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RELATIONSHIP OF B-MODE ULTRASONOGRAPHY FEATURES OF THE MAMMARY GLAND AND TEAT WITH CALIFORNIA MASTITIS TEST FINDING AND SOMATIC CELL COUNT IN DAIRY BUFFALOES (*Bubalus bubalis Linnaeus, 1758)* WITH SUBCLINICAL MASTITIS

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ABSTRACT

The ultrasound appearance of the udder from buffalo with subclinical mastitis consisted of hypoechoic parenchyma with anechoic gland cistern, alveoli, and lactiferous duct. In the normal mammary gland of buffalo, the udder revealed hyperechoic parenchyma with interspersed anechoic alveoli and lactiferous ducts. Teat of buffalo with subclinical mastitis revealed homogenous hypoechoic features of the three-discrete layer of the parenchyma. The teat in animals without subclinical mastitis showed hyperechoic to hypoechoic features of the teat wall. The mean ± SD echo mean values of the mammary gland and teat with subclinical mastitis were relatively higher than the echo mean values in the mammary gland and teat without subclinical mastitis. Pearson's correlation showed a positive correlation between ultrasound echo mean values of the udder and CMT finding and ultrasound echo mean values of the udder and porta SCC. Also, the study showed a positive correlation between the ultrasound echo mean values of the teat and CMT findings and between the ultrasound echo mean values of the teat and SCC results. The results of the study suggest that ultrasonography can be useful in detecting subclinical mastitis in dairy Murrah buffaloes.

Keywords: buffalo, CMT, subclinical mastitis, ultrasonography

INTRODUCTION

The primary source of non-Ultra-high temperature (non-UHT) milk in the Philippines, where the dairy sector is still in its infancy, are the smallholder farms, which include carabao-2,855,087, cattle-2,391,723, and goat-3,761,682 (PSA, 2016). Phil-Murrah cows and Murrah buffalo cows produced substantially more milk on average (1,563 kg and 1,412 kg, respectively) than Philippine carabaos did (555 kg). The genetic characteristics of the Murrah buffalo's milk production may be responsible for the Phil-Murrah cows' 254% increase in milk production. Compared to dairy cattle, dairy water buffalo are more suited to the tropical environment of the Philippines. The information suggested that buffaloes might serve as an alternative milk source (Mamuad *et al.*, 2001). Dairy production had an increase

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in value of production of almost 17%, but volume output fell by 1.5%. At current pricing, total dairy output reached a level of 26.30 thousand metric tons and generated more than P1,208 million. Sixty-nine thousand six hundred sixteen dairy animals in the area produce milk002E

Mastitis is simply defined as the inflammation of the mammary gland. Mastitis comes in two varieties: subclinical and clinical. Clinical mastitis manifests as udder abnormalities, including redness, heat, soreness, and loss of function. When milk has flakes, clots, or is runny in consistency, it is also abnormal. There is no noticeable alteration in the udder or anomaly in the milk when mastitis is subclinical. Mastitis is characterized by physical, chemical, and bacterial alterations in buffalo and other dairy animals' milk, which lead to decreased production of both high-quality milk and milk in sufficient quantities (Sharif *et al*., 2009). Animals can get mastitis due to a variety of reasons such as age and the number of parities (Salvador *et al*., 2013); udder hygiene and type of management, milking machine vacuum level and overmilking, milking practices, genetic predisposition, udder conformation, stage of lactation, incomplete treatment of cases (Sharma *et al*., 2011).

With incidences varying depending on lactation stage (2.3%, 2.3%, and 12.9% in early, mid, and late lactation, respectively) and lactation season (40.90% and 25.71% in winter and summer months, respectively), subclinical mastitis is one of the most common and economically significant diseases in dairy herds (Kotb *et al*., 2014). On bacterial isolates, conventional culture is typically used to diagnose mastitis pathogens. This is followed by biochemical assays (Oliver *et al*., 2004). For identifying bacteria in milk, conventional microbiological techniques have been the gold standard (Gillespie and Oliver, 2005). However, in some cattle, such as heifers and dry cows, it is impossible to detect mastitis by these tests done on milk.

A non-invasive imaging technique called diagnostic ultrasound, often known as ultrasonography, provides dynamic information on the shapes and appearance of a specific organ (Rambabu *et al*., 2009). Modern ultrasound technology allows for a more accurate study of the udder morphology in breastfeeding animals, whether or not they have mastitis (Slosarz *et al*., 2010). According to Abd Al-Galil and Khalil (2016), ultrasonography of a buffalo's mammary gland without mastitis revealed homogenous hypoechoic parenchyma with anechoic blood arteries, milk alveoli, a lactiferous duct, and a gland cistern interspersed. In contrast, the breast parenchyma appeared homogenous with poorly defined milk alveoli and a lactiferous duct in the examination results for subclinical mastitis. The study's findings led the researchers to the conclusion that ultrasonography could offer a quick method for correctly diagnosing subclinical mastitis in buffalo.

The study evaluated the relationship between ultrasound features of the mammary gland and teat and California Mastitis Test (CMT) findings and Somatic Cell Count (SCC) in dairy buffaloes with subclinical mastitis. To describe and differentiate the B-mode ultrasound appearance and echo mean values of the mammary gland and teat of dairy buffaloes with or without subclinical mastitis; to determine the mean value of California Mastitis Test findings and somatic cell count in milk; and to evaluate the relationship of echo mean values of the mammary gland and teat with California Mastitis Test findings and somatic cell count.

MATERIALS AND METHODS

Three hundred (300) animals from the Philippine Carabao Center-National Head

Quarters (Gene Pool) were considered as the study population. Open epi software (at 95% confidence interval and 5% absolute precision) was used to calculate the sample population of 38 buffalo cows which were grouped based on pre-assessment conducted with the use of Milko Scan, Foss Machine to determine the somatic cell count in the milk. From 38 buffalo cows, 25 normal mammary glands and 45 mammary glands with subclinical mastitis were subjected to the study. The age of the animal subject to the study was 2-4 years old, with parity numbers ranging from 1-3. The feeding management approach employed was "cut and carry", wherein forages such as elephant grass, along with hay and concentrates, constituted the feeding regimen.

Somatic Cell Count was determined using a somatic cell counter (the Porta SCC® Quick Somatic Cell Count Test, Porta Check, Inc., Moorestown, USA) separately for milk samples from each quarter. Animals were considered positive for subclinical mastitis when SCC was $250\text{-}500\times10^3$/ml milk (Djabri *et al.,* 2002*).* The color change indicates the somatic cell count estimate of scores ranging from ≤100, 250, 500, 750, 1500, and ≥3000.

California Mastitis Test or CMT (California Mastitis Test Kit, Immucell®) was conducted, and a CMT score was given to each buffalo as negative, trace, 1, 2, and 3 with a somatic cell range of 0 to 200,000; 400,000; 1,200,000 and 5,000,000, respectively (McFadden, 2011). California Mastitis Test uses a four-compartment paddle with one compartment used per quarter. After putting the milk samples from each quarter, three drops of CMT reagent are added, and a circular movement of the paddle is done to mix the reagent and the milk. Changes in the viscosity of the mixture or gel formation in the paddle indicate the degree or level of somatic cells in the milk samples.

B-mode ultrasonographic examination was performed on each quarter and its corresponding teat using a portable ultrasound machine (TRIUP, Ultrasound Scanner-Veterinary using the model: TBLP-500) equipped with 3.5 MHz convex scanners. The examinations were performed in the sagittal plane by direct contact method after applying the ultrasound gel. The entire udder was examined on the lateral surface of each quarter along its longitudinal axis, moving upwards and downwards (Flock and Winter, 2006). The teat was examined, and the scanner was placed lateral to each teat.

Quantitative assessment of udder parenchyma and echogenicity (echo mean values) was conducted by image brightness analysis (on grayscale units from 0 (black) to 255 (white) with the use of dedicated software Image J (Image J, NACL Co. Ltd., Tokyo, Japan)) to obtain the mean gray value of the analyzed ultrasonographic image (Mostafa *et al.*, 2015).

Mean ± SD measurements were analyzed by ANOVA. Differences between means were compared by Tukey's HSD ($P<0.05$). The relationship between echo mean values of the mammary gland and teat with CMT finding and somatic cell count, on the other hand, was determined by Pearson's correlation ($P<0.05$).

The research methodology has been submitted to the University Institutional Animal Care and Use Committee, College of Veterinary Medicine, University of the Philippines Los Baños with Assigned Protocol Number CVM-2022-003 and has been approved accordingly.

RESULTS AND DISCUSSION

Figure 1 depicts the ultrasound image of a buffalo's udder with subclinical mastitis. Subclinical mastitis in buffalo causes the udder to have hypoechoic parenchyma, anechoic alveoli, and a lactiferous duct. This conclusion agrees well with that of Hussein *et al.* (2015)

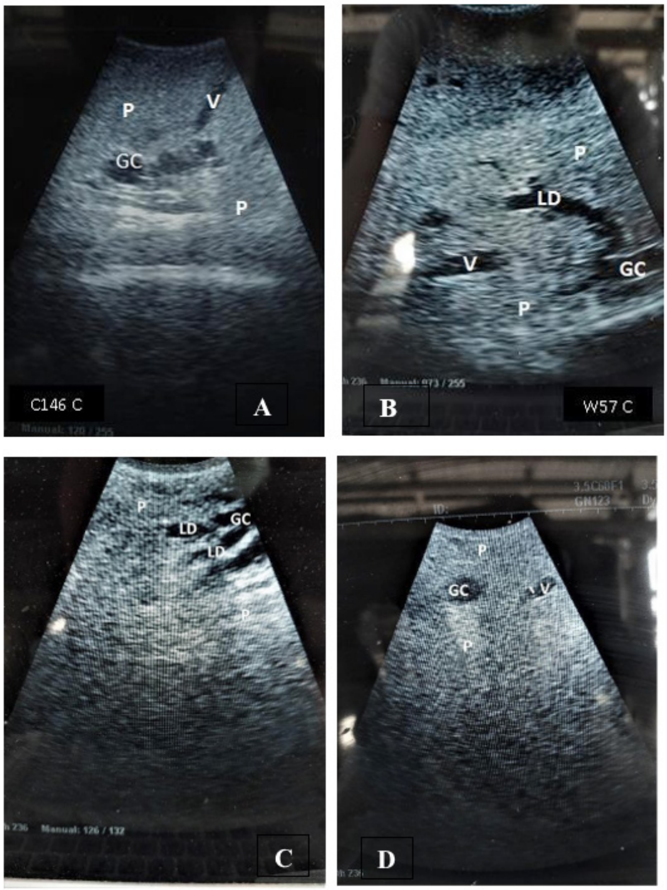

Figure 1. Ultrasound image of the mammary gland with and without subclinical mastitis. [A and B ultrasound images of the udder with subclinical mastitis show the hypoechoic-hyperechoic parenchyma (P), anechoic lactiferous duct (LD), gland cistern (GC), and vessels (V). Compared to images C and D, a normal udder, the parenchyma (P) has less echogenic (homogenous hypoechoic), whereas gland cistern (GC), lactiferous duct (LD), and vessels (V) are anechoic in appearance.]

and Kotb *et al.* (2014), who also noted that subclinical mastitis milk alveoli had anechoic fluid with suspended hypoechoic dots prior to milking and increased echogenicity after milking. The parenchyma of the udder with subclinical mastitis was described by Abd Al-Galil and Khalil (2016) as homogenous hypoechoic with poor visibility of milk alveoli and lactiferous duct. The gland cistern also developed mixed hypoechoic contents and lost its echogenicity. The parenchyma of the udder with mastitis emerged as hyperechoic in buffaloes in a prior study by Rambabu *et al.* (2009).

Buffalo with preclinical mastitis had ultrasonography of their udder that showed hypoechoic parenchyma with intermittent anechoic alveoli and a lactiferous duct. The lactiferous duct and milk alveoli cannot be seen clearly in the breast parenchyma, which appears hypoechoic, whilst the gland cistern lost its anechogeneity and mixed hypoechogenic contents (Abd Al-Galil and Khalil 2016). The mammary gland with mastitis shows non-homogenous in the mammary gland parenchyma, ranging from hypo to hyperechoic, according to a study by Fasulkov (2012). Additionally, the parenchyma of the udder with subclinical mastitis was hypoechoic with poorly defined milk alveoli and lactiferous duct in the study by Abd Al-Galil and Khalil (2016). Hussein *et al.* (2015) and Kotb *et al.* (2014) both backed up this conclusion.

The mammary parenchyma of a normal udder (Figure 1) showed intermediate echogenic features, which can be attributed to the connective tissue's equal distribution as an echogenic component and the udder parenchyma's lower echogenicity. The lactiferous ducts and anechoic alveoli are scattered throughout the uniform hypoechoic udder parenchyma. The vast homogenous anechoic area with hypoechogenic milk is also visible in the gland cisterns. These results matched those in the research of Hallowell (2012).

The gland cisterns revealed a sizable homogenous anechoic area with hypoechogenic milk, while the parenchyma of a typical udder looked homogenous hypoechoic. According to the study by Abd Al-Galil and Khalil (2016), the average buffalo mammary gland was

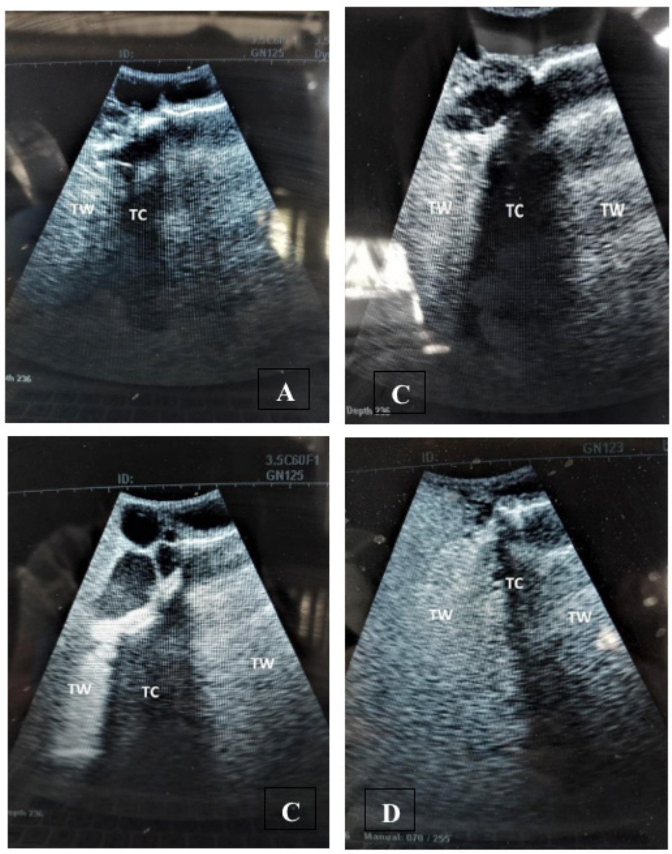

Figure 2. Ultrasound image of the teat with or without subclinical mastitis.

[Images A and B show the ultrasound features of the teat with subclinical mastitis; the teat wall (TW) is homogenous hypoechoic, while the teat cistern (TC) is hypoechoic to hyperechoic. Images C and D show the ultrasound features of the teat without mastitis; the teat wall (TW) reveals hyperechoic to hypoechoic, whereas the teat cistern (TC) has dilated anechoic area with hypoechoic dotes (milk).]

found to have homogenous hypoechoic parenchyma with anechoic blood vessels, milk alveoli, lactiferous ducts, and gland cisterns interspersed. The ultrasonography of healthy mammary gland parenchyma shows a homogeneous structure, according to numerous investigations (Flock, 2006; Franz *et al.*, 2003; Barbagianni *et al.*, 2017). The ultrasound features of the udder are amenable to sonographic imaging due to its superficial location hence, the technique has the potential to diagnose different diseases of the organ (Ragab *et al.,* 2016). The glandular parenchyma of the udder of buffalo appeared as homogenous and hyperechoic with anechoic alveoli (Rambabu *et al.,* 2008) similar to the udder of cows according to Ayadi *et al.* (2003).

Figure 2 depicts the subclinical mastitis teat's ultrasonographic appearance. The inner, middle, and outer layers are no longer differentiated and have been replaced with homogenous hypoechoic layers. Due to subclinical mastitis, the distinct three-layer of the teat parenchyma loses differences in echogenicity. Additionally, the lining contour of the lumen exhibits irregularity and the lumen's size diminishes. The teat sinus also seems to be anechoic to hypoechoic. According to a study by Abd Al-Galil and Khalil (2016), subclinical mastitis was characterized by irregular contour lining, homogenous hypoechogenic contents, a narrower lumen, a slightly thickened wall, and the absence of the recognizable three-layered appearance in the teat canal and cistern.

Due to subclinical mastitis, the teat lumen's contour lining was uneven, and the lumen's size shrank. In the study by Al-Galil and Khalil (2016), the teat cistern/sinus transitioned from being hypoechoic to anechoic; the teat cistern showed uniform hypoechoic, irregular contour lining, a narrower lumen, a little thickened wall, and the loss of the characteristics of the three-layered wall. Cows with subclinical mastitis have teat sinuses that are mildly hypoechoic (Javadi and Acorda, 2011). Also noted in the study by Kotb *et al.* (2014) were the uneven teat canal and sinus contour, disappearance of the three-layered teat walls, overlapped papillary duct, and rosette of Furstenberg in the buffaloes with subclinical mastitis.

Due to the teat wall's three distinct layers, the teat without subclinical mastitis had hyperechoic to hypoechoic characteristics (see Figure 2). A dilated anechoic characteristic and a few hypoechoic spots, which stand in for milk, can be seen in the teat cistern. Only when the teat cistern is full of milk is it classified as a dilated anechoic structure with few hypoechoic dots. Similar findings have been reported by Nak *et al.* (2005), Rambabu *et al.* (2008), Fasulkov (2012) and Kotb *et al.* (2014). The papillary duct appears as a single hypoechoic zone, and its boundary with the teat cistern exposes circular anechoic structures. In their work from 2012, Szencziova and Strapak demonstrated that the histological image of the teat wall appeared as a three-layered structure, with the teat skin layer on the outside, fibromuscular vascularization in the middle, and mucous membrane border on the inside. Ultrasonographically, the teat wall can be recognized due to the density differential. In the prior investigation, the teat wall appeared to have a three-layer structure. The muscle layer showed a larger, more homogenous, less echoic layer with anechoic cavities after the thick skin presented a bright, echoic line. A thin, brilliant line can be seen as the mucous membrane (Hospes and Seeh, 1999; Franz *et al.*, 2003).

Table 1 shows that the normal mammary glands' mean ± SD echo mean value was 56.937 ± 7.515, and the mammary gland with subclinical mastitis was 58.632 ±9.026. Additionally, the echo means value for a healthy teat was 56.22 ± 5.789, while the value for a teat with subclinical mastitis was 57.167 ± 8.883. As a result, subclinical mastitis in mammary glands and teats was associated with echo mean values that were considerably greater than those in healthy mammary glands and teats. The echo mean value of the udder cistern of cows with CMT readings of 2 and 3 was substantially greater than in cows without mastitis according to Javadi and Acorda (2011).

Table 1. Echo mean values of the mammary gland and teat, and mean values of CMT and porta SCC in dairy buffaloes with or without subclinical mastitis.

	Mean ± SD	
	Normal	**Subclinical**
Mammary Gland	56.94 ± 7.52	58.63 ± 9.03
Teat	56.22 ± 5.79	57.17 ± 8.88
CMT	0.00 ± 0.00	$2.82\times10^{6} \pm 2.22\times10^{6}$
Porta SCC	0.00 ± 0.00	851.46 ± 846.75

Compared to those without subclinical mastitis, the echo mean value of the udder and teat was comparatively greater. In comparison to the uninfected quarter, mastitis generally results in a higher level of heterogeneous echogenicity to the milk in the gland cistern and teat cistern (Radostits *et al.*, 2007).

Out of 70 udders tested using CMT and Porta SCC, 25 udders were verified as being subclinically mastitis-free using CMT and Porta SCC, whereas 45 udders were confirmed as being subclinically mastitis-positive. Results from CMT and Porta SCC were compared as well, but Porta SCC yielded negative results and vice versa. In comparison to the Porta SCC results, which had a total of 35 negative and 35 positive findings, the CMT results (Negative=35, SCC 100=4, SCC 250=12, SCC 500=0, SCC 750=7, SCC 1500=9, and SCC 3000=3) had a total of 28 negative and 42 positives. CMT had mean levels of $2.82\times10^{6} \pm 2.22\times10^{6}$ and 0.00 ± 0.00 for with and without subclinical mastitis, respectively. Meanwhile Porta SCC had mean values of 851.46 ± 846.75 and 0.00 ± 0.00 for with and without subclinical mastitis, respectively.

According to the study of Salvador *et al.* (2013), there is only a weak positive Pearson's coefficient-based association between the two tests (CMT and Porta SCC). The subjective interpretation of the CMT result and the color fluctuation of the Porta SCC result were both noted as potential factors that could affect the outcome. The pooling of milk samples is the alternative.

The somatic cell count and CMT scores were found to be positively correlated in the study by Costa *et al.* (2000) (r=1.000, p=0.0167). The two tests from cows and goats showed an 81% and 82% correlation, respectively, according to Hamann *et al.* (2010). Further, camel milk likewise revealed a substantial association between CMT score and SCC (Abdel Gadir Atif *et al.*, 2006).

Table 2 displays an association between CMT and Porta SCC, two echo textural features of the gland parenchyma that are positive. The P-value for Porta SCC was 0.011, and for CMT was 0.025. This finding indicates that subclinical mastitis in dairy buffaloes can be identified using the echo mean value and the ultrasonic characteristics of the mammary gland parenchyma. Four Pixel Standard Deviation qualities were highly significant and strongly correlated with SCC in the study by Zhang *et al.* (2022) between echo textural properties of the mammary gland parenchyma and milk obtained before and after milking ($p< 0.0001$). Table 2 also displays the findings of the Pearson association between the CMT, Porta SCC, and teat echo mean value. The connection between CMT:Teat and Porta SCC:Teat is found to be favorable. CMT vs. Teat had a P-value of 0.040, while Porta SCC vs. Teat had a P-value of 0.048.

Table 2. Pearson correlation coefficients between echo mean value of mammary gland parenchyma and teat parenchyma.

	Mammary Gland Parenchyma		**Teat Parenchyma**	
	R	***p*-value**	**R**	***p*-value**
CMT	0.137	<0.025	0.033	<0.040
Porta SCC	0.191	<0.011	0.006	<0.048

Pearson's correlation between ultrasound features of the udder with and without subclinical mastitis reveals a positive correction with CMT and Porta SCC results. A positive association between CMT results and echo mean value was demonstrated in the study by Javadi and Acorda (2011). This finding supported the hypothesis that the echo mean value of the udder with and without subclinical mastitis directly correlated with the CMT outcome. The SCC of the mammary gland parenchyma and the SCC of the milk obtained before and after milking also showed a highly significant and positive connection in the Zhang *et al.* (2022) investigation ($p< 0.0001$). The current study also demonstrates the positive link between the echo mean value of the teat and the results of the Porta SCC as well as CMT readings.

CONCLUSION

Therefore, in combination with CMT findings and Porta SCC results, ultrasonography can be beneficial in identifying subclinical mastitis in dairy buffalo (Murrah buffalo). Meanwhile, ultrasonography is capable of identifying subclinical mastitis in dairy buffalo on its own. Additionally, farmers can use their ultrasound equipment to find dairy buffalo with subclinical mastitis.

REFERENCES

Abd Al-Galil ASA and Khalil AH. 2016. Ultrasonographic diagnosis of subclinical mastitis in buffalo (*Bubalus bubalis*). *Res Opin Anim Vet Sci* 6(9):261-267.

Abdel Gadir Atif E, Hildebrandt G, Kleer JN, Molla B, Kyule MN and Baumann MPO. 2006. Comparison of california mastitis test (CMT), somatic cell counts (SCC) and bacteriological examinations for detection of camel (*Camelus dromedarius*) mastitis in Ethiopia. *Berl Munch Tierarztl Wochenschr* 119(1-2):45-9.

Ayadi M, Caja G, Such X and Knight CH. 2003. Effect of omitting one milking weekly on lactational performances and morphological udder changes in dairy cows. *J Dairy*

Sci 86(7): 2352-2358.

Barbagianni MS, Mavrogianni VS, Vasileiou NGC, Fthenakis GC and Petridis I. 2017. Ultrasonographic examination of the udder in sheep. *Small Ruminant Res* 152:86–99.

Djabri B, Bareille N, Beaudeau F, and Seegers H. 2002. Quarter milk SCC in infected dairy cows: a meta-analysis. *Vet Res* 33(4): 335-57.

Fasulkov I. 2012. Ultrasonography of the mammary gland in ruminants: A review. *Bulg J Vet Med* 15(1):1-12.

Flock M and Winter P. 2006. Diagnostic ultrasonography in cattle with disease of the mammary gland. *Vet J* 171(2): 314–321.

Franz S, Holfmann-Parisot M, Gutler S and Baumgartner W. 2003. Clinical and ultrasonographic findings in the mammary gland of sheep. *N Z Vet J*. 51(5):238-243.

Gillespie BE and Oliver SP. 2005. Simultaneous detection of mastitis pathogens, *Staphylococcus aureus*, *Streptococcus uberis*, and *Streptococcus agalactaiae* by multiplex real-time polymerase chain reaction. J Dairy Sci 88(10): 3510-3518.

Hallowell GD. 2012. *Ultrasound of the bovine mammary gland and its role in mastitis control*. Retrieved on 15 June 2022 from http://www.positiveaction.info/pdfs/articles/dt11.5p41.pdf

Hamann J, Lind O and Bansal BK. 2010. Determination of on-farm direct cell count and biochemical composition of milk in buffaloes. *Rev Vet* 21:541-542.

Hospes R and Seeh CM. 1999. *Sonographie und Endoskopie an der Aitze des Rindes*. Stuttgart, Germany: Schattauer Verlagsgesellschatt mbH.

Hussein HA, El-Khabaz KAS and Malek SS. 2015. Is udder ultrasonography a diagnostic tool for subclinical mastitis in sheep? *Small Ruminant Res* 129:121-128.

Javadi T and Acorda JA. 2011. Ultrasound features and echo mean values of udder and teat in dairy cows with mastitis. *Philipp J Vet Anim Sci* 37(2):167-176.

Kotb EEZ, Abu-Seida AM and Fadel MS. 2014. The correlation between ultrasonographic and laboratory findings of mastitis in buffaloes (*Bubalus bubalis*). *Glob Vet* 13(1):68–74.

Mamuad FV, Mamuad VL, Cruz LC, Lorenzo ND, Domingo IJ, Duran PG, Harada H and Ohashi T. (1996). Lactation pattern of Philippine carabao, murrah buffalo and its crosses. *Proceedings of the Second Asian Buffalo Association Congress, Makati, Philippines*.

McFadden M. 2011. *California mastitis test and milk quality*. Retrieved on 15 June 2022 from https://www.canr.msu.edu/uploads/234/76581/cmt.pdf

Mostafa MB, Al-Akraa AM and Khalil AH. 2015. Ultrasonographic assessment of superficial digital flexor tendon (SDFT) core lesion treated with platelet rich plasma (PRP) in donkeys (*Equus sinus*). *Benha Vet Med J* 29(2):274-282.

Nak D, Nak Y and Keskin A. 2005. Diagnosis of various teat lesion in dairy cows by ultrasonography. *Indian Vet J* 82(11):1163-1166.

Oliver SP, González RN, Hogan JS, Jayarao BM, Owens WE and Owens W. 2004. *Microbiological Procedures for the Diagnosis of Bovine Udder Infection and Determination of Milk Quality*. 4th ed. Verona, Wisconsin: National Mastitis Council.

Philippine Statistical Authority [PSA]. 2016. *Dairy Industry Performance Report*. ISSN:2012-0664. Quezon City, Philippines.

Radostits OM, Gay CC, Hinchcliff KW and Constable PD. 2007. *Veterinary Medicine – A Textbook of the Diseases of Cattle, Horses, Sheep, Pigs and Goats*. 10th ed. Philadelphia, USA: Saunders.

Ragab GH, Seif MM and Qutp MM. 2016. Ultrasonography of the mammary gland in ruminants. *J Vet Med Res* 23(2):125-132.

Rambabu K, Sreenu M, Kumar RVS and Rao TS. 2008. Ultrasonography of the udder and mastitis teat in buffaloes: A comparison of four methods. *Buffalo Bull* 27(4): 269-273.

Rambabu K, Sreenu M, Kumar RVS and Rao TSC. 2009. Ultrasonography of the udder and teat in buffaloes. *Buffalo Bull* 28(1):5-10.

Salvador RT, Garcia AAA, Abes NS and Mingala CN. 2013. Correlation of California mastitis test and somatic cell count on milk of water buffalo cows in the Philippines. *Trop Agric* 90(3):153-159.

Sharif A, Chhalgari MU and Muhammad G. 2009. Mastitis control in dairy production. *J Agric Soc Sci* 5(3):102-105.

Sharma N, Singh NK, and Bhadwal MS. 2011. Relationship of somatic cell count and mastitis: an overview. *Asian-Aust J Anim Sci* 24(3):429-438

Slorasarz P, Wojtowski J, Bienlinska S, Frackowiak A, Ludwiczak A, Krzysewski J, Bagnika E, and Strzalkowska N. 2010. Machine induced changes of caprine teats diagnosed by ultrsonography. *Afr J Biotechnol* 9(50):8698-8703.

Szencziova I and Strapak P. 2012. Ultrasonography of the udder and teat in cattle: perspective measuring technique. *Slovak J Anim Sci* 45(3):96-104.

Zhang X, Ahmad MJ, An Z, Niu K, Wang W, Nie P, Gao S and Yang L. 2022. Relationship between somatic cell counts and mammary gland parenchyma ultrasonography in buffaloes. *Front Vet Sci* 9:842105.