Advances in Environmental Biology, 5(7): 1871-1876, 2011
ISSN 1995-0756

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ORIGINAL ARTICLE

Comparison of the Sublingual Administration of Ultra Short Action Benzodiazepines Versus Ketamine for Cns Depression of Cat

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Eilyad Issabeagloo, Jamshid Ghiasi Ghalahkandi: Comparison of the Sublingual Administration of Ultra Short Action Benzodiazepines Versus Ketamine for Cns Depression of Cat

ABSTRACT

Ketamine is a suitable injectable anesthetic in human and animal that has a low intestinal absorption rate. The current study compared acceptance and behavioral responses to sublingual administration of midazolam and Ketamine in cat. Ten male cats received different doses of Ketamine or midazolam via sublingual route. Each animal was observed continuously for CNS depression as standard grades on the behavioral scale. Quantal data were obtained by determining the percentage of animals which lost the reflexes (graded as 5 scores) for each dose. Peak scores for each dose and the percentage of animals that reached each peak score is given. Animals accepted midazolam and Ketamine administered via the sublingual administration. Different doses of ketamine and midazolam showed dose dependent effect in CNS depression. Midazolam administration via the sublingual in any Doses could induce only Immobility in animals. But all of cats reacted to pain. Also 100% of animals showed no reaction to pain only in ketamine 80 mg/kg. Also Onset time of effect in midazolam administration was dose dependent but in regard with ketamine wasn't in this order. Sublingual administration of Ketamine is as effective as, and better Suppressed than, Sublingual midazolam as a sedative-hypnotic in cats.

Key words: Ketamine, Midazolam, Sublingual administration, CNS depression, Cat.

Introduction

Ketamine (KT) is a synthetic available anesthetic that has been used in human & animal operations for almost 35 years. Several studies had showed its wide margin of safety [2,6]. This agent blocks NMDA (N-methyl-D-Aspartate) receptors in CNS [5]. KT induces one form of anesthesia that called dissociative anesthesia in which there is a marked sensory loss and analgesia, as well as amnesia and paralysis of movement, without actual loss of consciousness [7]. KT has some other effects such as sedation, analgesia, and immobility. This drug has low intestinal absorption rate. It's bioavailability in human with oral administration is %20±7 [17] and with rectal administration in cat is %43.5±6/1 [18]. KT was stated to be metabolized to at least two major compounds of pharmacological interest: to norketamine (NK) by *N*-demethylation, which then is converted to dehydronorketamine (DHNK) by dehydrogenation [19]. Its major metabolite NK, however, is active with one-third of the potency of its parent drug as an anesthetic. Thus the first-pass effect after oral administration results in an active metabolite that can contribute to the pharmacological effects.

Oral KT has been used sporadically as premedication for anesthesia in children [20,1]. Also In fractious cats, is administered by squirting the drug into the mouth with a syringe when the animal is hissing [9,4]. KT in anesthetized cats cause psychotic symptoms, release of histamine and induce

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cardiovascular system hyper activity such as increase of hart rate and hypertension. Benzodiazepines are one group of sedative-hypnotic drugs that used to reduce hallucination induced by KT. Midazolam is one of ultra short action benzodiazepines that be given intravenously in anesthesia.

Midazolam, which induces anaesthesia in humans at IV. doses of 0.3 mg/kg, can not anaesthetize cats at doses higher than this dose [11]. Also this drug has little respiratory or cardiovascular depression effects [8]. Nevertheless it seems that co administration of Midazolam with ketamine improves CNS suppressing effect of KT and decreases some side effect of this drug (e.g. hallucination). The main and important aim of this study was evaluation of CNS suppressing effect of orally administrated ketamine & Midazolam in the manner of single and together.

Material and method

Animals:

Male, mature, sturdy free roaming and mixed breed cats selected randomly and were maintained as group housing in wide space (in a big room) to exhibit a wide range of complex behaviors. Animals had free access to food and water, and maintained on a 12-hour light-dark cycle. Temperature 25^{oc} with humidity between 45 and 65% provided for them allover the study. Food was withheld for 12 h and water for 2 h prior to the study to minimize the effects of gastric contents. They were kept one week before the examination in their room to achieve maximum adaptation to environmental situations. The numbers of cats in all of the treatment groups were ten animals.

Drugs:

Racemic ketamine (ketamine hydrochloride, Sigma, St. Louis, MO, U.S.A.) was dissolved in normal saline and the ph of each solution was adjusted to 5. Ketamine at a dose of 20, 40, 80 mg/kg (adams, 2001), was administered orally to cats in mixed with milk (40 cc) or meat (30 g) or sprayed in mouth by a ordinary syringe (when animals were rejecting the drug in mixture with milk or meat). For comparison, a similar study was performed with Midazolam. Midazolam (Roche, Switzerland) was dissolved in normal saline and different doses of Midazolam (0.3, 0.6, 1.2, 2.4, 4.8 mg/kg) [3] were administered as a mentioned method. To prevent absorption of the drugs from lower parts of gastrointestinal tract such as stomach or intestine, thus mentioned doses were balanced no exceed of 0.5 ml in treatment groups.

Be remembered because absence of scientific resources about oral doses of this two drugs in cat, therapeutic doses in IV. Administration rout, considered as minimum dose (base dose) for oral route of administration and according to this base, second, third and further doses estimated as two, three and more fold of first dose respectively. In first stage, drugs administered separately. In second step midazolam co-administered with Ketamine in treatment groups. In combination regimes high dose of each drug with low dose of other, also middle dose with other's middle dose was used. Hence treatment groups include:

1) Midazolam 0.3 mg/kg, 2) midazolam 0.6 mg/kg, 3) midazolam 1.2 mg/kg, 4) midazolam 2.4 mg/kg, 5) midazolam 4.8 mg/kg, 6) ketamine 20 mg/kg, 7) ketamine 40 mg/kg, 8) ketamine 80mg/kg, 9) ketamine 20 mg/kg plus midazolam 1.2 mg/kg, 10) ketamine 40 mg/kg plus midazolam 0.6 mg/kg, 11) ketamine 80 mg/kg plus midazolam 0.3 mg/kg.

Each animal was observed continuously by an educated expert for CNS depression as graded on the behavioral scales shown as fallow.

Scales for CNS depression were [16]:

1) No effect
2) impaired gait, prancing gat, some excitement
3) Lowered head, braced stance, hindquarter weakness
4) Sternal or lateral recumbency, some responsiveness to repositioning
5) lateral recumbency, no response to movement of limbs and painful excitements

Reflex to pain in cat is evaluated by painful excitation of tail or pads with clamp [12].

Also obtained results in administration of various doses of drugs were evaluated on the base of underneath parameters for each treatment group [15]:

- Onset time of effect: refer to initiation first effect result from drug, which generally reveals by relaxation and mild ataxia.
- Duration of effect: refer to drug effect length of time (from initiation of first drug effect and passing of peak score and then achieving to normal state in animal).
- Peak score for each dose: refer to the highest rate of CNS suppression in administrated dose.
- Percentage of animal reached peak score: lost the reflexes (upon scores) for each dose.
- Onset time of peak score: refer to peak score initiation time of each dose.
- Duration of peak score: refer to time that animal is in highest recordable score in administrated dose.

When ever score 2 recorded we did not recognize any time to Duration of Peak Score & onset time of Peak Score. Also Times more than 6 hours was not recorded in this study.

Statistical analysis:

The results (Onset Time and Duration of CNS depressant effects) are expressed as the Mean ± SE. Differences between the individual mean values in different groups were analyzed by one-way analysis of variance (ANOVA) and differences with a $p<0.05$ were considered significant.

Results and discussion

CNS suppression effect of orally administered ketamine (20, 40, 80 mg/kg) & Midazolam (0.3, 0.6, 1.2, 2.4, 4.8 mg/kg) in mixture with milk and meat in cat:

In these routs almost all of cats did not accept drugs in mixture with milk & meat or ice cream or chocolate. Therefore sublingual method was used over all study time.

Rate of CNS suppression of sublingual administration of Midazolam (0.3, 0.6, 1.2, 2.4, 4.8 mg/kg):

As shown in tables 1 & 2, Onset time of CNS suppression decrease with increasing dose of midazolam. So that in dose 4.8 mg/kg this time is half of 0.3 mg/kg (lowest administered dose). Also duration of CNS suppression was lasted with increasing dose of Midazolam. Midazolam in the highest administered dose only could create score 4 in cats. But the duration of time in which cats show score 4 is lasted with increasing dose of midazolam.

Rate of CNS suppression of sublingual administration of ketamine (20, 40, 80 mg/kg):

As shown in Tables 3 & 4, Onset time of effect decrease with increasing dose of ketamine. In dose of 80 mg/kg this time decrease to 1':24" that in comparison with ketamine 20 mg/kg was considered significant) $P<0.001$).

Also peak score of CNS suppression increased dose dependently so that in dose 80 mg/kg, in 50% of cats analgesia was sow (Score5). Onset time of peak score decreased dose dependently so that in dose of 80 mg/kg this time reached to 2.59±0.5 minute with almost in comparison, is half of 20 mg/kg.

Rate of CNS suppression in co-administration (as sublingual method) of ketamine (20, 40, 80 mg/kg) with midazolam (0.3, 0.6, 1.2 mg/kg):

Results of CNS suppression effects of co administration of ketamine 20 mg/kg + midazolam 1.2 mg/kg, ketamine 40 mg/kg + midazolam 0.6 mg/kg, ketamine 80 mg/kg + midazolam 0.3 mg/kg has been showed in tables 5 & 6. So as saw in table 5 in group ketamine 40 + midazolam 0.6, onset time of CNS suppression is faster than other two groups in comparison. Also duration of CNS suppression (duration of peak score & duration of effect) in mentioned group is considered significant more long-lasting in compare with other two groups ($P<0.001$). But peak score in group ketamine 80 + midazolam 0.3 was sow in further numbers of cats (80%) in comparison with other treatment groups.

Comparison of CNS suppression of ketamine (as sublingual administration) with co administration of it & midazolam: long late

On the base of graphs 1 & 2 midazolam in higher doses (0.6, 1.2 mg/kg) shows significant effect in lasting of ketamine's induced onset time of effect. But in lower doses (e.g. 0.3 mg/kg) has not such effect.

Also duration of CNS suppression in co administration of higher doses of ketamine (e.g. 40, 80) with midazolam is considered significant prolonger than ketamine alone in same doses ($P<0.001$). Extreme of this effect is in co administration of ketamine 40 + midazolam 0.6 ؛so that is more than 6 hours.

Discussion:

The results of present study show some notable facts:

1) Administration of ketamine or midazolam as mixed with milk, meat, ice cream or chocolate is not an appropriate rout of administration of these drugs in cat.
2) Intra oral (sublingual) spray of ketamine & midazolam in administrated doses absolutely causes a dose dependently CNS suppression.
3) CNS depression in co administration of midazolam with ketamine improved in compare with sporadically Ketamine administration.

Ketamine is a drug with high lipid solubility and rapidly leaves plasma to the CNS (brain). After i.v. administration, maximum within 1 minute it reaches to the highest brain concentration. There for this fact is compatible with its rapid onset time of effect that seems to be some seconds after IV administration [13,14]. I.v. administration of ketamine has some obvious CNS suppression effects in cat. In the present study sublingual ketamine spray induced CNS depression effects in 2.5-3 minutes in cats. These effects were dose dependently so that with dose of 80 mg/kg the cats reached to score5 (analgesia). This drug's Rapid onset time of effect with oral (sublingual) administration, indicates its high mucosal absorption from proximal parts of GI (e.g. oral cavity and esophagus).

Table 1: Effect of midazolam administration (0.3, 0.6, 1.2, 2.4, 4.8 mg/kg as sublingual spray). Onset time and duration of CNS suppression (since onset time of effect to the time of getting normal) were showed. Results are expressed as Mean±SE.

Dose Of Midazolam (mg/kg)	Onset Time Of Effect (min)	Duration Of Effect (min)
0.3	4.8 ± 0.77	24.3 ± 2.69
0.6	2.79 ± 0.24 **	25.5 ± 3.13
1.2	2.44 ± 0.24 **	26.7 ± 2.49
2.4	2.24 ± 0.21 ***	28.2 ± 3.48
4.8	2.15 ± 0.23 ***	37.2 ± 4.13

*** p<0.001, ** p<0.01 significantly different from the control group (Midazolam 0.3mg/kg).

Table 2: Effect of midazolam administration (0.3, 0.6, 1.2, 2.4, 4.8 mg/kg as sublingual spray). The highest rate of CNS suppression (Peak Score) & percentage of cats reached to seen peak score in each dose, also onset time and duration of peak score. Each group had at least 10 cats. Results are expressed as Mean±SE.

Dose Of Midazolam (mg/kg)	Observed Peak Score	Percentage Of Animals Reached Peak Score	Onset Time Of Peak Score (min)	Duration Of Peak Score (min)
0.3	2	100%	---	---
0.6	3	50%	6.52±1.07	6.02±2.06
	4	50%	7.76±1.16	9.28±1.4
1.2	3	30%	4.16±0.46	8.66±3.7
	4	70%	6.77±1.29	7.14±0.5
2.4	3	30%	5.3±0.66	13.33±1.76
	4	70%	5±0.43	8.41±0.89
4.8				
	3	20%	4.7±0.1	9±0.95
	4	80%	4.45±0.42	13.75±2.24

Table 3: Effect of ketamine administration (20, 40, 80 mg/kg as intra oral spray). Onset time and duration of CNS suppression (since onset time of effect to the time of getting normal) were showed. Results are expressed as Mean±SE.

Dose Of Ketamine (mg/kg)	Onset Time Of Effect (min)	Duration Of Effect (hour)
20	2.31 ± 0.41	0.56 ± 0.04
40	1.73 ± 0.23	1.87 ± 0.25 *
80	1.23 ± 0.16 *	2.41 ± 0.17 ***

*** p<0.001, * p<0.05 significantly different from the control group (Ketamine 20mg/kg).

Table 4: Effect of ketamine administration (20, 40, 80 mg/kg as intra oral spray). The highest rate of CNS suppression (Peak Score) & percentage of cats reached to seen peak score in each dose, also onset time and duration of peak score were showed. Each group had at least 10 cats. Results are expressed as Mean±SE.

Dose Of Ketamine (mg/kg)	Observed Peak Score	Percentage Of Animals Reached Peak Score	Onset Time Of Peak Score (min)	Duration Of Peak Score (min)
20	3	40%	4.9±0.7	11.15±1.9
	4	60%	3.63±0.7	23.46±6.9
40	3	50%	4.27±0.33	10.87±2.6
	4	50%	3.42±0.13	62.33±14.9
80	4	40%	2.86±0.4	86.2±19.8
	5	60%	2.32±0.6	114.4±24.8

Table 5: Effect of ketamine (20, 40, 80 mg/kg) & midazolam (0.3, 0.6, 1.2 mg/kg) co administration as sublingual spray. Onset time and duration of CNS suppression (since onset time of effect to the time of getting normal) were showed. Results are expressed as Mean±SE.

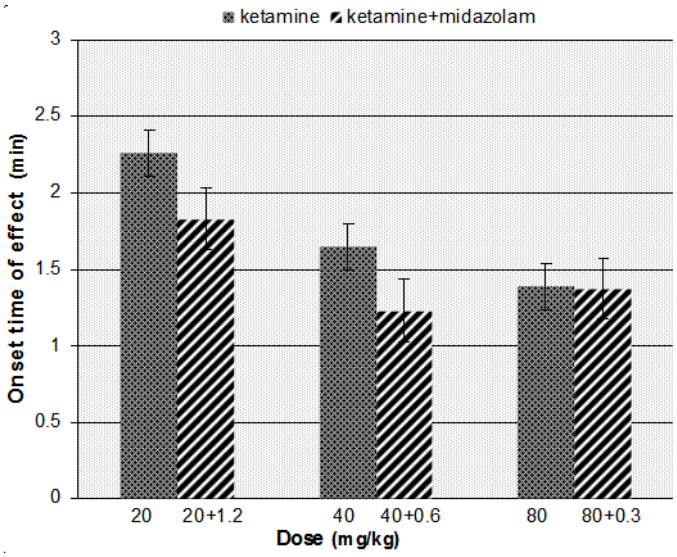
Ketamine (mg/kg)	+	Midazolam (mg/kg)	Onset Time Of Effect (min)	Duration Of Effect (hour)
20	+	1.2	1.83 ± 0.24	0.97 0.16
40	+	0.6	1.23 ± 0.21	† ***
80	+	0.3	1.37 ± 0.26	4.69 0.51 ***

*** p<0.001 significantly different from the control group (Ketamine 20mg/kg + midazolam 1.2 mg/kg).
†Times more than 6 hours was not recorded.

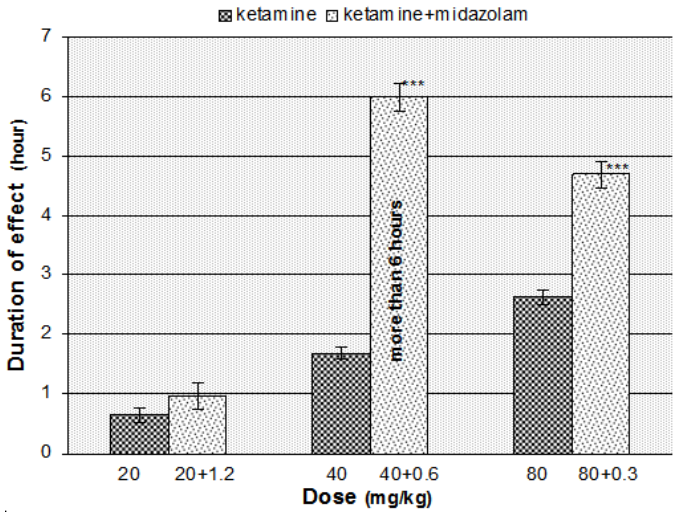
Table 6: Effect of ketamine (20, 40, 80 mg/kg) & midazolam (0.3, 0.6, 1.2 mg/kg) co administration as sublingual spray. The highest rate of CNS suppression (Peak Score) & percentage of cats reached to seen peak score in each dose, also onset time and duration of peak score were showed. Each group had at least 10 cats. Results are expressed as Mean±SE.

Ketamine (mg/kg)	+	Midazolam (mg/kg)	Observed Peak Score	Percentage Of Animals Reached Peak Score	Onset Time Of Peak Score (min)	Duration Of Peak Score (hour)
20	+	1.2	4	60%	3.35±0.36	1.01±0.2
			5	40%	2.95±0.73	1.87±0.22
40	+	0.6	4	50%	2.72±0.58	†
			5	50%	2.4±0.38	†
80	+	0.3	4	20%	5.3±0.5	1.65±0.55
			5	80%	2.22±0.39	3.9±0.52

†Times more than 6 hours was not recorded.



Graph. 1: Comparison onset time of CNS suppression between ketamine (20, 40, 80 mg/kg) & Ketamine (20, 40, 80 mg/kg) + Midazolam (0.3, 0.6, 1.2 mg/kg). Results are expressed as Mean±SE.



Graph. 2: Comparison duration of CNS suppression between ketamine (20, 40, 80 mg/kg) & Ketamine (20, 40, 80 mg/kg) + Midazolam (0.3, 0.6, 1.2 mg/kg). Results are expressed as Mean±SE. *** p<0.001

In other section of this study, sublingual spray of midazolam in different doses (even in very higher doses than recommended amounts) induced almost a weak & short-lasting CNS depression effects (maximum scores 4 in highest dose) maybe in result of its mechanism of action, low & slow distribution trough CNS in administered doses or etc. For clarify of it's exact mechanism need further studies.

But when this drug added to ketamine regime in mentioned doses (as sublingual spray), all of recorded parameters (except depth of CNS suppression) improved in comparison with ketamine and midazolam alone. So that it seems due to synergistic effects between them in suppression of CNS. These effects in group *ketmanie40mg/kg+midazolam0.6mg/kg* were considered significant (p<0/001) in compare with other combination protocols. But highest score of CNS suppression (score5) in *ketamine 80 mg/kg + midazolam 0.3 mg/kg* regime, was better & with highest percentage (80%) was seen. So that it seems ketamine has more effective role to induce peak score in combination protocols in compare with midazolam.

With due attention to that in present study, *ketamine 40 mg/kg + midazolam 0.6 mg/kg* protocol only could induce score5 in 50% of cats with undetectable duration of effect (more than 6 hours) and *ketmanie80mg/kg + midazolam0.3mg/kg* protocol induced analgesia (score5) in 80% of cats with short-lasting duration of effect (in compare) so that *ketmanie80mg/kg + midazolam0.3mg/kg* is more

suitable regime to induction anesthesia in cats as a non invasive method. But because of ketamine's hallucinogenic effect, when a mild sedation with short-last effect is needed, sublingual administration of midazolam in 2.4 or 4.8 mg/kg doses will be applicable non invasive method to sedate cats.

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