by Roshan et al, 84.5% had normal WBC while 80.8% had normal HB and 80.7% had normal ESR and 60.4% had normal CRP (Hasanjani Roushan et al., 2004).

Serologic Tests: Several serological tests were used for probable diagnosis of Brucella infection in evaluation of antibodies against Brucella. Tube standard agglutination test or Wright Test is cited as one of the oldest test invented by Wright and Smith in 1897 (Tohme et al., 2001). Coombs Wright Methods (Anti-human globulin test), complement fixation (CFT), agglutination 2 - mercaptoethanol (2-ME), Ring Rose Bengal test and fast agglutination on lam are cited as other serological valuable methods used for diagnosis of brucellosis (Ariza et al., 1992; Gazapo et al., 1989; Lulu et al., 1986; Peraza et al., 2004). Nowadays, countries such as Germany, Cuba, the United States and China proceeded to produce Elisa kits to detect immune response against brucellosis in humans and or



animals. According to research conducted on many industrial Elisa kits, human immunoglobulin antibodies such as IgM or IgG were solely used (Ferreira et al., 2003; Hajia 2006). In a number of these kits, IgM and IgG were used mutually as conjugate. In this case, the kits could detect both acute and chronic phases of the disease (Ferreira et al., 2003; Hajia 2006; Ertek et al., 2006; Henk et al., 2003).

In a study conducted by Araj et al. they not only compared tube standard agglutination test (SAT) with ELISA test, but also introduced ELISA test as the selected test for diagnosis in patients clinically suspected of having brucellosis (Araj et al., 2005). In a study conducted in Kuwait on application of ELISA for diagnosis of Brucella, sensitivity and specificity of IgG ELISA was detected as 98% for patients with acute or chronic brucellosis. The researchers also expressed that ELISA test is a rapid, sensitive and specific method for diagnosis of Brucella bacteria in humans provided that a view of immunoglobulin classes be prepared in diagnosis of acute and chronic brucellosis. Therefore, ELISA method can be described as a selected method for serological diagnosis of this disease (Ferreira et al., 2003). Chaudhuri et al. used a recombinant protein with 28 kilo dalton weight in outer membrane (OMP28) of Brucella melitensis as antigen. This antigen can induce immune system in cattle, sheep, goats and dogs. The sensitivity and specificity caused by this recombinant antigen were respectively equal to 88.7% and 93%, which were less than sensitivity and specificity compared to SLPS antigen used in this study (Chaudhuri et al., 2010).

A study was conducted in Iran using ELISA method with kit designed to diagnose Brucellosis in human and livestock on 40 serum samples (10 human serum and 30 livestock samples). It was reported that Wright test was positive. It showed that all samples with designed ELISA kits showed positive test results. In addition, 86 samples showed negative results among 89 negative serum samples (41 human serum samples and 48 livestock serum samples). In this study, sensitivity was obtained as 100 % and specificity as 95.83% for livestock kits while sensitivity was obtained as 100% and specificity as 97.56% for human kits and sensitivity was obtained as 100 % and specificity as 96.73% for combined kits. Accordingly, the threshold or (Cut off) was also determined as 0.13. These researchers showed that this kit could simultaneously diagnose brucellosis in animals or humans using two conjugates. High accuracy, sensitivity and specificity with fast testing procedure are cited as advantages of this method compared with other serological tests. Using a very small amount of patient's serum compared to other indices also resulted in high speed, high accuracy, ease of testing, high sensitivity, diagnosis of the disease in both acute and chronic phases and reducing the time for brucellosis diagnosis to 75 minutes from 24 hours with tube standard agglutination test (SAT) (samavati et al., 2012).

Type-1 immunity is important in controlling Brucella and macrophages infection. Interleukin- 10 is a type 2 cytokine, which deactivates macrophages and has adverse effects on the disease (Serre et al., 1987; Araya et al., 1989). Studies have shown that Interleukin-10 gene promoter polymorphisms can affect production of these cytokines (Cheers, 1984; Jiang and Baldwin, 1993; Jones and Winter, 1992; Baldwin and Parent, 2002). In a study conducted in 2008 in Iran, the effect of polymorphisms on susceptibility to brucellosis disease was examined. In the former study, 190 patients with brucellosis and 81 healthy ranchers who had infected animals and consumed infected dairy products were studied. All patients were genotyped in terms of two allelic polymorphisms in interleukin- 10 gene promoter region at positions of 1082 - (G/A), 819 - (T/C) and 592 - (A/C) using PCR-RFLP. The research results showed that distribution of CC genotypes and C alleles in positions of 592 and 819 of IL-10 gene were significantly higher in patients compared to healthy subjects (P-value was respectively equal to 0.034 and 0.008). Thus, ATA single and double haplotypes were significantly higher in control group compared to patients (P-value were respectively as 0.0278 and 0.013). Therefore, higher frequency of C alleles at positions of -819 and -592 of IL- 10 and lower frequency of ATA/ATA haplotype in patients are considered as predisposing factors for brucellosis disease (Rasouli et al., 2009).

Fernandes et al. also conducted a study and showed that neutralizing IL- 10 by monoclonal antibodies produces gamma interferon. In addition, lysing power of spleen cells increases against Brucella Abortus (Fernandes et al., 1995). Down-regulation effects of IL- 10 in immune system of the subjects infected with intracellular bacterial and parasitic infections were also observed (Bermudez et al., 1993; Wagner et al., 1994; Sher et al., 1991; Silva et al., 1992).

Molecular methods for diagnosis of brucellosis were also conducted. Khamesipour et al. conducted brucellosis molecular diagnosis on 135 blood samples from slaughtered sheep using polymerase chain reaction. Then, their DNA was extracted and isolated using PCR method for diagnosis of brucellosis. In total, 135 samples were studied in which 42 cases (31.11%) were infected with brucellosis. These were diagnosed positive in PCR tests (Khamesipour et al., 2013). In another study conducted by Sharifi-Zadeh et al. brucellosis and leptospirosis were molecularly diagnosed in abortion cases using multiplex PCR. In this study, multiplex PCR method was adjusted for simultaneous searching of these two bacteria. Then, these factors were directly determined from contents of aborted bovine fetuses. It was both simplicity and possible to search simultaneously for these two bacteria. Then, multiplex PCR method can be used as a convenient alternative culture method compared to conventional methods (Sharifi-Zadeh et al., 2010).

Raeisi et al. conducted a study in 2010. In this study, an indirect ELISA kit was designed for serological diagnosis of brucellosis and achieving higher sensitivity compared to other conventional methods. In this study, smooth lipopolysaccharide (S-LPS) of Brucella melitensis prepared commercially with high purity was used as antigen to coat microplates. Indirect ELISA method showed positive results in 194 serum samples among which 194 samples showed positive Wright test using the designed ELISA kit while 72 samples showed negative results from 75 negative serum samples. These respectively indicated 100% sensitivity and 96% specificity for the



designed kit. Accordingly, the threshold or cut off was determined as 0.19. This test showed high accuracy and speed in conducting the test compared to other serologic tests and foreign kits (Raeisi et al., 2010).

Pakzad et al. conducted a study conducted in 2011 on 700 blood samples collected from febrile patients with suspected brucellosis who visited ilam hospitals and laboratories for serological tests. These samples were screened using Rose Bengal Test. Then, 50 positive Rose Bengal samples were examined by Wright, Coombs Wright tests and PCR using two genes of 16srRNA and L7/L12 while 50 negative Rose Bengal samples were tested using PCR with two aforementioned genes. The results indicated that 125 samples were positive while 575 were negative among 700 samples tested by Rose Bengal. In addition, 50 Rose Bengal positive samples were positive in PCR using both genes while 50 Rose Bengal negative samples were negative in PCR using both genes. Moreover, 47 samples in Wright test and 49 samples in Coombs Wright test had high titers of 1:60. These researchers found out that PCR method has higher sensitivity and specificity compared to serological methods in human brucellosis diagnosis. PCR has similar sensitivity as 16srRNA gene using L7/L12 genes. It can be used for human brucellosis diagnosis (Pakzad et al., 2011).

CONCLUSION

Brucellosis is one of the most common diseases among human and animal, which is called thousand faces disease due to long lasting side effects. Certainly, identification and control of disease transmission methods can promote public health. No effective and safe vaccine is available for humans. Clinical signs alone are not sufficient for brucellosis diagnosis. Then, a sensitive, specific, rapid and inexpensive method is required. Early and appropriate diagnosis of this disease is effective in improving public health as well as disease control and eradication. Several serological tests for probable diagnosis of Brucella infection were used in evaluation of antibodies against Brucella. The oldest tests are tube standard agglutination test or Wright Test, Coombs Wright methods, anti-human globulin test, complement fixation test (CFT), agglutination 2 – mercaptoethanol (2-ME), Rose Bengal Test and rapid agglutination on lam. These are serological diagnosis methods for brucellosis. These are not appropriate for definitive diagnosis of brucellosis. Using new methods such as Elisa has higher sensitivity and specificity than standard SAT test and complement fixation, which can show both G and M immunoglobulins. It is also suitable for examining certain class of immunoglobulin. On the other hand, this method shows all antibodies generated in reaction with surface antigens of Brucella. It can also prevent the complexity created by glucan or incomplete antibodies. Therefore, acute brucellosis can be easily diagnosed from chronic brucellosis using this method. When interpreting agglutination test is met with confusion, the result can be confirmed using ELISA test. Although SAT method has relatively high sensitivity, it is time-consuming. The results should be read with focus and precision. However, ELISA is one of the methods for in vitro measurement of immune response in the solid phase. Therefore, many drawbacks of safety evaluation methods in liquid phase such as time-consuming manner, initial preparation and high non-specific connections are eliminated in this new method. It should be noted that IgG, IgM (IgG1, IgG2), IgA and partial amount of IgE are produced in Brucellosis humoral immunity response. IgG is particularly involved in serological tests. IgM appears on the fifth to seventh day of brucellosis infection and reaches the final amount during 13 to 21 days after bacteria penetrated the body. Low amount of IgA is also generated in the interval between emergences of above two immunoglobulins. IgG titer is higher and more durable during the disease. This is significant in serological survey of brucellosis when the serum is tested. If infected serum in the first week was tested, no immunoglobulin may be observed. Thus, the test result will be negative. IgM level increases in second week. IgG is generated between the second and third weeks. IgG reaches the maximum level after three weeks. This level is still high during infection. Research and studies have shown that ELISA is a complete method for in vitro detection of chronic samples, especially when other tests results are negative. In addition to this method, all unique and specific immunoglobulin in tested serum appear with high speed and accuracy. In recent years, indirect ELISA has considerably improved. In most experiments, a purified variable amount of S-LPS is used as antigen. Another diagnostic method is PCR, which has higher sensitivity and specificity in comparison with serologic methods for diagnosis of human brucellosis. PCR shows similar sensitivity as 16srRNA using L7/L12 gene. It can be used in diagnosis of human brucellosis. Another diagnostic method is identification of different forms of IL-10 gene, which is a cytokine. It inactivates macrophages and infects the susceptible subject with brucellosis. Therefore, identification of different forms of IL-10 gene, which affect production of these cytokines, is considered as effective method for diagnosis of the disease. It is concluded that due to limitations of serological and culturing methods, which are time consuming, risky and expensive, as well as importance of early detection of bacteria in epidemic cases, it is recommended to use this new and effective method because many of these methods can overcome limitations of traditional methods.

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INFLUENCE OF WEANING AGE ON THE ORGANOLEPTIC PROPERTIES OF PORK

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ABSTRACT: A trial was conducted to compare organoleptic properties of piglets weaned at 21, 28 and 35 days of age and slaughtered at 70 kg target body weight. A total of 24 pigs (Landrace x Large white x Topigs x Topigs cross) which were weaned at three weaning ages were randomly selected and slaughtered upon reaching target body weight. Each treatment comprised three replicates of two animals each. Out of this number, 18 carcasses were selected for organoleptic evaluation. Piglets were provided with creep diet from 10 to 35 days, a weaner diet from 36 to 70 days of age and pig grower diet from 71 days to target slaughter weight. Thereafter, pigs were sacrificed and their carcasses chilled for 20 hours at 5 °C before cuts were removed. The four meat cuts (the pork chop, chuck, Top sirloin and pork leg) were removed, cooked and tested for organoleptic evaluation, *i.e.*, texture, tenderness, juiciness, appearance and flavour. Data on organoleptic properties were analysed using frequencies and percentages in IBM SPSS statistics for Windows, version 20.0. Results showed that the panelists preferred pork from pigs weaned at 28 days followed by 21 days and 35 days. The appearance of pork was the most preferred characteristic across all weaning periods as it showed high rankings by panelists while juiciness appeared to be the less preferred attribute. These results suggest that weaning age influenced organoleptic properties of pork with 28 days weaning age giving better results.

Keywords: *Ad libitum*, Organoleptic Properties, Piglets, Pork, Weaning Age

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INTRODUCTION

The two major weaning methods that are practiced to improve production in pig operations are early and late weaning. In early weaning, piglets are weaned from 10 to 21 of days of age (Fangman et al., 1997; Hohenshell et al., 2000), whereas in late weaning piglets are weaned at 24 to 30 days of age (Drum et al., 1998; Hohenshell et al., 2000). Globally, weaning age varies between 14 to 56 days (English et al., 1988). In South Africa, weaning age ranges from 21 to 28 days (Grimbeek, 2004) while in Botswana it ranges from 35 to 42 days. It, however, appears that the common weaning age in large-scale pig operations in Botswana is 35 days.

Many producers and scientists are re-evaluating weaning age decisions, comparing growth differences and herd-health issues among pigs weaned at different ages (Smith et al., 2007). Therefore, this study was conducted to compare organoleptic properties of pigs weaned at 21, 28 and 35 days and slaughtered at a target body weight of 70 kg.

MATERIAL AND METHODS

Experimental site

The experiment was carried out at Chemaie Farm in Oodi-Matebeleng in Kgatleng District from January to April 2014. The site is situated on coordinates 24° 40' 54.12" S 26° 1' 3.55"E, at an altitude of 980 m above sea level. The area has an average daily temperature of 15 °C in winter (May to July) and 29 °C in summer (August to November).

Experimental animals and management procedure

The animals used in this study were obtained from an on-going experiment at Chemaie Farm. The experiment comprised three treatments (weaning ages), *i.e.*, 21, 28 and 35 days. Each treatment comprised four replicates of four animals (2 females and 2 males). All piglets were fed creep diet from 10 to 35 days. This feeding regime was in accordance with BOS 190:2006, which is the Botswana standard for pig feeds. Water and feeds were provided *ad libitum* throughout the study. Pigs were housed in a grow-to-finish facility in solid pens equipped with a long trough and nipple drinker. Body weight of piglets were recorded at birth and weekly after weaning until 70 kg target body weight was reached.



Processing procedure

A total of 24 pigs (Landrace x Large white x Topigs x Topigs cross) which were weaned at three weaning ages (*i.e.*, 21, 28 and 35 days) were randomly selected and slaughtered at 70 kg target body weight. Out of this number, 18 carcasses were selected for organoleptic evaluation. At target body weight, pigs were inspected to determine their fitness for slaughter and movement permit issued by the Department of Veterinary Services. Thereafter, pigs were transported in an open truck with rails to a slaughterhouse. Prior to slaughter, pigs were starved for 12 hours and offered water *ad libitum* and thereafter sacrificed. *Post mortem* inspection was conducted on the carcasses and carcasses were passed as fit for human consumption. Carcasses were then transported to Meat Science Laboratory at Botswana College of Agriculture (BCA) where they were chilled at 7 °C for 24 hours before meat cuts were removed. The four meat cuts (*i.e.*, pork chops, chuck, Top sirloin and pork leg) were removed and tested for sensory qualities with respect to texture, tenderness, juiciness, appearance and flavour. Meat samples were kept in the freezer at -8 °C for about a month. Prior to cooking meat was thawed at 4 °C for 24 hours (Aaslyng et al., 2007). Thereafter, meat samples were cooked at the College Refectory according to treatment with every cut put in its own tray. Before the meat samples were put on an oven, 0.41 g of salt, 0.43 g barbeque spice and 250 ml of cooking oil was mixed and spread all over the meat in each treatment. Meat temperature was brought to a room temperature before cooking. Meat samples were oven-fried at 80 °C for 60 minutes and turned every 30 minutes. After frying the samples were cut into small cubes and taken to the Meat Science Laboratory at BCA for assessment by panelists.

Sensory descriptive analysis

The panel for the sensory descriptive analysis which consisted of 10 assessors (panelists) from BCA, irrespective of gender was assembled. The panel underwent basic training in sensory assessment in accordance with ISO 4121, ASTM-MNL 13, DIN 10964 (Aaslyng et al., 2007) in order to familiarise it (panel) with sensory assessment of meat. During training, the profile of the sensory attributes was developed in cooperation with the assessors. The panel determined flavour, texture, tenderness, moisture (juiciness) and appearance of pork. Each panelist was allowed to assess each sample at a time and had a sip of water before he/she could assess another sample.

Statistical analysis

Data on organoleptic properties were analysed using frequencies and percentages in IBM SPSS Statistics for Windows, version 20.0 (2011).

RESULTS AND DISCUSSION

Texture

Eighty percent of the panelists ranked texture of chuck and sirloin as liked moderately to liked extremely followed by chop and pork leg with 70% each (Table 1). Chuck from pigs weaned at 28 days had 100% acceptability (liked moderately to liked extremely) followed by sirloin (80%), pork leg (80%) and chop (60%) (Table 2). Again, chuck from pigs weaned at 35 days had the highest frequency and percentage of the preferred texture ranging from like moderately to like extremely (90% ranking) followed by sirloin (80%), pork leg (80%) and chop with 60% (Table 3). Overall, pork from pigs weaned at 28 days ranked highest in texture with an average of 80% followed by 35 and 21 days with 77.5% and 75%, respectively. Beilken et al. (1990) attributed the differences in texture to physico-chemical processes occurring in the meat tissue during heating, which causes the next significant changes in its microstructure, texture and water-holding capacity. Dransfield and Macfie (1980) mentioned that the variation in texture within the *longissimus dorsi* in pork was possibly due to higher degree of localized muscle shortening. Furthermore, Warriss (2000) stated that dietary manipulations and cooking methods may offer greater potential to enhance meat texture.

Flavour

Eighty percent of the panelists ranked the flavour of chuck and sirloin from pigs weaned at 21 days between like moderately and like extremely while the chop and pork leg had the least ranking with 70% each (Table 1). Sirloin from pigs weaned at 28 days had acceptability of 100% while the chop had 70% acceptability (Table 2). The chop and sirloin from pigs weaned at 35 days had the highest acceptability of 90% each while the chuck and pork leg had lower acceptability of 80% each (Table 3). Overall, pork from pigs weaned at 28 days was ranked the highest in flavour with an average of 90% followed by 35 days and 21 days with 85% and 75%, respectively. It has been demonstrated that flavour can be influenced by precursors such as feed (Koutsidis et al., 2007), pre-slaughter stress (D´Souza et al., 1998) and ageing (Koutsidis et al., 2003). Bejerholm and Aaslyng (2003) studied the influence of cooking technique on the sensory evaluation of pork with different raw meat qualities and found that pan-frying gave more intense fried/roasted flavour compared to roasting in an oven. According to Mottram (1991), pork flavour is mainly generated during the heating process, with Maillard reactions involving reducing carbohydrates and amino acids, as one of the most important routes to flavour formation.

Tenderness

Over 85% of the panelists rated pork from pigs weaned at 21 days to be just about right to too tender (Table 1). All the respondents rated chuck to be tenderer compared to other cuts. According to Table 2, chuck from pigs weaned at 28 days was the most preferred (90%) and sirloin the least preferred (70%). In this study, chop from pigs



weaned at 35 days was less preferred (50%) while chuck was highly preferred (80%). On average pork from pigs weaned at 21 days was ranked the highest in tenderness with 85% followed by 28 days and 35 days with 80% and 65%, respectively. The current results indicate that tenderness decreases with age. According to Enfalt et al. (1977), the variation in tenderness could be due to the connective tissue surrounding the muscle fibre not breaking down easily during mastication. Tenderness can be influenced by cooking methods; long cooking times, in particular cooking by boiling can tenderize meat containing larger amount of connective tissues by converting it to gelatin (Warriss, 2000). Culler et al. (1978) reported myofibril fragmentation to be a more important effector of tenderness than sarcomere length or collagen solubility. For Seideman et al. (1986), one of the reasons of variation in tenderness of meat could be the muscle characteristics, including fibre type frequency, which may affect palatability.

Table 1 -Sensory evaluation of pork from pigs weaned at 21 days of age

Parameters	Chuck		Sirloin		Chop		Pork leg	
	Frequency	%	Frequency	%	Frequency	%	frequency	%
Texture								
Dislike moderately	1	1	0	0	0	0	1	10
Dislike slightly	0	0	2	20	2	20	2	20
Neither like nor dislike	1	10	0	0	1	10	0	0
Like moderately	3	30	2	20	3	30	2	20
Like very much	3	30	3	30	2	20	4	40
Like extremely	2	20	3	30	2	20	1	10
Total	10	100	10	100	10	100	10	100
Flavour								
Dislike slightly	0	0	2	20	2	20	0	0
Neither like nor dislike	1	10	0	0	1	10	1	10
Like moderately	6	60	3	30	4	40	5	50
Like very much	2	20	3	30	2	20	3	30
Like extremely	1	10	2	20	1	10	1	10
Total	10	100	10	100	10	100	10	100
Tenderness								
Too tough	0	0	2	20	2	20	2	20
Just about right	3	30	3	30	5	50	6	60
Too tender	5	50	3	30	2	20	2	20
Much too tender	2	20	2	20	1	10	0	0
Total	10	100	10	100	10	100	10	100
Juiciness								
Dislike very much	0	0	1	10	1	10	0	0
Dislike moderately	0	0	1	10	1	10	0	0
Dislike slightly	0	0	2	20	1	10	2	20
Neither like nor dislike	0	0	1	10	2	20	2	20
Like moderately	5	50	3	30	3	30	4	40
Like very much	1	10	1	10	1	10	0	0
Like extremely	4	40	1	10	1	10	2	20
Total	10	100	10	100	10	100	10	100
Appearance								
Dislike very much	0	0	0	0	1	10	0	0
Dislike moderately	0	0	1	10	0	0	0	0
Dislike slightly	0	0	1	10	0	0	0	0
Neither like nor dislike	0	0	0	0	1	10	1	10
Like moderately	4	40	4	40	3	30	3	30
Like very much	4	40	1	10	3	30	3	30
Like extremely	2	20	3	30	2	20	3	30
Total	10	100	10	100	10	100	10	100



Table 1 -Sensory evaluation of pork from pigs weaned at 28 days of age

Parameters	Chuck		Sirloin		Chop		Pork leg	
	Frequency	%	Frequency	%	Frequency	%	frequency	%
Texture								
Dislike very much	0	0	1	10	0	0	0	0
Dislike slightly	0	0	1	10	1	10	2	20
Neither like nor dislike	0	0	2	20	1	10	0	0
Like moderately	6	60	2	20	4	40	2	20
Like very much	3	30	2	20	3	30	6	60
Like extremely	1	10	2	20	1	10	0	0
Total	10	100	10	100	10	100	10	100
Flavour								
Dislike moderately	0	0	0	0	0	0	1	10
Dislike slightly	0	0	2	20	0	0	0	0
Neither like nor dislike	1	10	1	10	0	0	0	0
Like moderately	1	10	3	30	6	60	3	30
Like very much	4	40	3	30	3	30	4	40
Like extremely	4	40	1	10	1	10	2	20
Total	10	100	10	100	10	100	10	100
Tenderness								
Much too tough	0	0	0	0	0	0	1	10
Too tough	1	10	3	30	2	20	1	10
Just about right	5	50	5	50	4	40	4	40
Too tender	3	30	2	20	3	30	4	40
Much too tender	1	10	0	0	1	10	0	0
Total	10	100	10	100	10	100	10	100
Juiciness								
Dislike extremely	0	0	1	10	0	0	0	0
Dislike very much	0	0	1	10	1	10	3	30
Dislike moderately	0	0	0	0	0	0	0	0
Dislike slightly	1	10	1	10	3	30	1	10
Neither like nor dislike	0	0	1	10	0	0	0	0
Like moderately	4	40	4	40	3	30	5	50
Like very much	5	50	1	10	3	30	1	10
Like extremely	0	0	1	10	0	0	0	0
Total	10	100	10	100	10	100	10	100
Appearance								
Dislike moderately	0	0	1	10	0	0	0	0
Dislike slightly	1	10	1	10	0	0	2	20
Neither like nor dislike	1	10	2	20	0	0	0	0
Like moderately	3	30	0	0	3	30	4	40
Like very much	3	30	6	60	6	60	1	10
Like extremely	2	20	0	0	1	10	3	30
Total	10	100	10	100	10	100	10	100

Juiciness

All the panelists ranked chuck from pigs weaned at 21 days to be more juicy (*i.e.*, between like moderately and like extremely) while chop and sirloin were ranked less juicy with 50% each (Table 1). Again, 90% of the panelists ranked chuck from pigs weaned at 28 days to be more juicy compared to the chop, sirloin and pork leg which were ranked less juicy with 60% each (Table 4). Chuck and sirloin from pigs weaned at 35 days had 50% ranking each in terms of juiciness while chop was ranked 20% suggesting that the chop was the driest meat cut



(Table 3). FAO (2001) stated that dark, firm and dry (DFD) meat results from depletion of glycogen during handling, transportation and pre-slaughter, thus giving rise to little production of lactic acid. On average, pork from pigs weaned at 28 days had the highest acceptability (67.5%) in terms of juiciness followed by 21 days and 35 days with 60% and 40%, respectively. This finding suggests that pork from pigs weaned at 35 days was drier compared to other weaning periods. Toscas *et al.* (1999) observed that cooking loss alone does not explain the entire variation observed in juiciness but that the biochemical–biophysical state of the water in the meat, *i.e.*, water mobility and distribution play a major role in meat juiciness. According to Naude (1985), the variation in juiciness can be due to the amount and distribution (marbling) of intramuscular fat. Fat sustains the sensation of juiciness during chewing because it stimulates secretion of saliva. For Ngapo *et al.* (2003), juiciness can be influenced by low quality protein diets leading to slightly drier meat.

Table 3: Sensory evaluation of pork from pigs weaned at 35 days of age

Parameters	Chuck		Chop		Sirloin		Pork leg	
	Frequency	%	Frequency	%	Frequency	%	Frequency	%
Texture								
Dislike extremely	0	0	1	10	0	0	0	0
Dislike moderately	1	10	1	10	0	0	0	0
Dislike slightly	0	0	2	20	1	10	1	10
Neither like nor dislike	0	0	0	0	1	10	1	10
Like moderately	1	10	5	50	4	40	6	60
Like very much	6	60	0	0	3	30	2	20
Like extremely	2	20	1	10	1	10	0	0
Total	10	100	10	100	10	100	10	100
Flavour								
Dislike very much	0	0	0	0	0	0	1	10
Dislike moderately	0	0	0	0	1	10	0	0
Dislike slightly	1	10	0	0	0	0	1	10
Neither like nor dislike	1	10	1	10	0	0	0	0
Like moderately	0	0	8	80	3	30	5	50
Like very much	6	60	0	0	4	40	2	20
Like extremely	2	20	1	10	2	20	1	10
Total	10	100	10	100	10	100	10	100
Tenderness								
Too tough	2	20	5	50	3	30	4	40
Just about right	2	20	2	20	5	50	4	40
Too tender	6	60	3	30	2	20	2	20
Total	10	100	10	100	10	100	10	100
Juiciness								
Dislike extremely	0	0	0	0	0	0	1	10
Dislike very much	0	0	1	10	0	0	0	0
Dislike moderately	3	30	3	30	0	0	1	10
Dislike slightly	1	10	3	30	5	50	2	20
Neither like nor dislike	1	10	1	10	0	0	3	30
Like moderately	0	0	2	20	3	30	2	20
Like very much	5	50	0	0	2	20	0	0
Like extremely	0	0	0	0	0	0	1	20
Total	10	100	10	100	10	100	10	100
Appearance								
Dislike slightly	1	10	0	0	0	0	0	0
Neither like nor dislike	0	0	1	10	0	0	1	10
Like moderately	4	40	3	30	4	40	4	40
Like very much	2	20	4	40	4	40	2	20
Like extremely	3	30	2	20	2	20	3	30
Total	10	100	10	100	10	100	10	100



Appearance

Table 1 shows that sirloin from pigs weaned at 21 days was less preferred (80%) in terms of colour, whereas chuck was the most preferred (100%). However, sirloin had 100% acceptability in terms of colour compared to chop (60%) (Table 2). According to Table 3, sirloin had the highest responses (100%) while chuck, chop and pork leg had 90% ranking each. On average pork from pigs weaned at 35 days had the highest preference (92.5%) in all cuts in terms of appearance followed by 21 days (87.5%) and 28 days (80%). In the present study, meat appearance ranged from light brown to brown. Walker (2000) stated that the colour range of light brown to tan colour is ideal for cooked pork. According to van Oeckel et al. (1999), the perception of colour is very dependent on the observer and hence it is important to know the value of relative objective colour measurements to the subjective judgement of acceptable colour. Colour perception plays a major role in the consumer evaluation of meat quality (Lanari et al., 1995) and as such appearance of meat influences its acceptance by consumers.

CONCLUSION

Weaning age appeared to influence the eating quality of pork. Generally, the panelists preferred pork from pigs weaned at 28 days in terms of texture, tenderness, juiciness, flavour and appearance followed by 21 days and lastly 35 days. It seemed that the appearance of pork was the most preferred attribute across all weaning periods.

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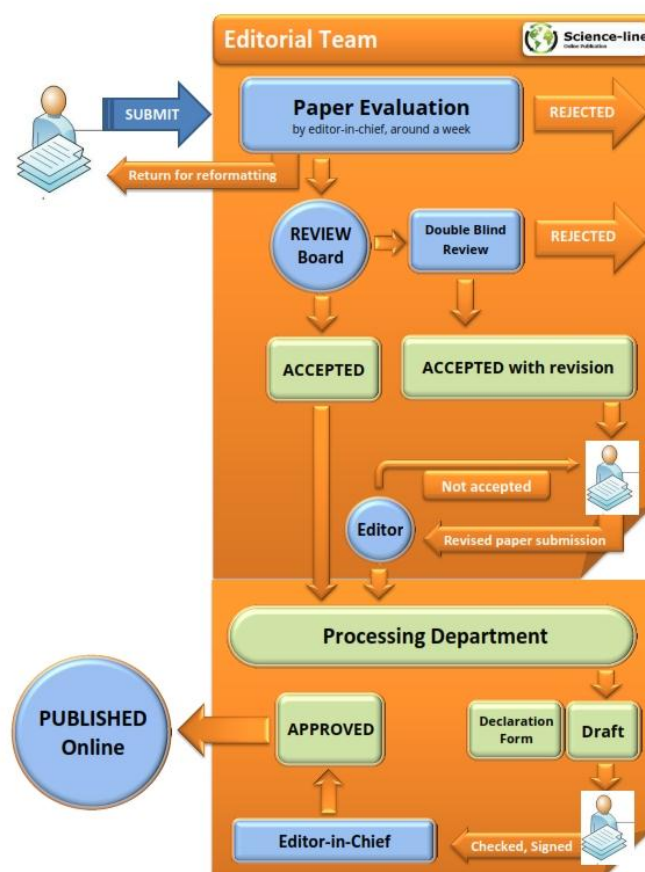
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