

Ultrasound-guided versus blind intraarticular injection of the foot of Egyptian buffaloes (Bubalus bubalis): A pilot study

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ABSTRACT

This study was designed to compare the effectiveness of US-guided and blind IA injection techniques of buffaloes foot. Twenty adult buffalo cadaveric hind feet were randomly assigned to blind (n=10) and US-guided (n=10) injections of the fetlock, pastern, and coffin joints. Methylene blue (1%) and Iopamidol® 300 (5 ml) were used as indicative markers for IA injection. The same injection strategy was also used *in vivo* on 10 live sound buffaloes. The injection criteria were comparatively evaluated between the two injection techniques. The US-guided injection technique showed a significant increase in the injection parameters of the fetlock, pastern, and coffin joints in the foot of buffaloes compared with the blind technique. However, the difficulties of the injection and several trials were significantly higher in the blind IA injection than in the US-guided injection. The performance time was significantly shorter with the US-guided injection as compared to blind IA injection. Compared to the blind approach, US-guided injection had the highest specificity for intra-articular injection procedures at 86.66%. In conclusion, US-guided IA injections of buffaloes feet showed promising results in enhancing the quality of diagnostic and therapeutic IA injections compared to blind injections.

Keywords: Blind, Buffalo, Foot, Intraarticular, Ultrasonography

In buffaloes, foot lameness is a significant problem that leads to financial losses due to high treatment costs, reduced milk production, and impaired fertility. About 80% of buffalo lameness cases are caused by hindlimb foot lameness (Enting *et al.* 1997, El-Shafaey *et al.* 2019). Reducing foot lameness can enhance animal health and productivity, substantially benefiting producers and national economies (Ettema and Østergaard 2006, El-Shafaey *et al.* 2021).

Various techniques have been used to diagnose and treat digit affections (joint injuries) in animals, including intra-articular (IA) injection, which is a simple and cost-effective method in practice (Smith *et al.* 1998, Courtney and Doherty 2009, Baxter and Stashak 2011, Al-Akraa

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et al. 2014, Alsobayil et al. 2015, Abdellatif et al. 2018). Traditionally, IA injections are performed using anatomical structures to locate the needle's positive path. Improper IA injections can cause post-injection discomfort, crystal synovitis, hemarthrosis, articular infection, and cartilage degeneration (Sethi et al. 2005, Bellamy et al. 2006, McGarry & Daruwalla 2011, AlSobayil et al. 2021). Therefore, it is essential to identify appropriate tools for proper needle localization during IA injections.

Recently, various imaging techniques have been employed to enhance the accuracy of IA injections, including ultrasonography, fluoroscopy, computed tomography, and magnetic resonance imaging (Hamed et al. 2020). Ultrasound (US)-guided injections are widely used to diagnose orthopedic disorders and assist with needle guidance during interventional procedures, improving their accuracy (Louis 2008, Epis et al. 2008). Nevertheless, the clinical efficacy of US-guided injections compared with blind injections is still controversial. Thus, this study aimed to evaluate and compare the accuracy and efficacy of US-guided IA injection with blind IA injection of buffalo feet by discriminating the injection criteria.

MATERIALS AND METHODS

Buffaloes: At the rural slaughterhouse in Dakahlia Governorate, Egypt, hind feet were extracted from 20 healthy adult Egyptian buffalo (48±12 months and



Fig. 1. Sites of needle placement for blind IA injection of the fetlock (A), pastern (B) and coffin joint (C) of an adult buffalo hind limb foot.

weighing 425±75 kg) immediately following animal slaughter. In addition, 10 live buffaloes that were clinically and radiographically healthy were drawn for the *in vivo* study. The cadavers used for this study were euthanized for reasons unrelated to orthopedic disorders, and the live animals showed no signs of hind limb lameness or orthopedic issues. The Mansoura University of Animal Care and Use Committee (VM.R.22.10.17) approved this study.

Cadaveric study: The cadaver feet were randomly divided into two groups: blind (n=10) and US-guided (n=10) injection of the fetlock, pastern, and coffin joints (Fig. 1). In this study, the locations for IA injection in each joint of the 20 cadaveric feet were identified and carefully prepared. The sole of each foot specimen was comparable to the ground surface to imitate the weight-bearing situation of a sedated standing buffaloes. Injections were performed using the dorsal approach by a well-trained veterinary surgeon (EE) with expertise in IA injection and ultrasound (US) examination. For blind IA injection, the joint space related to the joint was palpated, and a 20-gauge needle (Med, Eldawlia ico, Egypt) was precisely inserted into the joint until a positive injection was confirmed. Once the surgeon was satisfied that the correct position had been achieved, 2.5 mL of 1% methylene blue solution mixed with an identical volume of Iopamidol contrast agent (Scanlux®300, Sanochemia Pharmazeutika AG, Germany) was injected into the target joint space. The US-guided injection was carried out using an ultrasound machine (Mindray DP-2200Vet., PR China) with a 7.0 to 10.0 MHz linear probe in transverse and longitudinal planes. The foot was clipped, cleaned with water, soaked in alcohol, and coated in acoustic coupling gel. Utilizing the same landmarks as the blind approach, a 20-gauge needle was inserted in-plane using US guidance and guided at a 45° angle to the skin about the examined joint.

The location of the needle tip concerning the target joint capsule was checked on ultrasonography and adjusted if necessary (Fig. 2). On the ultrasound image, fluid entering the joint space was visible as a dark fluid wave. One hour after IA injection, the specimens were labeled with

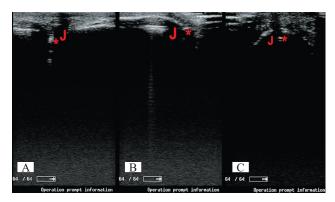


Fig. 2. Transverse ultrasonographic image for US-guided IA injection of fetlock (A), pastern (B) and coffin (C) joints of an adult buffalo hind limb foot. The needle tip is seen as hyperechoic interface (red asterisk) and positioned within the joint cavity of target joint (J).

numbers, packed in plastic bottles, and frozen for one week at -20°C to allow thorough staining of the joint capsule and associated pouches. Anatomical dissection of the stored specimens was then carried out with extensive care, and the occurrence of methylene blue in the synovial structures was reported and imaged. The presence of methylene blue in the joint after dissection confirmed a successful injection (Fig. 3A).

In vivo study: Ten healthy adult buffalo (n=10) were selected to compare the precision and reliability of US versus blind IA injection of the buffalo hind foot. The above mentioned technique was carried out on animals restrained with xylazine hydrochloride (Xylaject, Adwia, Egypt) at 0.05 mg/kg, IV. Each joint was processed in an aseptic manner and approached ultrasonographically (n=5) or blindly (n=5) with 5 ml of radiopaque Iopamidol contrast agent. Subsequently, lateral and dorsoplantar (DP) X-rays of each joint were taken to confirm the accuracy of the injection by a radiologist who was blind about the

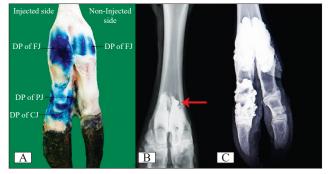


Fig. 3. A: Gross dissection of a buffalo hind limb foot following IA injection of the methylene blue dye showing successful staining of the sites for needle placement for IA injection of the fetlock, pastern, and coffin joint. Dorsalo-planter radiograph of a buffalo hind limb foot. B: Contrast dye remained outside joint pouches indicating inaccurate injection (arrow). C: Successful IA injection confirmed by presence of contrast dye within the dorsal and palmar joint pouches. DP of FJ: dorsal pouch of fetlock joint. DP of PJ: dorsal pouch of pastern joint. DP of CJ: dorsal pouch of coffin joint.

approach to injections. A radiography device with 70 kVp, 2.0 mAs, and a 70 cm focal film distance (Samsung-dong, SY-31-100-P, Seoul, Korea) was used for this purpose. The injection of the contrast agent revealed a small amount of resistance after the needle was successfully inserted into the joint area. Noticeable swelling of a joint pouch or fluid might be detected, followed by injection. A contrast agent in the studied joint on contrast arthrography was considered a confirmation of successful injection (Fig. 3B, C). Buffaloes were monitored for three days after the injection to check for any problems such as infection or hematoma.

Assessment of injection techniques: Individual practitioners evaluated the IA injection criteria. The experts assessed confidence in injection, judged and scored on a subjective scoring system for the ease of correct needle penetration, the difficulty of injection, number of trials, and performance time on a scale from 0 to 2 (Tables 1-4), according to El-Shafaey *et al.* (2017).

Statistical analysis: Statistical analysis was performed using the GraphPad Prism statistical software (GraphPad Prism for Windows, version 5.0, GraphPad Software Inc., USA). The Mann–Whitney non-parametric t-test was used to compare injection criteria scores between the two injection techniques. Furthermore, variations between the median and range were considered significant.

RESULTS AND DISCUSSION

In the present study, the anatomical markers defining the site of needle insertion for each joint were positively identified and accurately stained with methylene blue in all instances that was proven by cadaver division.

IA injection techniques with a high success rate are crucial

Table 1. Effect of correct penetration on the injection scores for intraarticular injection of the buffaloes' foot

Technique	Target joint		
	Fetlock	Pastern	Coffin
Blind	0 (0-1) ^b	0 (0-1) ^b	0.5 (0-1) ^b
US-guided	2 (1-2) ^a	1 (1-2) ^a	2 (1-2) ^a
P value	0.0004***	0.0001***	0.001**

Correct penetration. 0: Poor, out of the target joint capsule; 1: Good, in the way but did not enter the target joint capsule; 2: Excellent, in the target joint capsule. $^{a \text{ and } b}$ Medians and ranges with different superscript letters at the same column are significantly different at P < 0.05.

Table 2. Effect of difficulty of injection on the injection scores for intraarticular injection of the buffaloes' foot

Technique	Target joint			
	Fetlock	Pastern	Coffin	
Blind	0.5 (0-1) ^b	0 (0-1) ^b	1 (0-1) ^b	
US-guided	2 (1-2) ^a	1 (1-2) ^a	2 (1-2) ^a	
P value	0.0009***	0.0002***	0.0006^{**}	

Difficulty of injection. 0: Difficult, several attempts with low confidence; 1: Moderate, several attempts until successful injection; 2: Easy, immediate and confident injection. $^{\rm a}$ and $^{\rm b}$ Medians and ranges with different superscript letters at the same column are significantly different at P < 0.05.

Table 3. Effect of number of trials on the injection scores for intra-articular injection of the buffaloes' foot

Technique	Target joint		
	Fetlock	Pastern	Coffin
Blind	0.5 (0-1) ^b	0 (0-1) ^b	1 (0-1) ^b
US-guided	2 (1-2) ^a	1 (1-2) ^a	2 (1-2) ^a
P value	0.0002^{***}	0.0003**	0.0001***

Number of trials. 0: five trials, 1: three trials, 2: one trial. $^{\rm a}$ and $^{\rm b}$ Medians and ranges with different superscript letters at the same column are significantly different at P < 0.05.

Table 4. Effect of performance time (mins) on the injection scores for intra-articular injection of the buffaloes' foot

Technique	Target joint		
	Fetlock	Pastern	Coffin
Blind	0 (0-1) ^b	0 (0-1) ^b	1 (0-1) ^b
US-guided	1.5 (0-2) ^a	1 (1-2) ^a	2 (1-2) ^a
P value	0.0058**	0.0005***	0.0013***

Performance time (min). 0: 15 min, 1: 10 min, 2: 15 min. $^{a \text{ and } b}$ Medians and ranges with different superscript letters at the same column are significantly different at P < 0.05.

for appropriately managing animal joint disorders (Blaser et al. 2012, Alsobayil et al. 2015). Although IA injection techniques for horses and cattle are commonly used for other animals, such as buffalo, the technique's success is often influenced by anatomical and genetic differences that affect limb conformation and joint alignment (Desrochers et al. 1997, Al-Akraa et al. 2014). However, searching for an accurate imaging modality for IA injection in veterinary practice is controversial. Therefore, the present study was designed to compare the efficacy and feasibility of US-guided injection techniques for IA injection of buffalo feet with the blind technique. According to the authors' data, this is the first study to use US-guided injection techniques for IA injection of buffalo feet.

The cadaveric study aimed to develop and confirm an appropriate method for puncturing the fetlock, pastern, and coffin joints in buffalo feet while minimizing the risk of accidental injury to the orthopedic, vascular, and neurological systems. Dissections were performed to ensure proper needle placement and evaluate potential harm to the surrounding structures. Before each injection, an arthrocentesis was performed to ensure correct needle insertion and avoidance of vascular structures. However, blood was aspirated from three live buffaloes using the blind technique. Similar findings were reported by Hamed *et al.* (2020) and AlSobayil *et al.* (2021).

In the current study, IA injection into the target joints was safely accomplished using the dorsal approach. This is due to the anatomical connection between the lateral and medial synovial compartments in the buffalo's distal plantar portion of the fetlock joint. Our results are consistent with the findings of Desrochers *et al.* (1997) and Hamed *et al.* (2020). The pastern joint can be easily punctured by the dorsal approach by placing the needle far from the lateral branch of the long digital extensor tendon, as defined by

Alsobayil *et al.* (2015, 2021). Moreover, the coffin joint of buffaloes can be easily punctured through the dorsal approach with a needle implanted at a 45° angle ~ 1.5 cm above the coronet. These results are similar to those found in cattle (Van Amstel and Shearer 2006).

Using US-guided IA injection resulted in better outcomes across all injection measures and greater specificity than the blind method in this study. This might be attributed to the positive non-invasive visualization of the joint cavity and the ideal needle position inside the cavity, both of which increase accuracy and reduce the time required for IA. These results are consistent with those of Sites *et al.* (2007) and Shilo *et al.* (2010).

The live investigation aimed to eliminate post-mortem changes in cadaveric specimens and demonstrate critical parameters, including the mood, discomfort, and behaviour of live buffaloes during the injection. Apart from faster synovial aspiration preceding IA injection in live buffaloes, no significant differences were observed between IA injection procedures in cadavers and live animals. This could be attributed to the absence of weight-bearing outcomes in cadaveric limbs. Our results align with those of Piccot-Crézollet *et al.* (2005) in horses and Alsobayil *et al.* (2015 & 2021) in camels.

Comparing US-guided injection techniques to blind injection procedures, sensitivity 86.66%, specificity 46.60%, odds ratio 0.176, confidence interval 0.049–0.628 and P value 0.011. There was a significant increase (P<0.05) of the injection parameters in the US-guided injection of the fetlock, pastern, and coffin joints in buffalo's foot compared with the blind technique. The injection criteria's median and range scores in both injection procedures are listed in Tables 1-4.

Needle insertion and localization are the obstacles to a safe and successful US-guided IA injection method (Sites *et al.* 2007). Our study showed that US-guided injections were significantly more accurate than blind injections. This could be attributed to the feasibility of US-guided injection in directing the needle to the joint space and avoiding vital structures by visualizing the needle tip and fluid diffusion during IA injection. Our findings were consistent with Sites *et al.* (2007), Maecken and Grau (2007), and Rabba *et al.* (2011).

Non-guided IA injection, relying on the palpation of surface anatomic markers, is challenging. In the present study, blind techniques significantly increased IA injection difficulty (p<0.05) compared to US-guided techniques. This might be due to difficulty in correctly identifying the anatomic location for needle insertion, leading to improper needle placement and insufficient IA injection (Badawy and Eshra 2015). Thus, the present study provide a reference base for using US-guided injection for IA injection of buffalo feet as a model for refining IA injection methods in large ruminants.

An ideal sonogram is critical for proper US-guided arthrocentesis, and ultrasound image quality highly influences the number of injected joints. In this study, the average number of trials required to inject the joint successfully with US-guided techniques was significantly lower than with blind injection techniques. This could be attributed to the small number of low-quality ultrasound images. Similar findings were reported in horses (Nottrott *et al.* 2017).

In our investigation, the time required for IA injections of target joints was significantly lower with US-guided than blind methods. This finding is consistent with that of Moyer *et al.* (2007) and Shilo *et al.* (2010). However, the regular time for injecting the infraspinatus bursa (IB), bicipital bursa (BB), and scapulohumeral joint (SHJ) in horses using US-guided methods was significantly longer. This was thought to be directly related to the operator's lack of expertise (Schneeweiss *et al.* 2012).

In conclusion, US-guided IA injection in buffalo feet has shown promising results in improving diagnostic and therapeutic IA injections and clinical outcomes compared to blind injections. Not only does it improve injection accuracy in the target joint, but it also reduces side effects and procedure time. Most US-guided joint injections are straightforward techniques that are easy to learn and can be used in field conditions.

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