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Research Paper

Phenotypic Detection of Methicillin-Resistant *Staphylococcus Aureus* (MRSA) in Horses in Maiduguri, Borno State, Nigeria.

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ABSTRACT*: The Methicillin-resistant Staphylococcus aureus (MRSA) in Horses in Maiduguriwere determined. A total of 150 nasal swabs sample were collected from Horses for laboratory analysis, among the sample collected, 142 (94.7%) were positive for staphylococcus aureus, while 8 (5.3%) do not show the presence of staphylococcus aureus in the nasal flora on Mannitol salt agar (MSA) and Biochemical test. The result from this study revealed a total of 36 (24%) isolates were positive for MRSA and appeared intense blue on ORSAB media while 114 (76) were negative. The highest occurrence of 22 (14.7%) of MRSA in horses were recorded in Adult, although no statistically significant association observed with respect to Age ($p > 0.05$). Also this research revealed the highest occurrence of 20 (13.3%) in the horses from southern region of Maiduguri, with no statistically significant association observed in the occurrence of MRSA in the horses with respect to Region ($p > 0.05$). However, the prevalence of 15 (10%) and 21(14) were recorded from the male and female of horses respectively, there is statistically significant association observed with the occurrence of MRSA in horses with respect to Gender, with highest occurrence of 21(14%) in female ($p < 0.05$).*
KEYWORD:*Methicillin-resistant Staphylococcus aureus (MRSA), Oxacillin Resistance Screening Agar Base (ORSAB), and apparently healthy.*

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I. INTRODUCTION

Staphylococcus aureus (S. aureus) is one of the species of the genus staphylococcus. The organism was first discovered in the United Kingdom in 1961 (Fayomi *et al.*, 2011). *Staphylococcus aureus (S. aureus)* is normally found on the skin or in the nares of about 30% of healthy individuals. When *(S. aureus)* is present without causing symptoms it is called an infection (Fact sheet Review 2007). Multiple body sites can be colonized but the anterior nares are the most frequently colonized sites (Wertheim *et al.*, 2005). For humans, this organism is an important cause of foodborne intoxication, pneumonia, post-operative wound infections, and nosocomial bacteremia. *Staphylococcus aureus* is considered the most resistant of all non-spore forming pathogens, with well-developed capacities to withstand high salt concentrations (7.5-10%) (Talaro and Talaro, 2002). *Staphylococcus aureus* is known to be notorious in the acquisition of resistance to new drugs and continues to defy control measures (Ikaegwu *et al.*, 2008). Many strains of *S. aureus* that carry a wide variety of multi-drug resistant genes on their plasmids are known as methicillin-resistant *Staphylococcus aureus* (MRSA) (Ikaegwu *et al.*, 2008). Methicillin-resistant *S. aureus* is isolates of *S. aureus* which have acquired genes encoding antibiotic resistance to all penicillins including methicillin. The emergence of multiresistant staphylococcus and particularly methicillin-resistant Staphylococcus aureus (MRSA) has bring an attention on the need for a better understanding of the epidemiology of staphylococcal disease and the development of more effective methods for their treatment and control (DeLeo and Chambers 2009; Loeffler and Lloyd 2010).

In Maiduguri some research on MRSA have been conducted for some species but such research on horses is limited. Therefore detection of MRSA in horses is critical in order to prevent the spread of the infection to wider population.This paper is designed with aim of detection MRSA amongst *staphylococcus aureus* isolates in horse nares in Maiduguri. The result of the project will contribute to the knowledge require for improving the antibiotic usage in treating *Staphylococcus aureus* infection in horses, and may provide an insight for addressing Antimicrobial resistance challenges at local and national levels.

II. MATERIAL AND METHOD

2.1 STUDY AREA

The study was conducted in Maiduguri, Borno State which lies between latitude 10.20N and 13.40N longitude 9.80E and 14.40N with an area of 69,436 sq km located in the Northeastern corner of Nigeria sharing borders with Niger to the North, Chad to the Northeast and Cameroon to the east (Musa and Pindar, 2005). The State has Sahel vegetation in the North and a Sudan Savanna in the South.

2.2 SAMPLING TECHNIQUE

A stratified sampling technique was employed in this study, considering North and Southzonesof the state as strata. A total of 150 Nasal swabs samples were collected.Fifty (75)samples from horses in each strata within the study area using simple random sampling. Information on each of the sampled horses was recorded which include as age, sex, and zone name.

2.3 SAMPLE COLLECTION PROCEDURE

After putting on clean gloves, a sterile swab stick was introduced into the nostril of the Sampled Animals and rolled against the nasal mucosa and then replaced into the swab stick case. The swabs were aseptically transported in an ice pack container to the Department of Veterinary Medicine Laboratory and analysis was done according to Cheesbrough, 2002.

2.4 BACTERIAL ISOLATION AND IDENTIFICATION

Isolation of *S. aureus* was made by culturing the sample on Mannitol salt agar prepared according to the conventional technique. The cultured plate was incubated at 37^0C aerobically for 24 hours and thereafter examined for the presence of *Staphylococcus* like colonies as described by Cheesbrough (2002). The suspected *Staphylococcus* colonies (yellowish colonies from Mannitol fermentation) were selected and sub-cultured on to Mannitol salt agar. Colonies typical of *Staphylococcus* were confirmed using Gram staining, catalase and coagulase test.

2.5 OXACILLIN RESISTANCE SCREENING AGAR BASE (ORSAB)

ORSAB is used for the screening and isolation of methicillin-resistant *S. aureus* from clinical samples. MRSA routine detection methods are typically based on those described for humans, often involving selectiveenrichment broth and oxacillin-supplemented agar (Brown, D. F., et al.2005). The medium was allowed to reach room temperature, a colony of *Staphylococcus* was inoculated over the plate containing the media, and then incubated aerobically at 35^0C and examine after 24 hours for typical MRSA colonies. MRSA colonies appeared as intense blue colonies on the agar surface, the growth of other bacteria was inhibited but those able to grow on the media were typical colonies.

3.7 STATISTICAL ANALYSIS

Data generated were analyzed using IBM SPSS statistics software. Variables were assessed for association with MRSA colonization using Chi Square at 95% confidence interval for statistical association. Also tables and Graphs were used for data descriptions and presentations.

III. RESULT

Among the One hundred and fifty horse nasal swabs collected 142 (94.7%) were positive for *staphylococcus aureus*, while 8 (5.3%) do not show the presence of *staphylococcus aureus* in the nasal flora on Mannitol salt agar (MSA) and Biochemical test (Table 1). The result from this study revealed a total of 36 (24%) horses sampled were positive for MRSA and appeared intense blue on ORSAB media while 114 (76) were negative (Table 2). The highest occurrence of 22 (14.7%) of MRSA in horses were recorded in Adult, although this study has shown no statistically significant association observed with the occurrence with respect to Age. ($p > 0.05$) (Table 3). Also this research revealed the highest occurrence of 20 (13.3%) in the horses from southern region of Maiduguri, with no statistically significant association observed with the occurrence of MRSA in horses with respect to Region ($p > 0.05$) (Table 4). The prevalence of 15 (10%) and 21(14) were recorded from the male and female of horses respectively, however, there is statistically significant association observed with the occurrence of MRSA in horses with respect to Gender, with highest occurrence of 21(14%) in female ($p < 0.05$) (Table 5), thus female horses appeared to be more susceptible to MRSA than male horses in Maiduguri.

Table 1: Prevalence of S. aureus

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Positive	142	94.7	94.7	94.7
	Negative	8	5.3	5.3	100.0
	Total	150	100.0	100.0	

Table 2: Prevalence of MRSA

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Positive	36	24.0	24.0	24.0
	Negative	114	76.0	76.0	100.0
	Total	150	100.0	100.0	

Table 3: Occurrence of MRSA among horses with respect to Age (P= 0.185)

			Age			
			Young	Growing	Adult	Total
MRSA	Positive	Count	1	13	22	36
		% of Total	0.7%	8.7%	14.7%	24.0%
	Negative	Count	15	32	67	114
		% of Total	10.0%	21.3%	44.7%	76.0%
Total		Count	16	45	89	150
		% of Total	10.7%	30.0%	59.3%	100.0%

Table 4: Occurrence of MRSA among horses with respect to Region (P= 0.444)

			Region		
			North	South	Total
MRSA	Positive	Count	16	20	36
		% of Total	10.7%	13.3%	24.0%
	Negative	Count	59	55	114
		% of Total	39.3%	36.7%	76.0%
Total		Count	75	75	150
		% of Total	50.0%	50.0%	100.0%

Table 5: Occurrence of MRSA among horses with respect to Gender (P= 0.444)

			Gender		
			Male	Female	Total
MRSA	Positive	Count	15	21	36
		% of Total	10.0%	14.0%	24.0%
	Negative	Count	84	30	114
		% of Total	56.0%	20.0%	76.0%
Total		Count	99	51	150
		% of Total	66.0%	34.0%	100.0%

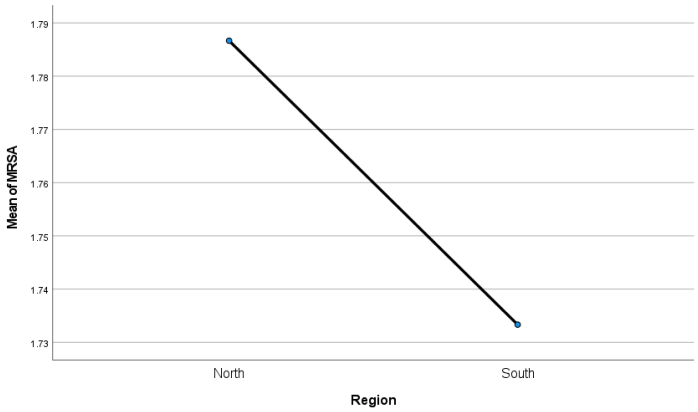

Figure 1. Mean plot of MRSA with respect to region

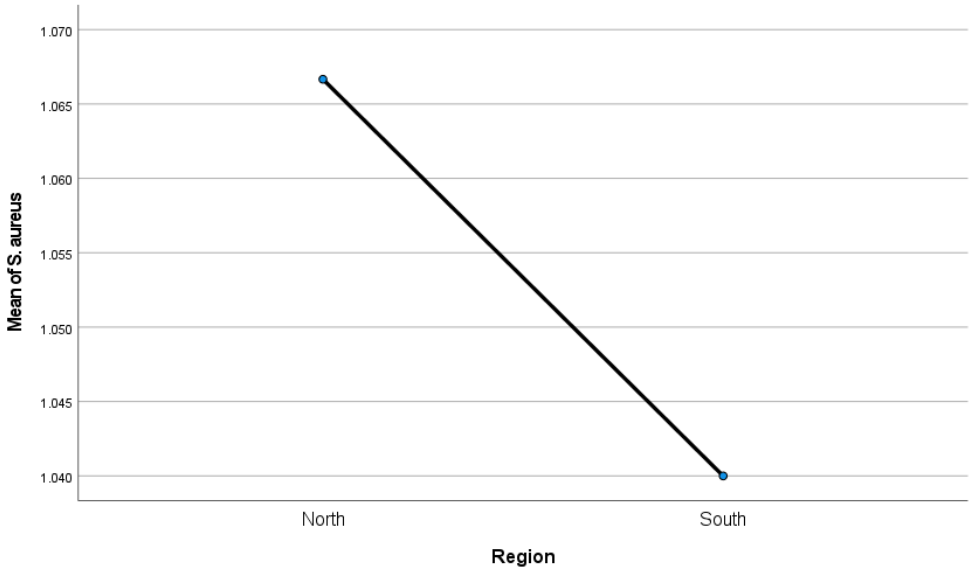

Figure 2. Mean plot of S. aureus with respect to region

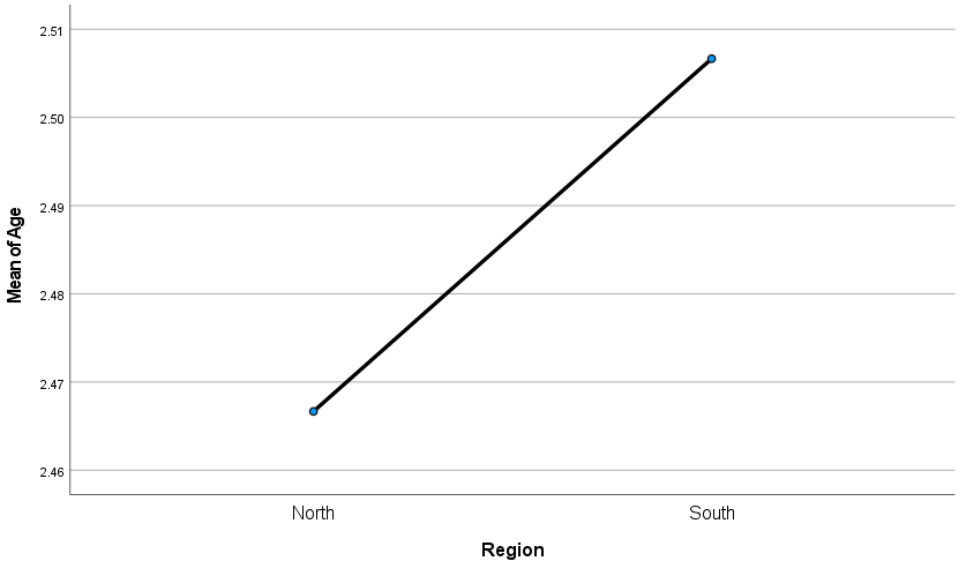

Figure 3. Mean plot of Age with respect to region

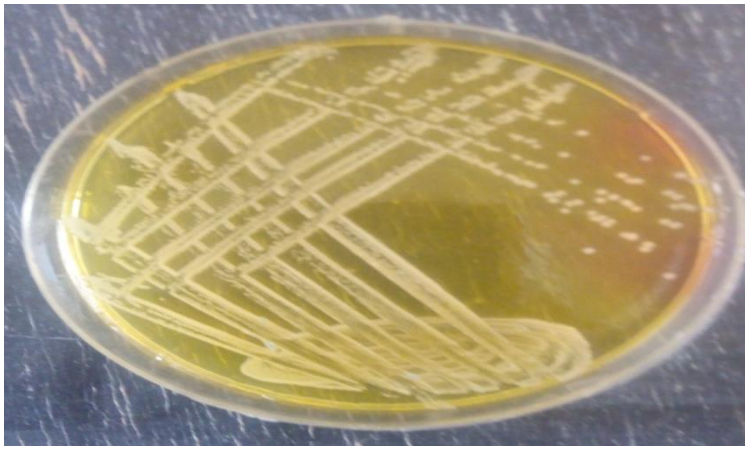

Figure 4: Photomicrograph of *Staphylococcus aureus* on manitol salt agar (MSA)

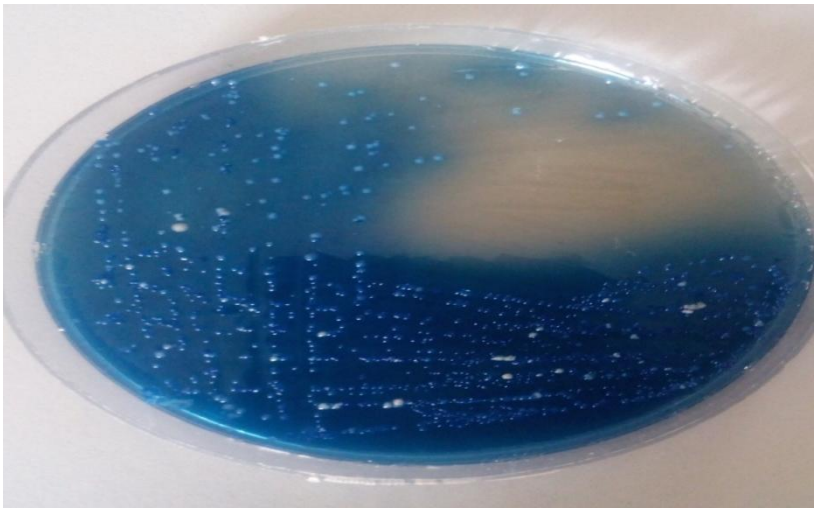

Figure 5: Photomicrograph of *Staphylococcus aureus* on Oxacillin Resistance Screening Agar Base (ORSAB)

IV. CONCLUSION

The phenotypic results in this study showed out of the 150 horses sampled 142 (94.7%) were positive which closely agree with the work of Suleiman et al. (2013) 83%, Owuna et al., (2015) 72.5%, and Kwoji et al. (2017) 72.1%. who reported occurrence of this bacteria in the same target population from the same study area. However, our finding is greater than that of Abdulrahman et al., (2018) and ***Kwoji et al.***, *(*2019)who reported 39.3% and 38.3% occurrence of *S. aureus* respectively. This variations could be due to differences in the type of sampled population and/or study period.It is concluded that MRSA are present in the nostrils of apparently healthy Horses in Maiduguri Local Government, Borno state, with the prevalence of 36%. There is need for further study on MRSA in horses in the study area to investigate the antimicrobial susceptibility and molecular characterization.

REFERENCES

[1]. Abdulrahman H. I., YA Geidam, MB Abubakar, MM Gashua A Gulani, HB Galadima. Phenotypic detection and antibiogram of *Staphylococcus aureus* from poultry processing units in Maiduguri, Borno State, Nigeria. Asian J Res Anim Vet Sci 2018; 1(1):1–8.

[2]. Brown, D. F., et al., Guidelines for the laboratory diagnosis and susceptibilitytesting of methicillin-resistant *Staphylococcus aureus* (MRSA). J. Antimicrob.Chemother. 2005. **56:**1000–1018.

[3]. Cheesbrough, M., District laboratory practice in tropical countries. ECBS edition.Cambridge University Press 2002. 2: PP.97-182.

[4]. DeLeo, F.R. and H.F Chambers. Reemergence of antibiotic-resistant Staphylococcus aureus in the genomics era. J Clin Invest 2009. 119, 2464–2474.

[5]. [5]. Fayomi, O. D., E. I. O. Oyediran, A. T. Adeyemo, and A. T Oyekale. "Prevalence and antibiotic resistance pattern of methicillin- resistant *Staphylococcus aureus* among in-patients at a tertiary health facility in Ido-Ekiti, Nigeria," *InternationalJournal of Laboratory Medicine*, 2011. 2 (4): 21-30.

[6]. John Hwa Lee DOI**:** 10.1128/AEM.69.11.6489-6494.2003

[7]. Ikeagwu, I. J., E. S. Amadi, and I. R Iroha, "Antibiotic sensitivity pattern of *Staphylococcus aureus* in Abakaliki, Nigeria", *Pakistan Journal of Medical Science*, 2008. 24: 230-235.

[8]. Kwoji ID, S, Jauro, JA Musa, YM Lekko, SI Salihu, HA Danchuwa. Phenotypic detection of methicillin-resistant *Staphylococcus aureus* in village chickens from poultry markets in Maiduguri, Nigeria. J Adv Vet Anim Res 2019. 6(2):163–7.

[9]. Kwoji ID, FM Tambuwal, MB Abubakar, Y Yakubu, AA Bitrus, S Jauro. Occurrence of methicillin resistant *Staphylococcus aureus* in chickens and farm personnel in Sokoto, North-western Nigeria. J Adv Vet Anim Res 2017. 4(3):255–60; https://doi.org/10.5455/ javar.2017.d220

[10]. Loeffler, A. and D.H. Lloyd, Companion animals: a reservoir for methicillin-resistant Staphylococcus aureus in the community, Epidemiol Infect, 2010. 138, 595–605.

[11]. Musa, A.H., T.Y. Pindar, Geological history of Borno State ministry of local government and chieftancy affair algon diary, 2005. pp:450.

[12]. Owuna G, RH Abimiku, IH Nkene, GW Joseph, O Ijalana. Isolation and antibiotic susceptibility of *Staphylococcus aureus* from Fresh Poultry Meat Sold in Keffi Metropolis, Nigeria. Int J Res Stud Biosci 2015. 3(11):1–5.

[13]. Rahimi F, M Katouli, MR. pourshafie.Characteristics of hospital and community acquired Methicillin resistant *Staphylococcus aureus* in Tehran, Iran. J med microbial. 2014. 63 (pt 6): 796-804.

[14]. Suleiman A, LT Zaria, HA Grema, P. Ahmadu. Antimicrobial resistant coagulase positive *Staphylococcus aureus* from chickens in Maiduguri, Nigeria. Sokoto J Vet Sci 2013. 11(1):51–5; https://doi. org/10.4314/sokjvs.v11i1.8

[15]. Talaro, P. Kathleen, and Arthur Talaro. *Foundations in Microbiology*. 4th ed. Boston: McGraw-Hill, 2002.

[16]. Wertheim, H. F., D. C. Melies, M. C. Vos, W. van Leeuwen, A. van Belkum, H. AVerbrugh. and J. L. Nouwen. The role of nasal carriage in Staphyiococcus aureus infections. *LancetInfectious Diseases*, 2005. 5: 751-762.