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# Evaluation of midazolam-ketamine with dexmedetomidine and fentanyl for injectable anaesthesia in dogs

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**ABSTRACT**

A prospective randomized blinded study was conducted on 12 clinically healthy adult dogs of both sexes (mean weight of 18.34 ± 0.78 kg) divided into three groups (n = 4). The animals received 0.4 mg/kg midazolam and 10 µg/kg dexmedetomidine (group A), 0.4 mg/kg midazolam and 20 µg/kg dexmedetomidine (group B) and 0.4 mg/kg midazolam + 20 µg/kg dexmedetomidine + 4 µg/kg fentanyl (group C) intramuscularly, using separate syringes. Ten minutes later Ketamine was administered intravenously in all the groups. A significantly ($P<0.05$) shorter weak time (onset of sedation) and down time (onset of recumbency) were recorded in animals in group C as compared to the animals of groups A and B. Muscle relaxation was excellent in group C. The pedal reflex was abolished up to 30 min in groups A and B and up to 60 min in group C. Intubation was only possible in groups B and C. The anaesthetic induction dose of ketamine was minimal in group C. Standing recovery time was shortest in the animals of group C. Respiratory rate (RR) decreased significantly ($P<0.05$) throughout the observation period, but rectal temperature (RT) decreased significantly ($P<0.05$) towards the end of the observation period in all the groups. Heart rate decreased significantly ($P<0.05$) in the animals of group B. Mean arterial pressure (MAP) was maintained within the physiological range in all the groups. It was concluded that dexmedetomidine (10 µg/kg)-midazolam-ketamine can produce anaesthesia for about 20 min in dogs. Increasing the dose of dexmedetomidine did not enhance the duration of anaesthesia, but the further addition of fentanyl not only reduced the induction dose of ketamine but also increased the duration of anaesthesia up to 50 min. Dexmedetomidine-midazolam-fentanyl-ketamine can be used for prolonged duration of injectable anaesthesia in dogs.

**Key words:** anaesthesia, dexmedetomidine, dogs, fentanyl, ketamine, midazolam

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## Introduction

Alpha-2 adrenoceptor agonists are frequently used as pre-anaesthetics in veterinary practice. Dexmedetomidine is a highly selective and potent $\alpha_2$-adrenoceptor agonist that offers sedative and analgesic benefits over racemic medetomidine (KUUSELA et al., 2000). Co-administration of alpha-2 adrenoceptor agonists with opioids or benzodiazepines may result in pronounced sedation and analgesia (MUIR, 1998). Midazolam, a benzodiazepine derivative, is used as a premedicant, sedative and an anaesthetic induction agent, with minimal effect on cardiac function (LEMKE, 2007). Fentanyl citrate, a highly potent synthetic μ agonist, rapidly crosses the blood-brain barrier, which is responsible for its characteristic rapid onset and short duration of action (HUG and MURPHY, 1979). Opioids like butorphanol, hydromorphone, and buprenorphine, have been recommended for use in combination with dexmedetomidine and ketamine in dogs and cats (KO et al., 2009). A recent study suggested that dexmedetomidine-ketamine-buprenorphine was a suitable injectable anaesthesia combination for castration in dogs (BARLETTA et al., 2011). A review of the literature did not show any published report on evaluation of midazolam-dexmedetomidine-ketamine with fentanyl for anaesthesia in dogs. However, the combination of fentanyl-dexmedetomidine-midazolam resulted in excellent analgesia, sedation and muscle relaxation with favourable conditions for intubation in dogs (AHMAD et al., 2011). As small animal practitioners commonly use injectable anaesthetics alone or with inhalation anaesthetics for short-duration surgical procedures, it is worthwhile to investigate the use of dexmedetomidine-ketamine-opioid-based anaesthetic combinations for this purpose (BARLETTA et al., 2011). The present study was therefore designed to compare dexmedetomidine-midazolam-ketamine with dexmedetomidine-midazolam-fentanyl-ketamine for injectable anaesthesia in dogs.

## Materials and methods

A prospective randomized blinded study was conducted on 12 client-owned, mixed breed adult dogs of either sex weighing 18.34 ± 0.78 kg. An informed consent was obtained from the owners prior to subjecting the animals to the study. The animals were deemed healthy through physical examination, complete blood count, estimation of plasma urea and creatinine, and recording of electrocardiogram.

*Design of work.* The dogs were divided randomly into three equal groups, designated as group A, B and C. In the animals of group A, dexmedetomidine (Dexdomitor; 0.5 mg/mL; Orion Pharma, Turku, Finland) 10 μg/kg and midazolam (Mezolam; 1 mg/mL; Neon Laboratories, Thane, India) 0.4 mg/kg were administered simultaneously in the thigh muscles using separate syringes. In the animals of group B, dexmedetomidine 20 μg/kg and midazolam 0.4 mg/kg were administered in the same manner as in group A. In the animals of group C dexmedetomidine 20 μg/kg, midazolam 0.4 mg/kg and fentanyl

(Fendrop; 50 mcg/mL; Sun Pharmaceuticals India Ltd., India) 4 µg/kg were administered in the thigh muscles using separate syringes. Ten minutes later, ketamine (Ketmin 50; 50 mg/mL; Themis Medicare Ltd., Uttarakhand, India) was administered i.v. until the abolition of pedal reflex in all the groups.

Base values for different parameters were recorded before administration of pre-anaesthetic drugs. After administration of pre-anaesthetic drugs, the animal was left loose in a room to allow onset of the effects of the drugs and to record weak time (onset of sedation) and down time (onset of recumbency). Ten minutes later the animal was secured on the examination table in right lateral recumbency, and ketamine was administered in the cephalic vein until onset of anaesthesia was achieved, which was confirmed by loss of pedal reflex.

*Clinical observations.* Weak time was recorded as the time elapsed from the time of injection of pre-anaesthetic drugs to the time of onset of in-coordination / ataxia or drowsiness. Down time was recorded as the time that elapsed from the time of injection of pre-anaesthetic drugs to the time when the animal attained sternal recumbency.

Relaxation of the jaw was taken as a measure of muscle relaxation. It was evaluated by observing the resistance to opening of the jaws while pulling the jaws apart. The status of jaw relaxation was recorded at 0 min (base value), 10 min (just before administration of ketamine) and then at 15, 20, 30, 45, 60, 75, 90, 105 and 120 min on a 0 to 4 score scale, as shown in Table 1. At each interval, the mean value for jaw relaxation score was calculated and the muscle relaxation was graded as nil if the mean score was 0, very mild when the score was >0 but <1, mild when the score was ≥1 but <2, moderate when the score was ≥2 but <3 and excellent when the score was 3.

Status of palpebral reflex was recorded, at the same interval as for the relaxation of the jaw, by observing blink of the eye lids on touching the area around medial canthus of the eyes with an index finger, on a 0 to 3 score scale (Table 1).

The status of pedal reflex was recorded as a measure of the depth of analgesia. It was assessed by observing the withdrawal reflex to pinching the interdigital skin of the hind foot of the animal (KUUSELA, 2004). The response of the animal to pinching was graded on a 0 to 3 score scale (Table 1) at the same interval as for the jaw relaxation. The mean value for pedal reflex score was calculated and the analgesia was graded at each interval as: no analgesia if the mean score was 0, very mild analgesia when the score was >0 but <1, mild analgesia when the score was ≥1 but <2, moderate analgesia when the score was ≥2 but <3, and complete analgesia when the score was 3.

Response to intubation was recorded to assess the status of laryngeal reflexes and feasibility of intubation during different stages of sedation/anaesthesia in all the animals. The response to intubation was recorded by attempting intubation, at the same intervals

as for the jaw relaxation, until the animal allowed easy intubation using a 0 to 4 score scale (Table 1). The mean value for intubation score was calculated at each interval and the status of laryngeal reflex was graded as: strong if the mean score was <1, very mild depression when the score was ≥1 but <2, mild depression when the score was ≥2 but <3, moderate depression when the score was ≥3 but <4, and complete depression of laryngeal reflex when the score was 4. Extent of salivation was recorded at different intervals as for the other reflexes and was graded from 0 to 3 using the score scale shown in Table 1. The reflex responses were allotted by a person blinded to the treatment.

Table 1. System of recording of various reflexes and responses (Adapted and modified after AMARPAL et al., 1996)

| | Score (Numbers) | | | | |
|---|---|---|---|---|---|
| Parameter | 0 | 1 | 2 | 3 | 4 |
| Relaxation of jaw | Not allowing to open the jaws | Resistant to opening the jaws and closed quickly | Less resistance to opening the jaws and closed slowly | No resistance and jaws remain open | – |
| Palpebral reflex | Intact and strong (quick blink) | Intact but weak (slow response) | Very weak (very slow and occasional response) | Abolished (no response) | – |
| Pedal reflex | Intact and strong (strong withdrawal) | Intact but weak (animal responding slowly) | Intact but very light (slow and occasional response) | Abolished completely (no response) | – |
| Response to intubation | Not permitting entry of tube in the mouth | Allowing entry but chewing | Allowing deeper entry but coughing | Difficult intubation with coughing | Easy intubation without coughing |

Recovery time was recorded as the time elapsed from injection of the drugs to the time of reappearance of pedal reflex. The time to the return of righting reflex was recorded as the time from injection of drug until the animal regained sternal recumbency. Standing recovery time was recorded as the time interval elapsed from the time of injection of the drugs until the time when animal attained standing position. Complete recovery time was recorded as the time elapsed from the time of reappearance of pedal reflex to the time when the animal stood and walked unassisted. Duration of anaesthesia was recorded as the time interval that elapsed from the time of abolition of pedal reflex to the time of reappearance of the pedal reflex.

The total dose of ketamine required to induce anaesthesia was recorded and dose per kg body mass was calculated for each group.

*Physiological observations.* Heart rate (HR), respiratory rate (RR), rectal temperature (RT), oxygen saturation ($SpO_2$) and mean arterial pressure (MAP) were recorded before administration of the drug(s) at 0 min (base value) and at 10 min (just before administration of ketamine) and then at 15, 20, 30, 45, 60, 75, 90, 105, and 120 min after administration of the drugs. Respiratory rate was recorded by counting the excursions of thoraco-abdomen. Rectal temperature was recorded with the help of a digital thermometer, as per the standard procedure. Heart rate (HR) and oxygen saturation ($SpO_2$) were measured by means of a pulse oximeter (Model 8600, pulse oximeter; Nonin Medical Inc. MPLS, Minnesota) applied to the toe web of the forelimb after clipping the hair around the site and cleaning with 70% alcohol (HUSS et al., 1995). Mean arterial pressure was recorded by applying the cuff of a non-invasive blood pressure (NIBP) (Surgivet®, Smith's medical PM, Inc. Waukesha, USA) monitor around the digital artery.

*Statistical analysis.* The data were analysed for statistical significance using SPSS software, version 15.0 (SPSS, Inc., Chicago, Illinois). The means at different time intervals were compared among different groups, using one way analysis of variance and Duncan's multiple range test (DMRT). The mean values at different intervals were compared with their base values in each group using the Paired "t" test. The subjective data generated from the scoring of various parameters were analysed using the Kruskal Wallis test. In each analysis, the differences were considered significant at a value of $P<0.05$.

**Results**

*Clinical observations.* Weak time was significantly ($P<0.05$) shorter in the animals of group C (2.25 ± 0.25 min) as compared to group B (3.25 ± 0.25 min) and group A (4.37 ± 0.23 min). Down time in the animals of group C (4.00 ± 0.40 min) was significantly ($P<0.05$) shorter than that in group A (5.87 ± 0.125 min), but did not differ significantly from that in group B (4.50 ± 0.28 min) (Fig. 1).

Excellent muscle relaxation was observed up to 30 min in groups A and B, and up to 90 min in group C. Muscle relaxation score was significantly ($P<0.05$) higher in group C as compared to groups A and B from the 45 to 90 min observation periods. Palpebral reflex was more sluggish in the animals of groups A and C as compared to group B (Fig. 2). The pedal reflex was lost completely after the administration of ketamine at 10 min in animals in all the groups, which indicated onset of surgical anaesthesia (score 3). The pedal reflex was lost completely up to 30 min in groups A and B and up to 60 min in group C (Fig. 3) suggesting anaesthesia of 20 min (time of ketamine injection to 30 min) in groups A and B and 50 min (time of ketamine injection to 60 min) in group C. Intubation was not possible in the animals of group A at any time interval of the study. Easy intubation was possible in the animals of groups B and C, and loss of laryngeal reflex persisted up to the 45 min interval in group B and 90 min in group C (Fig. 4).

Intubation scores were significantly higher ($P<0.05$) in group C 45 to 90 min interval. Mild to moderate salivation was observed in animals of all the groups.

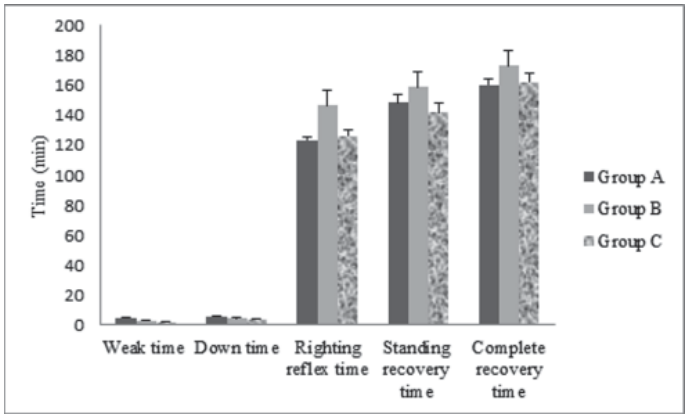


Fig. 1. Mean weak time, down time, righting reflex time, standing recovery time and complete recovery time in groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine)

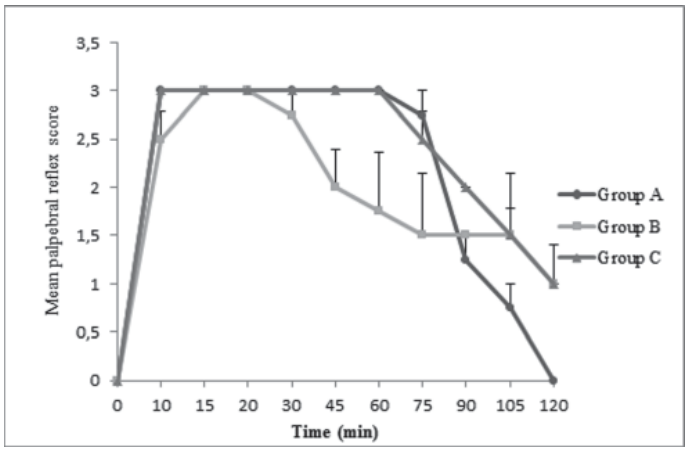


Fig. 2. Mean ± SE palpebral reflex score after drug administration in groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

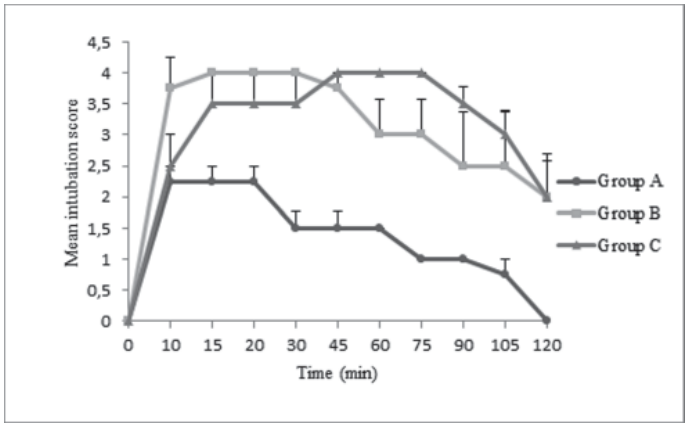


Fig. 3. Mean ± SE pedal reflex score after drug administration in groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

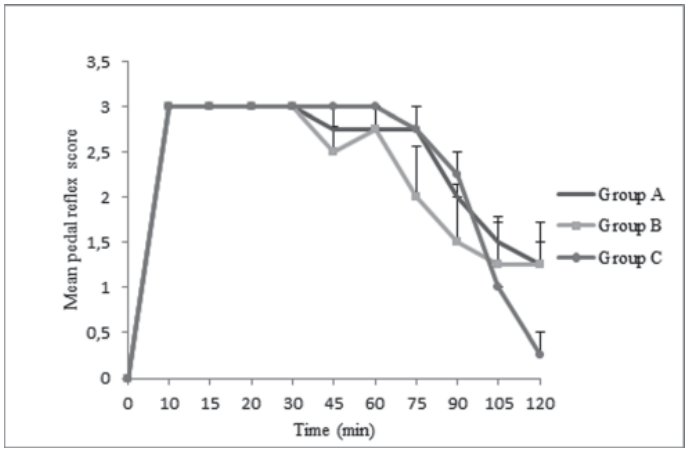


Fig. 4. Mean ± SE intubation score after drug administration in groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

The dose of ketamine needed for induction of anaesthesia was reduced from 9.04 ± 0.0.59 mg/kg in group A to 8.93 ± 0.83 mg/kg in group B and 7.98 ± 0.22 mg/kg in group C.

Recovery time was significantly (P<0.05) shorter in the animals of group A (51.75 ± 4.49 min) as compared to group B (74.50 ± 5.42 min) and group C (83.75 ± 3.44 min). The time to the return of righting reflex in group B (146.50 ± 10.33 min) was significantly (P<0.05) longer than that in group A (123.25 ± 2.28 min) and C (126.25 ± 3.7 min). Standing recovery time was shortest in the animals of group C (142.00 ± 6.62 min) followed, in increasing order, by group A (148.50 ± 5.51 min) and in group B (158.75 ± 10.71 min), but the differences between the groups were not significant. The complete recovery time was longer in the animals of group B (173.00 ± 10.07 min) than that in group C (162.00 ± 6.28 min) and in group A (160.50 ± 3.92 min). The differences in the complete recovery time between the groups were not significant (Fig. 1).

Table 2. Mean ± SE of heart rate in the animals of groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

| Groups | Intervals (Min) | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | 0 | 10 | 15 | 20 | 30 | 45 | 60 | 75 | 90 | 105 | 120 |
| Group A | 101.00 ± 12.65 | 66.50$^{a}$ ± 14.97 | 107.00 ± 22.18 | 83.50 ± 9.84 | 86.50 ± 7.29 | 75.25 ± 10.29 | 73.25$^{a}$ ± 8.66 | 65.20 ± 8.13 | 68.25 ± 7.89 | 69.50$^{a}$ ± 6.03 | 76.50$^{a}$ ± 3.00 |
| Group B | 105.50 ± 8.14 | 48.75$^{a}$ ± 9.10 | 93.00 ± 13.41 | 88.50 ± 11.90 | 77.00 ± 8.82 | 56.75$^{*}$ ± 9.10 | 43.75$^{b**}$ ± 1.37 | 41.25$^{**}$ ± 1.75 | 44.25$^{*}$ ± 4.55 | 40.00$^{b**}$ ± 1.15 | 41.75$^{b**}$ ± 2.50 |
| Group C | 115.00 ± 15.5 | 109.50$^{b}$ ± 15.26 | 111.75 ± 3.35 | 95.00 ± 11.09 | 90.00 ± 12.51 | 62.00 ± 4.88 | 56.25$^{ab}$ ± 7.12 | 48.75 ± 9.09 | 46.25 ± 9.28 | 54.00$^{ab}$ ± 6.55 | 56.25$^{b*}$ ± 8.50 |

*Significantly different from base value (P<0.05); **Significantly different from base value (P<0.01); Values with different alphabets differ significantly at respective intervals (P<0.05)

*Physiological observations.* In animals of group A, heart rate remained decreased throughout the study period, although this decrease was not significant as compared to the baseline value. In animals of group B, heart rate decreased significantly (P<0.01) at the 10 min interval i.e. just before the administration of ketamine. HR showed some improvement from 10 min to 30 min after administration of ketamine, however, the values were still lower as compared to the base value. At 45 min interval HR was again significantly (P<0.05) lower than the base value and remained so up to 120 min of the study (P<0.01). In animals of group C, heart rate decreased and remained below the baseline throughout the study period but this decrease was significant (P<0.05) only at 120 min. Mean heart rate was significantly (P<0.05) lower in group B at 60, 105 and 120 min, when compared to group A (Table 2).

Table 3. Mean ± SE of respiratory rate in the animals of groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

| | Intervals (Min) | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Groups | 0 | 10 | 15 | 20 | 30 | 45 | 60 | 75 | 90 | 105 | 120 |
| Group A | 33.50 ± 7.54 | 16.00* ± 2.65 | 14.25 ± 0.85 | 14.00 ± 1.35 | 15.00* ± 1.78 | 14.70* ± 2.50 | 14.75* ± 3.00 | 15.00* ± 2.65 | 15.00* ± 3.37 | 15.50* ± 4.25 | 21.50* ± 8.75 |
| Group B | 34.50 ± 5.04 | 13.25* ± 0.75 | 12.25* ± 0.63 | 11.50* ± 0.96 | 11.25* ± 0.85 | 10.50* ± 1.20 | 10.50* ± 1.20 | 10.50* ± 1.20 | 11.25* ± 0 .95 | 10.50* ± 0.65 | 11.00* ± 1.08 |
| Group C | 26.00 ± 3.03 | 11.00* ± 0.41 | 11.75* ± 1.18 | 11.00* ± 0.82 | 12.25* ± 1.25 | 11.50* ± 1.20 | 15.00* ± 1.47 | 16.00* ± 0.71 | 16.50* ± 0.96 | 17.25* ± 0.63 | 19.25* ± 1.32 |

*Significantly different from base value (P<0.05); **Significantly different from base value (P<0.01); Values with different alphabets differ significantly at respective intervals (P<0.05)

Table 4. Mean ± SE of rectal temperature in the animals of groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

| | Intervals (Min) | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Groups | 0 | 10 | 15 | 20 | 30 | 45 | 60 | 75 | 90 | 105 | 120 |
| Group A | 38.59 ± 0.12 | 38.77 ± 0.19 | 38.48 ± 0.41 | 38.41 ± 0.38 | 38.15 ± 0.37 | 37.95 ± 0.38 | 37.70 ± 0.43 | 37.57* ± 0.42 | 37.40* ± 0.40 | 37.21* ± 0.39 | 37.65* ± 0.39 |
| Group B | 38.99 ± 1.19 | 39.03* ± 0.14 | 38.93** ± 0.12 | 38.66 ± 0.15 | 38.56 ± 0.13 | 38.40 ± 0.12 | 38.24 ± 0.09 | 37.86 ± 0.20 | 37.73 ± 0.22 | 37.56 ± 0.22 | 37.43 ± 0.22 |
| Group C | 38.98 ± 0.08 | 38.78 ± 0.07 | 38.75 ± 0.06 | 38.61* ± 0.09 | 38.52* ± 0.08 | 38.43* ± 0.09 | 38.21* ± 0.15 | 37.91* ± 0.15 | 37.87** ± 0.08 | 37.56** ± 0.05 | 37.59** ± 0.03 |

*Significantly different from base value (P<0.05); **Significantly different from base value (P<0.01)

Table 5. Mean ± SE of mean arterial pressure in the animals of groups A (dexmedetomidine 10 µg/kg-midazolam 0.4 mg/kg-ketamine), B (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-ketamine) and C (dexmedetomidine 20 µg/kg-midazolam 0.4 mg/kg-Fentanyl 4 µg/kg-ketamine) at different time intervals

| | Interval (Min) | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Groups | 0 | 10 | 15 | 20 | 30 | 45 | 60 | 75 | 90 | 105 | 120 |
| Group A | 116.75 ± 6.70 | 118.25 ± 8.22 | 117.75 ± 11.56 | 119.80 ± 12.37 | 120.50 ± 10.31 | 122.00 ± 10.54 | 106.25* ± 8.73 | 111.25 ± 10.88 | 105.00** ± 5.87 | 107.25 ± 8.13 | 110.50 ± 9.04 |
| Group B | 128.25 ± 8.37 | 113.07 ± 15.25 | 157.00 ± 9.84 | 160.00* ± 14.42 | 131.00 ± 4.69 | 131.50 ± 8.79 | 114.42 ± 4.09 | 111.50 ± 1.65 | 121.50 ± 6.53 | 118.25 ± 9.49 | 120.50 ± 5.17 |
| Group C | 124.75 ± 8.90 | 138.75 ± 3.40 | 152.75 ± 4.93 | 162.50 ± 10.47 | 144.75 ± 4.11 | 124.75 ± 7.20 | 120.75 ± 3.81 | 111.25 ± 6.10 | 106.75 ± 3.92 | 102.75 ± 1.18 | 110.50 ± 3.08 |

*Significantly different from base value ($P<0.05$); **Significantly different from base value ($P<0.01$)

RR decreased significantly ($P<0.05$) as compared to the respective baseline values in all the groups. Comparison of RR between the groups did not reveal any significant difference at any time interval (Table 3).

RT increased initially up to 15 min in the animals of group B ($P<0.05$). Thereafter RT decreased gradually and remained just below the base line throughout the study period. In the animals of groups A and C, RT decreased gradually to become significantly ($P<0.05$) below the baseline from 75 to 120 min in group A and from 20 min to 120 min in group C (Table 4). However, RT did not differ significantly between the groups.

Oxygen saturation values did not differ significantly ($P>0.05$) from the baseline value throughout the study period, except for occasional decreases in groups B and C up to the 45 min interval.

Mean arterial pressure (MAP) increased initially and then decreased gradually towards the end of study in all the groups. In group A, mean arterial pressure reached significantly below the baseline at the 60 min ($P<0.05$) and 90 min ($P<0.01$) intervals. Mean arterial pressure remained near the baseline values throughout the study period in the animals of group B, except at 20 min, where a significant ($P<0.05$) increase was observed. MAP in the group C animals fluctuated around baseline values without significant changes (Table 5). MAP did not differ significantly between the groups.

**Discussion**

Dexmedetomidine, like medetomidine, has rapid onset of action owing to its lipophilic properties (AMARPAL et al., 1996). The fast onset of sedation and recumbency recorded

in the present study conformed to the observations made in earlier studies following the administration of medetomidine/dexmedetomidine (AMARPAL et al., 1996; AHMAD et al., 2011).

Ketamine, when used alone for anaesthesia in dogs, produces spontaneous movement, muscle rigidity and violent recovery (LIN, 2007). Clinically it is used in combination with or after tranquilizers or sedatives to eliminate its side effects. In the present study, 20 µg/kg dose of dexmedetomidine with fentanyl and midazolam as pre-anaesthetics to ketamine produced better muscle relaxation for a longer duration, as compared 10 and 20 µg/kg dose of dexmedetomidine with midazolam. The findings of the present study conformed to the observations of earlier researchers, who reported greater muscle relaxation when dexmedetomidine or medetomidine was combined with opioid and/or ketamine in cats or dogs (KO et al., 2000; SELMI et al., 2003).

Abolition of palpebral reflex was more consistent in group C as compared to group B. The lower palpebral reflex score recorded in the animals of group B, as compared to group A, could be attributable to $\alpha_1$-adrenergic receptor activation mediated arousal and vigilance due to the increased dose of dexmedetomidine (PUUMALA et al., 1997). It has been reported that $\alpha_1$- adrenoceptor effect will predominate with increased dose of α-2 agonists, which can antagonize the hypnotic action of even potent α-2 agonists like dexmedetomidine (SINCLAIR, 2003). The greater depression of the palpebral reflex in the animals of group C as compared group B might be attributed to the action of fentanyl. Fentanyl, basically an analgesic agent, could have increased the depth of the sedation by its synergistic interaction with dexmedetomidine/midazolam. Synergistic interactions have been reported between alpha-2 agonists, opioids and benzodiazepines (SALMENPERA et al., 1994; AMARPAL et al., 1996; BOL et al., 2000).

Analgesia was excellent, with complete loss of pedal reflex, in all the groups after 10 min but persisted for longer in the animals of group C. Complete loss of pedal reflex was plausibly attributed to the action of ketamine in all the groups. Ketamine itself is a short acting anaesthetic drug, but its relatively long duration of anaesthesia in the present study could be attributable to the action of dexmedetomidine/gmidazolam administered as pre-anaesthetics in groups A and B. The further increased duration of anaesthesia in group C could be attributed to the additional action of the µ opioid agonist, fentanyl. A synergistic interaction between alpha-2 agonists, opioids and benzodiazepines has been reported in earlier studies (SALMENPERA et al., 1994; AMARPAL et al., 1996; BOL et al., 2000).

Dexmedetomidine and midazolam, by themselves, are not general anaesthetics and are incapable of completely abolishing the laryngeal reflex, and also laryngeal and pharyngeal reflexes are reasonably well maintained during ketamine induced anaesthesia in all species (HASKINS et al., 1975). This might have prevented intubation in group A. Increase in the dose of dexmedetomidine might have led to further depression of laryngeal reflexes to allow intubation in the animals of group B. In group C, the complete and

prolonged depression of laryngeal reflex might have been achieved due to the synergistic interaction of dexmedetomidine and midazolam with fentanyl (SALMENPERA et al., 1994).

Recovery from ketamine anaesthesia occurs through tissue redistribution and hepatic metabolism of the drug making it a short acting anaesthetic drug (STEPHENSON et al., 1978). However, the higher dose of dexmedetomidine in group B and group C, resulting in deeper sedation, might have led to reduced metabolic activity to delay redistribution and metabolism of the drugs, resulting in prolongation of recovery time (JACOBSON and HARTSFIELD, 1993; KO et al., 2000).

The total dose of ketamine required to induce anaesthesia was minimal in group C, but the differences in groups A and B were marginal. It suggested that increasing the dose of dexmedetomidine has no dose sparing effect on ketamine, but further addition of fentanyl can reduce the dose of the anaesthetic agent.

Bradycardia was observed in all the groups, but it was more obvious in groups B and C. Dexmedetomidine, like medetomidine, causes dose dependent depression of the cardiovascular system, with maximal depression at a higher dose (KUUSELA et al., 2001). Fentanyl may also cause bradycardia by activation of cardiac vagal efferents (THURMON et al., 1999). The greater decrease in HR recorded in the animals of groups B and C could thus be attributable to the higher dose of dexmedetomidine in group B and dexmedetomidine plus fentanyl in group C.

Alpha-2 agonists are known to induce RR depression, which is mediated by activation of the alpha-2 adrenergic pathway, leading to inhibition of locus coeruleus neurons (OYAMADA et al., 1998). Similarly, μ receptors agonists induce analgesia, accompanied by some respiratory depression, but in therapeutic doses fentanyl has minimal side effects (THURMON et al., 1999). Respiratory rates decreased markedly in all the three groups. Profound respiratory depression has been reported when medetomidine and ketamine were used in dogs by KO et al. (2001).

Sedative/anaesthetic drugs may plausibly induce a decrease in rectal temperature by decreasing heat production, as a result of decreased muscular activity, and by direct action on the hypothalamus (VIRTANEN, 1989). In spite of the decrease in last phase of the study, RT remained within physiological limits in all the groups, and differences in RT were not significant among the groups.

Low pulse oximeter readings are indicative of reduced arterial oxygenation and diminished tissue perfusion. However, as reported by LEPPANEN et al. (2006) vasoconstriction may also lead to low pulse oximeter readings. A decrease in oxygen saturation after the administration of dexmedetomidine in humans and swine has been reported by SANO et al. (2010). However, in the present study $Spo_2$ was fairly maintained,

except slightly lower oximeter readings at a few intervals in groups B and C, which could be attributable to vasoconstriction due to the higher dose of dexmedetomidine.

Mean arterial blood pressure increased initially as compared to baseline values, which was followed by a decrease towards the end of the study period in all the groups. Dexmedetomidine/medetomidine has been associated with high systemic vascular resistance due to alpha-2 mediated vasoconstriction, which maintains arterial blood pressure in the face of lowered cardiac output (LAWRENCE et al., 1996). In the present study blood pressure was also maintained within the physiological limits in all the groups, which could be attributable to the effect of dexmedetomidine and ketamine.

## Conclusion

It was concluded that dexmedetomidine (10 µg/kg)-midazolam-ketamine induces surgical anaesthesia for 20 min in dogs. Increasing the dose of dexmedetomidine did not enhance the depth and duration of anaesthesia but further addition of fentanyl not only reduced the induction dose of ketamine but also increased the duration of anaesthesia. The combination may be suitable for injectable anaesthesia in dogs.

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**SANTOSH, K. M., AMARPAL, R. A. AHMAD, P. KINJAVDEKAR, H. P. AITHAL, A. M. PAWDE, D. KUMAR: Prosudba učinka midazolam-ketamina s deksmedetomidinom i fentanilom za injekcijsku anesteziju u pasa. Vet. arhiv 83, 509-523, 2013.**

**SAŽETAK**

Poduzeto je prospektivno istraživanje na 12 slučajno odabranih klinički zdravih pasa i kuja (prosječne tjelesne mase 18,34 ± 0,78 kg) podijeljenih u tri skupine (n = 4). Životinjama skupine A bio je intramuskularno primijenjen midazolam u dozi od 0,4 mg/kg i deksmedetomidin u dozi od 10 µg/kg. Životinjama skupine B bio je i/m primijenjen midazolam u dozi od 0,4 mg/kg i deksmedetomidin 20 µg/kg, a životinje skupine C primile su i/m 0,4 mg/kg midazolama, 20 µg/kg deksmedetomidina i 4 µg/kg fentanila. Deset minuta nakon toga svim je životinjama intravenski bio ubrizgan ketamin. Značajno (P<0,05) kraće vrijeme smirivanja (nastup sedacije) i vrijeme lijeganja ustanovljeno je u životinja skupine C u usporedbi sa skupinama A i B. Opuštanje mišićja bilo je izvrsno u skupini C. Nožni refleks nestao je nakon 30 minuta u skupinama A i B, a nakon 60 minuta u skupini C. Intubacija je bila moguća samo u životinja skupine B i C. Doza ketamina potrebna za početak anestezije bila je najmanja u životinja skupine C. Vrijeme potrebno za ponovno ustajanje bilo je najkraće u životinja skupine C. Frekvencija disanja značajno se smanjila (P<0,05) u čitavom razdoblju promatranja, dok se rektalna temperatura u svih životinja značajno smanjila (P<0,05) na kraju razdoblja promatranja. Frekvencija bila znatno se smanjila (P<0,05) u životinja skupine B. Srednji arterijski tlak bio je u fiziološkim granicama u svih životinja. Može se zaključiti da kombinacija deksmedetomidin (10 µg/kg)-midazolam-ketamin može u pasa dovesti do anestezije za oko 20 minuta. Povećanje doze deksmedetomidina nije povećalo trajanje anestezije. Ipak, daljnja primjena fentanila ne samo da je smanjila početnu dozu ketamina već je povećala trajanje anestezije na 50 minuta. Deksmedetomidin-midazolam-fentanil-ketamin mogu se rabiti za produženo trajanje injekcijske anestezije u pasa.

**Ključne riječ:** anestezija, deksmedetomidin, fentanil, ketamin, midazolam, psi