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Effect of steroids as an adjunct to local anesthetics in conventional and ultrasound guided brachial plexus nerve block in calves

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Abstract
The powerful anti-inflammatory as well as analgesic property of steroids through inhibition of phospholipids A_2 and by blocking transmission in nociceptive C-fibers but not in myelinated A-beta fibers have been well documented in the literature. We tried to investigate the effect of isoflupredone acetate as an adjuvant to 2% xylocaine hydrochloride. Twenty four young cow calves were divided randomly in 4 groups A, B, C and D. In group A, 20 ml of 2% Xylocaine hydrochloride was injected as in a traditional manner without under ultrasound guidance. In group B, the total dose of local anaesthetic was decreased and the calves were injected 18ml of 2% xylocaine hydrochloride mixed with 2ml (4mg) isoflupredone acetate. In groups C and D, brachial plexus nerve block was performed under the guidance of ultrasound scanner by using 20ml of 2% xylocaine hydrochloride and 18ml of 2% xylocaine hydrochloride mixed with 2ml (4mg) isoflupredone acetate respectively. The onset and duration of sensory/motor blockade parameters were observed at time interval 5, 10, 15, 20, 30, 45, 60, 75, 90, 120, 135, 150, 165 and 180 min after local anesthetic injection. The decrease in the total dose of local anesthetic did not significantly decrease the onset and duration of anesthesia indicating analgesic effect of isoflupredone. Hence isoflupredone acetate can be used as an adjuvant to local anesthetic for prolonging the duration of anesthesia.

Keywords: brachial plexus, corticosteroids, isoflupredone acetate, xylocaine hydrochloride, calves, sensory block, motor block

1. Introduction
Steroids have a powerful anti-inflammatory, analgesic effect and the topical application of corticosteroids cause skin vasoconstriction. The effect of vasoconstriction by steroids is mediated by occupancy of classical glucocorticoid receptors rather than by nonspecific pharmacological mechanisms [1, 2]. The effect of steroids as per the traditional theory of steroid action is mediated by their binding to intracellular receptors and modulation of nuclear transcription. Dexamethasone was used as an adjuvant with local anesthetic produced a relatively rapid effect which cannot be explained by the above mechanism [3]. Therefore, vasoconstriction, the presumed mechanism of action for epinephrine's adjunctive effect on local anesthetics, is probably not responsible for block prolongation by dexamethasone. The local effect of corticosteroid on the nerve may be responsible for relative rapid analgesic property [4]. The systemic effects of dexamethasone may be responsible for prolongation of local anesthetic block because they suppress inflammation through inhibition of phospholipase A_2. Methylprednisolone when applied locally has been found to block transmission in nociceptive fibers and the effect was reversible, suggesting a direct membrane action of steroids [5].

Regional anesthesia as nerve blocks is preferred over general anesthesia in ruminants due to obvious inherited problems and it serves as a multimodal approach to pain management in these animals. It involves minimally invasive procedures that don't impair patient consciousness and have aa negligible effect on cardiovascular and respiratory systems and reduced surgical stress [6]. Peripheral nerve blocks provide effective anesthesia of areas of the body innervated by individual plexuses which is accomplished by inhibiting nerve conduction using local anesthetics. The low mortality and morbidity rates in nerve blocks [7], little necessity for tracheal intubation [8] and a long-lasting postoperative analgesia [9] makes regional anesthetic technique better than general anesthesia.

The surgical significance of upper extremity in cattle reveal that for the correction of conditions namely fracture of scapula, brachiocephalic abscess drainage, lymph node biopsy puncturing of shoulder joint, fracture of humerus, amputation of front limb, radial nerve paralysis and triceps tendon stretching reflex and knuckling demand, brachial plexus is one of the most popular and suitable block for performing such operations in the forelimb in cattle [10]. The brachial plexus is formed by ventral branches of spinal nerves cervical (C6, C7 and C8) and thoracic (T1) and is located between the ribcage and medial aspect of scapula cranial to first rib. Brachial plexus supplies motor and sensory fibres to the muscles and structures of the front limb. Brachial plexus block provide good anesthesia and analgesia but of shorter duration. So various adjuvants like opioids, clonidine, neostigmine, midazolam, etc. have been added to local anesthetics in brachial plexus block to achieve quick, deep and prolonged block, but the results are either inconclusive or associated with side effects [11].

Brachial plexus nerve block under ultrasound guidance is an emerging aspect of regional anesthesia that can be used to precisely deliver drug around a nerve thus reducing the number of painful pricks. Nerves appear as hypoechoic structures surrounded by a thin hyperechoic rim and containing discontinuous hyperechoic bands [12] whereas fat and muscles are seen as heterogenous, hypoechoic structures. Fluids appear anechoic whereas air is shown as bright, hyperechoic image. Ultrasonography requires the use of high frequency sound waves in range of 10-14MHz. Perineural injection of glucocorticoid along with local anesthetics is reported to influence the onset and duration of sensory and motor block [13]. Corticosteroids also suppress ectopic neuronal discharge [4]. Keeping in view the above scenario, study was undertaken to evaluate the effect of steroid isoflupredone acetate as an adjunct to 2% xylocaine hydrochloride.

2. Materials and Methods

Twenty four crossbred cow calves housed under similar managemental conditions since birth were divided randomly in 4 groups *viz.* A, B, C and D. Each group consist of 6 animals each, irrespective of sex with varying age of six months to one year formed the subjects of the study.

2.1 Brachial plexus block in group a animals

The group A included calves in which brachial plexus was blocked without the use of ultrasound using 20ml of 2% xylocaine hydrochloride. The area 10 cm cranial to the acromion process of the scapula and axillary lymph node was surgically scrubbed shaved and infiltrated 3 ml of local anesthetic solution to form insensitive wheel. A 16 cm, 18-gauge needle, was inserted through the desensitized area and pushed horizontally till it striked the lateral surface of the first rib, where approximately 10 mL of a 2% xylocaine hydrochloride solution was injected. The needle was then withdrawn 5 to 10 cm, and then its tip was redirected 3 cm more distal to the first injection site, where an additional 10 ml of the anaesthetic was deposited. Care was exercised that no air, blood, or cerebrospinal was withdrawn upon needle aspiration.

2.2 Brachial plexus block in group B animals

In Group B, the animals were restrained in lateral recumbancy, 18 ml of 2% xylocaine hydrochloride and 2ml (4mg) of isoflupredone was administered by using 16 cm 18 gauge needle as in group A.

2.3 Brachial plexus block in group C animals

The scapula and the area around the scapular region was surgically scrubbed and prepared aseptically. The animals were restrained in lateral recumbancy which was followed by application of copious gel over the prepared site. After standardization of procedure from different possible angles and borders of scapula and anatomical area in vicinity the window was identified. The exact area where from the brachial plexus was visible was by placing the transducer along the medial aspect of scapula over the triceps and latissimus dorsi muscle. The axilary lymph node was identified and the needle was inserted under the guidance of ultrasound scanner. The needle was slowly pushed forward above the level of axillary lymph node so that the bevel of the needle was nearer to the plexus close to the radial nerve and the anaesthetic agent was injected and its spread around the brachial plexus was clearly monitored on the screen of the ultra sound scanner**.** Ultrasonography was performed by using TELEMED CAB with a 5-10MHz linear transducer. 20 ml of 2% xylocaine hydrochloride was deposited near the brachial plexus in instalments to effect. The deposition of the anaesthetic agent at brachial plexus was monitored on USG screen.

2.4 Brachial plexus block in group D animals

In Group D, the procedure performed was same as that in group A. Ultrasonography was performed by using TELEMED CAB with a 5-10MHz linear transducer. The animals were subjected to brachial plexus blockade using a combination of 18ml 2% xylocaine hydrochloride plus 2ml (4mg) isoflupredone acetate which was deposited near the brachial plexus in instalments to effect and duly monitored on USG screen while the amount of agent was being deposited. A 3 point scale using pin prick test was used to assess sensory blockade of the musculocutaneous, median, radial and ulnar nerves in the corresponding dermatomal areas. Sharp pin sensation felt at the skin suface was given grade 0, a null response to needle inserted into the skin was given grade 1 and absence of response to needle inserted through the skin was allotted grade 2. For assessment of motor blockade a grade 0 was assigned to animals showing normal motor function/normal gait while walking or no abnormal sign while standing., a grade 1 for animals that can walk while bearing mild to moderate weight and no abnormal sign while standing and grade 2 for animals which were not able to bear weight. These effects was done by different investigators with no information about the limb that has been blocked and the score was determined by their consensus. Grade 1 score was considered as onset of sensory and motor blockade with Grade 2 as their peak. The duration of sensory block was defined as the time interval between the onset of sensory block and the first postoperative pain. The time interval between the onset of motor block and complete recovery of motor functions was considered as duration of motor block.

2.5 Statistical analysis

The data was statistically analyzed by Dunken's Multiple Comparison Test using software SPSS 11.5 and inferences were drawn.

3. Results

The animals of group A were subjected to anesthesia of brachial plexus in conventional manner at the site 10 centimeter cranial to acromian of scapula. The dose regimen for inducing anesthesia was 20 ml 2% Xylocaine hydrochloride. The onset of brachial plexus blockade was marked by anesthesia of Radial nerve and was recorded at 20 minute post injection graded 0.17±0.17 thereafter the anesthetic effect increased to 1.17±0.17.It significantly (p<0.05) increased at 45 to 75 minutes (2.00±0.00). Thereafter the anesthetic effect declined to 1.00±0.34, 0.83±0.17,0.67±0.21 at 90,105 and 120 minutes respectively. No anesthetic effect was evidenced at 135 minutes and onwards. On comparative basis the anesthesia in this group was induced later compared to group C and D (at minute 15). As for the depth of anesthesia was concerned it was more in the animals of group D at 15 minutes (1.00± 0.00). The depth of anesthesia was significantly p<0.05 lower in this group A and B at 20 minutes (0.17±0.17) compared to the depth of anesthesia in animals of group C and D (1.83±0.17 for group C and 2.00±0.00 for group D) respectively. The duration of anesthesia was significantly lower in group A and the effect got terminated 120 minutes onwards however the anesthetic effect lasted up to 165 minutes in the animals of group C and D respectively (Table 1).

The ulnar and median nerve blockade was observed at 20 minutes after injection of anesthesia in the animals of group A and B (Table 2 and 3). The depth of anesthesia was appreciated for both the nerves at 30 minutes, 1.17±0.17 for ulnar and 1.33±0.21 for median nerve respectively. The effect significantly (p<0.05) increased at 45 minute to 60 minute in the nerves ulnar and median respectively and was maximum at this interval during the stipulated time of study. Thereafter the decline followed the same trend at 90 and 105 minute (1.67±0.21) and 0.83±0.17 for both ulnar and median nerves. The anesthesia lasted till 120 minutes for both the nerves in group A and thereafter at no interval of observation the anesthetic effect was detected on comparative basis. The duration of anesthesia for ulnar and median nerve lasted for 100 minutes only which was significantly (p<0.05) less than the duration of anesthesia observed for the animals of group C and D respectively. The signs of musculocutaneous nerve desensitization started at 20 minute (0.83±0.17).The level of anesthesia was comparable to that in the animals of group C at 20 minute interval, however was significantly higher than in the animals of all other groups at this interval during the course of study. The peak effect of musculocutaneous nerve desensitization was recorded at 45 and 60 minutes post injection (2.00±0.00). The effect non-significantly p>0.05 decreased to 1.83±0.17, 1.67±0.21 at 75, 90 and then significantly p<0.05 decreased to 0.83±0.17, 0.83±0.17 at 105 and 120 minutes respectively. No anesthetic effect of musculocutaneous nerve was recorded 120 minutes of anesthetic injection in group A (Table 4).

The signs of motor block in group A appeared at 20 minute with the mean value of 0.50±0.22.It showed peak effect at 60 to 90 minute (2.00±0.00) there after the effect showed the declined trend with the mean values 1.50±0.22, 1.33±0.21, 1.71±0.31, 0.50±0.22, 0.17±0.17 at 105, 120, 135, 150 and 165 minutes respectively. The effect of motor block could not be detected 165 minute of anesthetic administration (Table-5).

Table 1: The Mean ± SE scoring values of radial nerve desensitization in different groups at various intervals of brachial plexus block in calves

Group	Time															
	0	**5**	**10**	**15**	**20**	**30**	**45**	**60**	**75**	**90**	**105**	**120**	**135**	**150**	**165**	**180**
A	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00± 0.00^{Ca}	0.17± 0.17^{Ba}	1.17± 0.17^{Bbc}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	1.50± 0.34^{Ab}	0.83± 0.17Bce	0.67± 0.21^{Be}	0.00 ± 0.00^{Ba}	0.00 ± 0.00^{Ba}	0.00 ± 0.00^{Ba}	0.00± 0.00^{Aa}
B	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00± 0.00^{Ca}	0.17± 0.17^{Ba}	1.17± 0.17^{Bb}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.83± 0.17^{Ac}	1.83± 0.17^{Ac}	1.00± 0.00^{Bdb}	0.83± 0.17^{Bda}	0.17± 0.17^{Ba}	0.00 ± 0.00^{Ba}	0.00 ± 0.00^{Ba}	0.00± 0.00^{Aa}
C	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.83± 0.17^{ABb}	1.83± 0.17^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.17± 0.17^{Ad}	0.83± 0.17^{Ab}	0.00± 0.00^{Aa}
D	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	1.00± 0.00^{Ab}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.67± 0.21^{Ba}	1.00± 0.00^{Ab}	0.00± 0.00^{Aa}

Different small superscript indicate significant difference (p<0.05) within the groups
Different capital superscript indicate significant difference (p<0.05) among the groups

Table 2: The Mean ± SE scoring values of ulnar nerve desensitization in different groups at various intervals of brachial plexus block in calves

Group	Time															
	0	**5**	**10**	**15**	**20**	**30**	**45**	**60**	**75**	**90**	**105**	**120**	**135**	**150**	**165**	**180**
A	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00± 0.00^{Ca}	0.50± 0.22^{Bb}	1.17± 0.00^{Bc}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	1.83± 0.17^{Ad}	1.67± 0.21^{Ad}	0.83± 0.17^{Bbc}	0.50± 0.22^{Bb}	0.00 ± 0.00^{Bb}	0.00 ± 0.00^{Ba}	0.00 ± 0.00^{Ca}	0.00 ± 0.00^{Aa}
B	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00± 0.00^{Ca}	0.83± 0.17^{Bb}	1.50± 0.22^{Cc}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	1.83± 0.17^{Acd}	1.67± 0.21^{Acd}	1.67± 0.21^{Acd}	1.00± 0.00^{Cb}	0.17± 0.17^{Ba}	0.00 ± 0.00^{Ba}	0.00 ± 0.00^{Ca}	0.00 ± 0.00^{Aa}
C	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.67± 0.21^{ABb}	1.67± 0.21^{Ac}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Acd}	1.17± 0.17^{Ae}	0.67± 0.21^{Ab}	0.00 ± 0.00^{Aa}
D	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.83± 0.17^{ABb}	1.83± 0.17^{Acd}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Bc}	1.67± 0.21^{Ad}	1.00± 0.00^{Bb}	0.00 ± 0.00^{Aa}

Different small superscript indicate significant difference (p<0.05) within the groups
Different capital superscript indicate significant difference (p<0.05) among the groups

Table 3: The Mean ± SE scoring values of median nerve desensitization in different groups at various intervals of brachial plexus block in calves

Group	Time															
	0	**5**	**10**	**15**	**20**	**30**	**45**	**60**	**75**	**90**	**105**	**120**	**135**	**150**	**165**	**180**
A	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	1.00± 0.00^{Bb}	1.33± 0.21^{Bc}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	1.83± 0.17^{Aed}	1.67± 0.21^{Ae}	0.83± 0.17^{Bbf}	0.67± 0.21^{Bf}	0.00± 0.00^{Ba}	0.00± 0.00^{Ba}	0.00± 0.00^{Ca}	0.00 ±0.00^{Aa}
B	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00± 0.00^{Aa}	1.00± 0.00^{Bb}	1.67± 0.21^{ABcd}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.83± 0.17^{Ac}	1.50± 0.22^{Ad}	1.17± 0.17^{Bb}	0.67± 0.21^{Ce}	0.00± 0.00^{Ba}	0.00± 0.00^{Ca}	0.00 ±0.00^{Aa}
C	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.17± 0.17^{Aa}	1.17± 0.17^{ABb}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.17± 0.17^{Ab}	0.67± 0.21^{Ad}	0.00 ±0.00^{Aa}
D	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.00 ±0.00^{Aa}	0.67± 0.21^{Bbc}	1.67± 0.21^{Ac}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	2.00± 0.00^{Ad}	1.17± 0.17^{Ae}	1.00± 0.00^{Be}	0.00 ±0.00^{Aa}

Different small superscript indicate significant difference (p<0.05) within the groups
Different capital superscript indicate significant difference (p<0.05) among the groups

Table 4: The Mean ± SE scoring values of musculocutaneous nerve desensitization in different groups at various intervals of brachial plexus block in calves

Group	Time															
	0	**5**	**10**	**15**	**20**	**30**	**45**	**60**	**75**	**90**	**105**	**120**	**135**	**150**	**165**	**180**
A	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.83± 0.17^{Ab}	1.33± 0.21^{Bc}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	1.83± 0.17 ^{Ad}	1.67± 0.21 ^{Ad}	0.83± 0.17^{Bb}	0.83± 0.17^{Bbc}	0.00 ± 0.00^{Ba}	0.00 ± 0.00 ^{Ba}	0.00 ± 0.00 ^{Ca}	0.00 ± 0.00^{Aa}
B	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	1.00± 0.00^{Ab}	1.83± 0.17^{Acd}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	1.67± 0.21 ^{Ad}	1.17± 0.17 ^{Cb}	1.00± 0.00 ^{Cb}	0.00 ± 0.00 ^{Ba}	0.00 ± 0.00 ^{ca}	0.00 ± 0.00^{Aa}
C	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.83± 0.17^{Ab}	1.83± 0.17^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	2.00± 0.00^{Ac}	1.17± 0.17^{Ad}	0.67± 0.21^{Ab}	0.00 ± 0.00^{Aa}
D	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.00± 0.00^{Aa}	0.33± 0.21^{ABb}	1.17± 0.31^{Ac}	1.83± 0.17^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	1.00± 0.00^{Ac}	1.00± 0.00^{Bc}	0.00 ± 0.00^{Aa}

Different small superscript indicate significant difference (p<0.05) within the groups
Different capital superscript indicate significant difference (p<0.05) among the groups

Table 5: The Mean ± SE scoring values of motor block in different groups at various intervals of brachial plexus block in calves

Group	Time															
	0	**5**	**10**	**15**	**20**	**30**	**45**	**60**	**75**	**90**	**105**	**120**	**135**	**150**	**165**	**180**
A	0.00± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00 ^{Ba}	0.50± 0.22 ^{Bb}	1.00± 0.00 ^{Ac}	1.83± 0.17 ^{Ade}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	1.50± 0.22^{Aef}	1.33± 0.21 ^{Bcf}	1.17± 0.31 ^{Acf}	0.50± 0.22 ^{Cb}	0.17± 0.17 ^{Cba}	0.00 ± 0.00^{Aa}
B	0.00± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00 ^{Ba}	0.83± 0.17 ^{BbCc}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	1.50± 0.22 ^{Ae}	1.17± 0.17 ^{Bce}	1.17± 0.17 ^{Ace}	0.50± 0.22 ^{Cbf}	0.33± 0.21 ^{Cfa}	0.00 ± 0.00^{Aa}
C	0.00± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.67± 0.21 ^{Ab}	1.67± 0.21 ^{Ac}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	2.00± 0.00 ^{Ad}	1.83± 0.17 ^{Acd}	1.67± 0.17 ^{Ae}	1.00± 0.00 ^{Ae}	0.00 ± 0.00^{Aa}
D	0.00± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.00 ± 0.00^{Aa}	0.67± 0.21 ^{Ab}	1.67± 0.21 ^{Acd}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Ac}	2.00± 0.00 ^{Bc}	2.00± 0.00 ^{Bc}	1.50± 0.22^{Bd}	0.50 ± 0.22 ^{Bb}

Different small superscript indicate significant difference (p<0.05) within the groups
Different capital superscript indicate significant difference (p<0.05) among the groups

In the animals of group B, conventional brachial plexus block was done with the supplementation of Injection isoflupredone acetate, the blockade of radial nerve was rather similar as in group A with the mean value of 1.17 ±0.17 at 30 minutes interval. The onset of desensitization of this nerve was significantly p<0.05 delayed compared to the animals of group D in which the same combination and the onset of radial nerve desensitization was recorded at 15 minute and lasted till 165 minutes. The anesthetic effect for the radial nerve in this group of animals started a declining trend from 90 minute onward up to 135 minutes beyond which the anesthetic effect disappeared. Comparing the duration of anesthesia of radial nerve between this group and group D animals, the duration was shorter in group B (90 minutes) against 150 minutes in the animals of group D (Table 1). The desensitization of ulnar nerve in the animals of group B was recorded at 20 minute after injection with the mean value 0.83±0.17.The effect non- significantly (p>0.05) increased at 45, 60, 75, 90 and 105 minutes post injection with mean values 0f 2.00±0.00, 2.00±0.00, 1.83±0.00, 1.67±0.21 and 1.67±0.21 respectively. After that the anesthetic effect significantly (p<0.05) decreased at 120 minutes to 1.00±0.00.Thereafter the effect decreased to 0.17±0.17 at 135 minute beyond which the ulnar nerve was free from the influence of anesthetic effect. Comparing the radial nerve blockade of the animals in the same group, the onset of anesthesia for ulnar nerve was significantly higher and much earlier than the radial nerve and the duration of anesthesia of this nerve was shorter 90 minutes as compared to ulnar nerve blockade in the animals of group C (150minutes) and group D (150 minutes). However, no significant difference was observed between groups C and D in the duration of anesthesia (Table-2)

In the animals of group B, the desensitization of the median nerve and musculocutaneous nerve as marked by loss of sensation in the areas supplied followed a similar pattern in both the nerves. The desensitization started at 20 minutes and the peak effect was reached at 45 minutes (mean value 2.00±0.00) in both the nerves in this group. It remained at the same level up to 75 and 90 minutes for median and musculocutaneous nerve respectively. In both the nerves, the influence of anesthesia was over after 135 minutes i.e. 0.67±0.21 and 1.00±0.00 for median and musculocutaneous nerve respectively (Table 3 and 4). Motor blockade as was marked by the loss of weight bearing on the anesthetized limb was noted at 20 minutes post injection which significantly

$p<0.05$ increased from 30 minutes and continued for 90 minutes with a mean value (2.00±0.00). Thereafter, the motor block showed a declined trend towards the terminal intervals with the mean values 1.17±0.17, 0.50±0.22, and 0.33±0.21 at 135.150 and 165 minutes post injection beyond which no motor block was detected. As for the duration of motor block in the animals of group B are concerned, it is comparable to the animals of group A however it is lesser than the animals of groups C and D In the animals of group C, the brachial plexus blockade was done by injecting 20ml, 2% Xylocaine hydrochloride under ultrasound guidance using the site selected after standardization of scanning window. The site selected was near the caudal border and on the distal aspect of scapular area with the transducer placed on the long head of triceps muscle. A 5 inches 16 gauge needle was directed between thoracic wall and scapula through triceps above the level of axillay lymh node. The first nerve to get anesthetized and revealed desentization was radial nerve at 15 min (0.83±0.17) post injection. This anesthesia was noticed by inability to extend the segments distal to the revealed nerve with resultant development of dropped elbow and overextension of the limb without abduction of shoulder. The results were pronounced and maximum effects at 20 minutes (1.83±0.17) at 30 minutes (2.00±0.00) continued upto 135 minutes thereafter declined to 0.083±0.17 and normal reflexes returned at 180 minutes (Table-1). The ulnar nerve was desensitized at 15 minutes post injection, however the degree of desensitization was comparatively lesser (0.67±0.21) compared to radial nerve (0.83±0.17) in the same group. (Table-2).The desensitization of the median nerve as evidenced by the pinpricks started at 15 minutes (0.17±0.17) and the effect was significant at 20 minutes (1.17±0.17). The effect of anesthesia remain steadily highly significant ($p<0.05$) from 30 minutes to 135 minutes post injection thereafter the anesthetic effect decreased significantly at 150 minutes and the effected lasted upto 180 minutes post injection (Table-3). The desensitization of the medial aspect in the distal third of humerus marked the desensitization of musculocutaneous nerve which started at 20 minutes (0.83±0.17). It significantly increased at 20 to 30 minutes post injection interval (1.83±0.17). The effect increased to 2.00±0.00 and continued upto 135 minutes post injection, thereafter, it significantly ($p<0.05$) decreased at 150 and 165 minutes 1.17±0.17 and 0.67±0.21 respectively) and the anesthetic effect of musculocutaneous nerve got abolished at 180 minutes post injection (Table-4). In the animals of group C, the signs of motor block namely walking while bearing mild weight, abnormal posture, flexion of fetlock joint appeared at 15th minutes post injection (0.67±0.21).The effect was much pronounced clinically at 20 minutes (1.67±21) and it significantly remained higher from 20 to 120 minutes (2.00±0.00).Thereafter, it significantly decreased ($p<0.05$) to 1.83± 0.17, 1.67± 0.17 and 1.00±0.00 at 135, 150 and 165 minutes post injection respectively (Table-5). The dose regimen used in the animals of group C (20ml, 2% Xylocaine hydrochloride) was quite comparable to the groups A however was slightly more than in the groups where isoflupredone acetate was incorporated (groups D and B). The anesthesia was completely abolished at 180 minutes in this group. The application of transcutaneous electrical nerve stimulator at the calliberated frequency for 0.9 seconds revealed no response during the phase of anesthesia. However, the animals of this group responded to the stimulation by transcutaneous electrical nerve stimulation at the preset frequency for 0.9 seconds which confirmed the return of reflexes after 180 minutes post injection.

In the animals of Group D, the brachial plexus was blocked by injecting a combination of local anesthetic and a corticosteroid (2 % xylocaine hydrochloride + injection isoflupredone acetate 4 mg) using a 5 inches long, 16 gauge needle inserted at the same site and scanning window as in the animals of group C. In the animals of group D, among the nerves emerging from the brachial plexus, the radial nerve was the first to get desensitized which was evidenced by the signs elucidating radial paralysis type posture by the animal. The onset of effect of the nerve was noted at 15 minutes post injection (1.00 ± 0.00). The depth of desensitization significantly increased from 20 minutes post injection (2.00±0.00) and the anesthetic effect showed declining trend from 150 minutes post injection till 165 minutes (1.67± 0.21 and 1.00±0.00). The anesthetic effect got completely abolished at 180 minutes as was evidenced when the animal could walk without any difficulty and showing any abnormal gait. On comparison of the anaesthetic effect at various intervals in the groups C and D, although the radial nerve showed anesthetic effect at 15 minutes post injection, however the effect was significant in group D animals than in the animals of group C as the number of animals showing onset of sensory blockade in group D was more than in group C. Similarly, the levels of anesthesia were significantly higher in the radial nerve of group D, than group C at any hour of observation during the period of study (Table-1).

The anesthetic effect of the ulnar nerve followed the same pattern of desensitization at 15 minutes post injection which increased significantly ($p<0.05$) from minute 15 (0.83±0.17) to 20 minutes (1.83±0.17). After that the effect remained constant from minutes 30 to 135 post injection period (2.00±0.00). The effect showed declined trend from 150 minutes to 165 minutes (1.67±0.21 and 1.00±0.00 respectively). The anesthetic effect ended there after and at 180 minute post injection the animals were free from the effect of anesthesia as for the ulnar nerve is concerned. On comparative basis the anesthetic effect of ulnar nerve, although followed the same trend as for the duration is concerned yet the degree of desensitization was higher at any hour of the observation during the course of study (Table-2).

The desensitization of median nerve in the animal of group D followed the same trend as for the duration of anesthesia is concerned. It showed desensitization at 15 and 20 minutes post injection with mean values of 0.067±0.21, 1.67±0.21 respectively. The anesthetic effect significantly and steadily increased from 30 minutes post injection period and continued upto 135 minutes (2.00±0.00) thereafter the anesthetic effect declined to1.17 ±0.07 at 150 and 1.00±0.00 at 165 post injection period respectively and the anesthesia lasted thereafter so that towards the terminal period of observation the animal was free from anesthetic effect (Table-3). The anesthetic effect of musculocutaneous nerve showed mild desensitization (0.33±0.21) at 15 minutes and the effect significantly ($p<0.05$) increased to 1.83±0.17.The maximum effect of anaesthesia was from 45 to 135 minutes (2.00±0.00) thereafter the anaesthetic effect showed a declining trend from 150 to 165 minutes post injection and the effect was completely abolished at 180 minutes. As for the degree of desensitization at 45 to 135 minutes is concerned both the groups C and D followed the same patterns and the degree of depth (2.00±0.00), however the effect was rather less at 165 minutes in the musculocutaneous nerve of group A

(0.67±0.21) than in the animals of group D at the same hour (1.00±0.00). In the animals of group D motor blockade was not appreciated up to 10 minutes post injection. It appeared at 15 minutes and significantly increased up to 30 minutes post injection where from it showed a significantly a steadily depth of anesthetic motor block up to 150 hours. The depth showed a declining trend and decreased 1.50±0.22 at 165 minutes post injections, so much so that the motor block did not abolish by 180 minute of observation (Table-5).

4. Discussion

The aim of the study was to block the brachial plexus and to evaluate the effect of isoflupredone acetate as an adjunct to local anesthetic. Steroids being the powerful anti-inflammatory and analgesic agents have been used as an adjuvant with local anesthetic drugs in human medicine. The binding of steroids to intracellular receptors and modulation of nuclear transcription cannot explain their relatively rapid effect when used as an adjunct to local anesthetics [3]. The effect of vasoconstriction by steroids is mediated by occupancy of classical glucocorticoid receptors rather than by nonspecific pharmacological mechanisms [2]. Therefore, vasoconstriction, the presumed mechanism of action for epinephrine's adjunctive effect on local anesthetics, is probably not responsible for block prolongation by steroids. The local effect of corticosteroid on the nerve may be responsible for relative rapid analgesic property [4].The systemic effects of dexamethasone may be responsible for prolongation of local anesthetic block because they suppress inflammation through inhibition of phospholipase A2. Methylprednisolone when applied locally has been found to block transmission in nociceptive fibers and the effect was reversible, suggesting a direct membrane action of steroids [5]. In our study, we observed that the block performance time did not differ between the groups and ranged in all the groups between 15-20 minutes and lasted differently. In the animals of group C onset of anesthesia in all the nerves examined started at 15 minutes post injection except median and musculocutaneous nerve which started at 20 minutes post injection. The possible reason being that these nerves were at a farthest point from the site of the deposition of anaesthesia owing to its anatomical position related to the site of anesthesia injected. The duration of anesthesia varied in the nerves of the same plexuses from 145 for nerves radial and ulnar to 150 minutes for median and musculocutaneous nerves. In group D Animals all the nerves were blocked after 15 minutes and the duration of the anesthesia remained up to 150 minutes. The possible reason for much higher significant effect was due to incorporation of injection isoflupredone acetate. The anaesthetic agent use was otherwise 2 ml less than that of group C.

In the animals of group A the onset of sensory block for radial and ulnar nerve was 30 and 20 minutes respectively with the duration of anaesthesia lasting for 90 and 100 minutes respectively. Median and musculocutaneous nerves of the same group showed onset of sensory analgesia at 20 minute post injection and the anesthetic effect lasted for 100 minutes. The results of this group when compared with the results of group B which was done in conventional manner with the incorporation of steroid as an adjuvant, No significant effect was recorded as for onset of the anesthesia s concerned however there was a significant increase in the duration of anesthesia. The onset of motor blockade for group C and D was same (15 minutes), however the duration of block in group D was much longer as compared to group C. In group D, motor block was significantly more intense which even extended beyond 165 minutes.

5. Conclusions

Addition of steroid to local anesthetic agent in regional anesthesia of ruminants as an adjunct reduces its overall dosage but the extent up to which the steroid can replace the anesthetic drug without significantly decreasing the sensory and motor blockade effects needs to be evaluated.

6. References

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