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Title: Guanabenz in the horse: Clinical effects and its comparison with other α_2 agonists

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Abstract:

In veterinary medicine, a number of the α2-receptor agonists are marketed as sedatives/hypnotics and analgesics, with their principal use being the chemical restraint of large and small animals. In human medicine, however, members of the α_2-receptor agonist family are used primarily as anti-hypertensive agents.

Guanabenz, an α2-receptor agonist, is marketed for humans as Wytensin®, an anti-hypertensive agent. Recently, there have been reports that guanabenz has been administered pre-race to horses in small doses (0.04 mg/kg) for its anti-hypertensive effects. In this study, intravenous administration of much large doses (0.2 mg/kg) of guanabenz produced a rapid and intense sedative effect, with sagging of the lower lip, sunken eyelids, and marked head droop. The head droop response was quantified as an indication of the intensity and duration of the pharmacological responses to guanabenz. Despite the intensity of the sedation, all horses remained standing and were able to walk following guanabenz administration, and the sedation and head droop responses were rapidly reversed by intravenous administration of yohimbine, consistent with suggestion that the major pharmacological responses to guanabenz are mediated through alpha-2 receptors. Furthermore, this investigation was extended to other members of the alpha-2 agonist group of agents, including clonidine.

These results strongly suggest that the α2-receptor agonists have abuse potential in racing horses, therefore the pharmacological responses of guanabenz, amitraz, clonidine, detomidine, medetomidine, romifidine, and xylazine were compared. While all of these agents showed qualitatively similar pharmacological responses, only guanabenz produced an unusually intense and prolonged analgesic response. These studies show that a 100 mg i.v. dose of guanabenz can rapidly induce sedation and analgesia that are more intense and considerably longer-lasting than that produced by other α2-receptor agonists. These experiments suggest considerable clinical potential for guanabenz as a sedative and a relatively long-lasting analgesic in equine medicine.

Introduction:

The autonomic nervous system controls involuntary body functions and elicits either inhibitory or excitatory responses within the parasympathetic and sympathetic divisions. The sympathetic division also elicits either inhibitory or excitatory effects with different receptors for the adrenergic agonists divided into subtypes α and β. Both subtypes have been further subdivided; in particular, the α subtype is divided into α_1 and α_2 receptors. The α_2 receptors are prejunctional inhibitory receptors within the sympathetic nervous system[1].

The α_2 receptor agonists, of which clonidine is the best-known example, were first synthesized in the early 1960's and found to produce vasoconstriction when applied topically, apparently mediated through α_2 receptors. During clinical testing of clonidine as a nasal decongestant, it caused hypotension, sedation, and bradycardia in man, which led to its introduction as an antihypertensive in human medicine. The α_2 agonists reduce arterial blood pressure by mediating both cardiac output and peripheral resistance[2]. One outcome of this study was that among these agents, guanabenz is particularly and uniquely effective as a long-lasting sedative and analgesic agent in the horse.

Guanabenz (Wytensin®, Wyeth Pharmaceuticals, Collegeville, PA) has been marketed for human use for a number of years as a centrally acting α_2 agonist to decrease blood pressure. Presumably, its well-identified action as an anti-hypertensive in human medicine led to its administration in small doses (0.04mg/kg) as an anti-hypertensive agent in horses.

The rationale for this administration is that most horses experience pulmonary hypertension during running, leading to exercise induced pulmonary hemorrhage (EIPH), a considerable problem in the horse racing industry. EIPH acutely interferes with the racing performance of horses by compromising the exchange of O_2 and CO_2 in the alveolar capillaries, and repeated bouts of EIPH result in chronic and cumulative damage to the lung[3]. For these reasons, horsemen have long sought effective prophylactic approaches to the alleviation or prevention of EIPH.

Among the possible prophylactic approaches to EIPH, equine veterinarians and horsemen reportedly have administered ground-up guanabenz tablets re-suspended in an alcohol solution[4]. This is administered in small (10-20 mg) doses shortly before post time to reduce the intensity of the racing-related pulmonary hypertension and, by extension, the associated EIPH.

Guanabenz is listed by the Association of Racing Commissioners International as an ARCI class 3 agent[5]. As such, guanabenz is considered to have significant potential to influence the outcome of a race, and its administration to a horse shortly before post time would clearly contravene the rules of racing in most jurisdictions. The objective of this study was to determine the pharmacological effects of guanabenz and other α_2 adrenergic agonists including xylazine, detomidine, medetomidine, clonidine, romifidine, and amitraz. Pharmacological effects measured included head droop, analgesia, heart rate, and urine output.

Materials and Methods

Horses

Five mature Thoroughbred mares weighing 448 to 489 kg were used for this study. The animals were maintained on grass hay and feed (12% protein), which was a 50:50 mixture of oats and an alfalfa-based protein pellet. Horses were fed twice a day. The animals were vaccinated annually for tetanus and were dewormed quarterly with ivermectin (MSD Agvet, Rahway, NJ). A routine clinical examination was performed before each experiment to assure that the animals were healthy and sound. Because of the critical role of superficial skin temperature in these experiments, no heat lamp experiments were performed when the ambient temperature was less than 10°C. Animals used in these experiments were managed according to the rules and regulations of the University of Kentucky Institutional Animal Care Use Committee, which also approved the experimental protocol.

Pharmacologic effects were determined following intravenous administration of 0.2 mg/kg of guanabenz acetate (approximately equivalent to 100 mg guanabenz to a 1,200 lb horse) dissolved in 3 ml ethanol. Head droop, hoof withdrawal reflex latency, locomotor activity, heart rate, and urine chemistry were measured. These results were compared to similar effects measured following administration of broadly equivalent doses of other α_2 agonists including: amitraz (0.15 mg/kg), clonidine (0.02 mg/kg), detomidine (0.04 mg/kg), medetomidine (0.01 mg/kg), romifidine (0.1 mg/kg, and xylazine (1mg/kg). Although guanabenz and clonidine were administered to five horses each, the comparative doses of other agents were administered to one horse, and no statistical analyses were performed. At least one week elapsed between all treatments.

Measurement of pharmacological effects

Head droop

Sedation was assessed by measuring the degree of head droop following administration of α_2 agonists. A pre-treatment height (from floor to chin) was determined at -10, -5 and 0 min before intravenous injection of the α_2 agonists, and head droop was measured periodically during the subsequent 1.5-8 h. In a separate experiment to evaluate the reversal effect of an α_2 receptor antagonist, the same protocol was followed as described above, but 30 min after injection of guanabenz and clonidine the horses received intravenous injections of 0.12 mg/kg yohimbine powder dissolved in 2 ml of dimethyl sulfoxide (DMSO).

Analgesia

Thermal antinociception, which has been used as a measure of analgesia, was determined with a heat projection lamp described previously[6]. Briefly, focused radiant light/heat was used as a noxious stimulus and was directed onto the pastern of the horse, from a constant distance, to elicit the classic flexion-withdrawal reflex. Hoof withdrawal reflex latency (HWRL) was defined as the time between lamp illumination and withdrawal of the hoof. The reflex times were adjusted by varying the intensity of the heat output with a rheostat so that the HWRL for control measurements was 3-4 sec, with the actual HWRL recorded on an electronic timer built into the lamp. The duration of light exposure to the pasterns was limited to 10 sec to prevent skin damage. A secondary unfocused light beam (sham light) was used to confound the horse, reducing the possibility that the flexion-withdrawal reflex was to visual rather than thermal perception of the focused light beam.

HWRL was measured at -30 and -15 min and immediately before injection, and these times (-30, -15, and 0 min) were used to establish a baseline value for HWRL in each horse. The HWRL was then measured at 7.5, and 15 min after injection and every 15 min until HWRL returned to control values. The HWRL was expressed as a percent of the baseline values.

Heart Rate

Heart rates (HR) were recorded at 1 min intervals during each experiment by an on-board heart rate computer (Polar CIC Inc, Port Washington, NY). An elastic strap with a receiver and transmitter attached was placed around the chest of the horse. The transmitter was connected to two electrodes placed on shaved areas of the sternum and left side of the anterior chest. Electrode gel was used to insure proper conduction of the HR signal.

Urine chemistry

Urine glucose, production, and specific gravity were monitored following intravenous injection of the α_2 agonists. Urine was collected using a Foley catheter for 8 hr after administration. The catheter was drained into a volumetric flask to measure urine volume. Urine specific gravity was measured with a refractometer (URC-NE, Atago, Japan), and glucose was measured with DiaScreen 3 urine sticks (ChroniMed, Minneapolis, MN).

Results

Although horses remained standing and were able to walk following IV guanabenz administration (0.2 mg/kg), the agent produced rapid, profound sedation as evinced by relaxation of the lower lip, sunken eyelids, and extreme head droop (Figure 1). As suggested by the rapidity of the head droop, the horses were clinically depressed within minutes of the guanabenz administration, and the profound head droop persisted for 3.5 h after injection (Figure 1a). This figure also illustrates the rapid reversal following yohimbine injection; 2 min after IV injection yohimbine (0.12 mg/kg), head height was ~50% of pre-treatment value, and by 5 min after injection, head height was within normal limits, and the horse was clinically alert. Clonidine and

detomidine also produced profound head droop for 1-1.5 h, with the other α_2 agonists producing head droop of shorter duration.

Figure 2 illustrates the rapid onset of analgesia following injection of some of the α_2 agonists. Intravenous administration of 0.2 mg/kg of guanabenz provided maximal measurable analgesia (300% of control value) by 10 min after injection, which persisted through the 0.75 h testing point. Analgesia had returned to control values by 6 h after administration, by far the longest analgesic response to any of the α_2 agonists evaluated (Figure 2a). Medetomidine, romifidine, and xylazine provided maximal analgesia for 0.5, 0.25, and 0.25 h respectively (Figure 1b-d). Detomidine and clonidine failed to provide maximal analgesia, although the limited analgesic effect persisted for ~1.5 and 0.75 h, respectively (Figures 2e-f).

Figure 3 shows the decreased HR following injection of guanabenz, which persisted for ~3.5 h. Amitraz and romifidine decreased HR for ~1.5 h. Clonidine, detomidine, medetomidine, and xylazine decreased HR for ~1.5, 1.5, 1, and 0.75 h, respectively.

Guanabenz produced a significant hyperglycemia (data not shown) and glucosuria (normal blood and urine glucose ranges are 70-140 and 0 mg/dL, respectively)[7]. At ~2 h after administration, the transport maximum for glucose was apparently reached in the kidney tubules, and glucose started to appear in the urine (Figure 4a). Urine glucose continued to rise late in the experiment, even though blood glucose returned to normal values by 8 h after administration. Amitraz, romifidine, and detomidine also produced a glucosuria.

The hyperglycemia and the corresponding glucosuria resulted in a significant diuresis, as shown by the cumulative urine volume. Peak production occurred between 1.5 and 3 h after administration, as indicated by the steeper slope of the urine volume curve during that period. Thereafter, production was minimal. Similarly, urine specific gravity dropped to a low of about 1.006 at 2 h after administration, remained at this level for ~1 h, and gradually returned to control value. Note that the lowest specific gravity values corresponded with peak urine production. Urine pH remained at 8, and urine protein was negative throughout the testing (data not shown).

The other α_2 agonists also caused an increased urine production with a concomitant decrease in specific gravity. The effect of guanabenz (Figure 4a) had the longest duration on increased urine volume, lasting about 3 h. Xylazine (Figure 4g) had the shortest polyuric effect, persisting for about 1 h.

Discussion

Sedation

The sedative effects of the α_2 agonists were similar and included lowering of the head, drooping of the eyelids and lower lip, and ataxia. Male horses occasionally relax the penis[8].

Different studies have used various criteria to measure sedation[9-12]. The sedative effects of many of the α_2 agonists have been quantified with an equine locomotor chamber[13]. Amitraz (25 mg, iv) significantly reduced locomotor activity for about 120 min after administration. Larger doses (50 and 75 mg, iv) reduced spontaneous activity for 180 and 240 min, respectively. Detomidine (10 and 20 mg, iv) reduced locomotor activity for about 120 and 240 min, respectively, and xylazine (167 and 500 mg, iv) reduced locomotor activity for about 15 and 30 min, respectively. Guanabenz (100 mg, iv) significantly reduced locomotor activity for about 240 min after administration[14].

In a similar study measuring head droop[15], amitraz produced a dose-dependent response. The lowest dose (0.05 mg/kg) produced significant droop from 5-60 min after administration, and the highest doses administered (0.15 mg/kg) produced significant head droop from 5-150 min after administration.

Analgesia

It is difficult to compare the efficacy of analgesic agents between studies. Pain can be superficial, deep, or visceral, and the different types of pain have varying responses to analgesic drugs. Xylazine and detomidine are routinely used for the relief of colic (visceral) pain in horses. One study found that romifidine produced limited analgesia for visceral pain in a dose-related manner[a], and another study concluded that the limited analgesic effect was neither dose-related nor consistent[b]. One study comparing romifidine and detomidine concluded that the analgesic effects of both agents were similar[c]; another comparative study concluded that romifidine had no analgesic effect on peripheral pain[16]. Medetomidine has not been evaluated previously as an analgesic agent in horses, although its analgesic effect has been well demonstrated in other species[17]. In a study that used a heat lamp similar to the one described in this paper, amitraz produced analgesia to peripheral pain in a dose-related manner[15]. The least administered dose (0.05 mg/kg) produced significant anesthesia 30-45 min after injection. The highest administered dose (0.15 mg/kg) produced significant analgesia from 5-150 min after injection.

Urine chemistry

A dose-dependent hyperglycemia has been noted following administration of xylazine[18], detomidine[19], and romifidine[d], and elevated levels of urine glucose have been measured in some, but not all, of the studies. The increased urine production has been attributed to an osmotic diuresis from spillover of glucose into the urine for some α_2 agonists. However, there is a suggestion that the polyuria is also mediated by an inhibition of arginine vasopressin release, which could explain the increased urine volume following administration of α_2 agonists that do not show glucosuria (e.g., medetomidine, clonidine, xylazine; Figure 4)

Conclusion:

These studies suggest that intravenous guanabenz (0.2 mg/kg) induces sedation more rapidly than that produced by other α_2 agonists, and that sedation and analgesia were generally more intense and always considerably longer-lasting than responses to other members of this group. These experiments suggest considerable clinical potential for guanabenz as a sedative and as a relatively long-lasting analgesic in equine medicine.

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Figure 1 Head droop following IV injection of the α_2 agonist agents. The sedative effects of a) guanabenz and b) clonidine were rapidly reversed by yohimbine.

Figure 2 Hoof Withdrawal Reflex Latency (HWRL) following IV injection of the α_2 agonists.

Figure 3 Heart rates following IV administration of the α_2 agonists.

Figure 4 Cumulative urine volumes, specific gravity, and urine glucose following IV administration of the α_2 agonists.

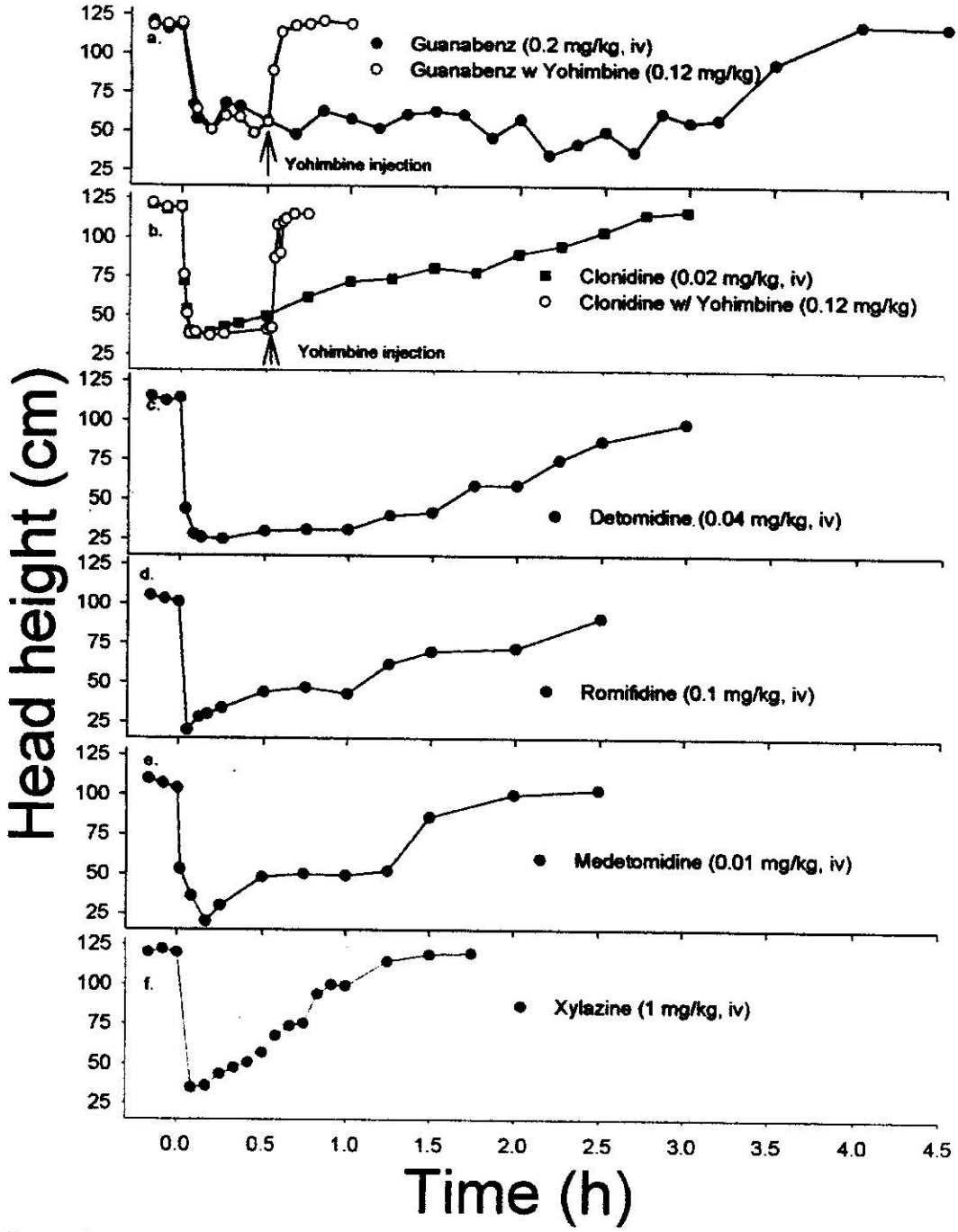
Head height (cm)
Time (h)
a.
Guanabenz (0.2 mg/kg, iv)
Guanabenz w Yohimbine (0.12 mg/kg)
Yohimbine injection
b.
Clonidine (0.02 mg/kg, iv)
Clonidine w/ Yohimbine (0.12 mg/kg)
Yohimbine injection
c.
Detomidine (0.04 mg/kg, iv)
d.
Romifidine (0.1 mg/kg, iv)
e.
Medetomidine (0.01 mg/kg, iv)
f.
Xylazine (1 mg/kg, iv)
125
100
75
50
25
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
4.0
4.5

Figure 1

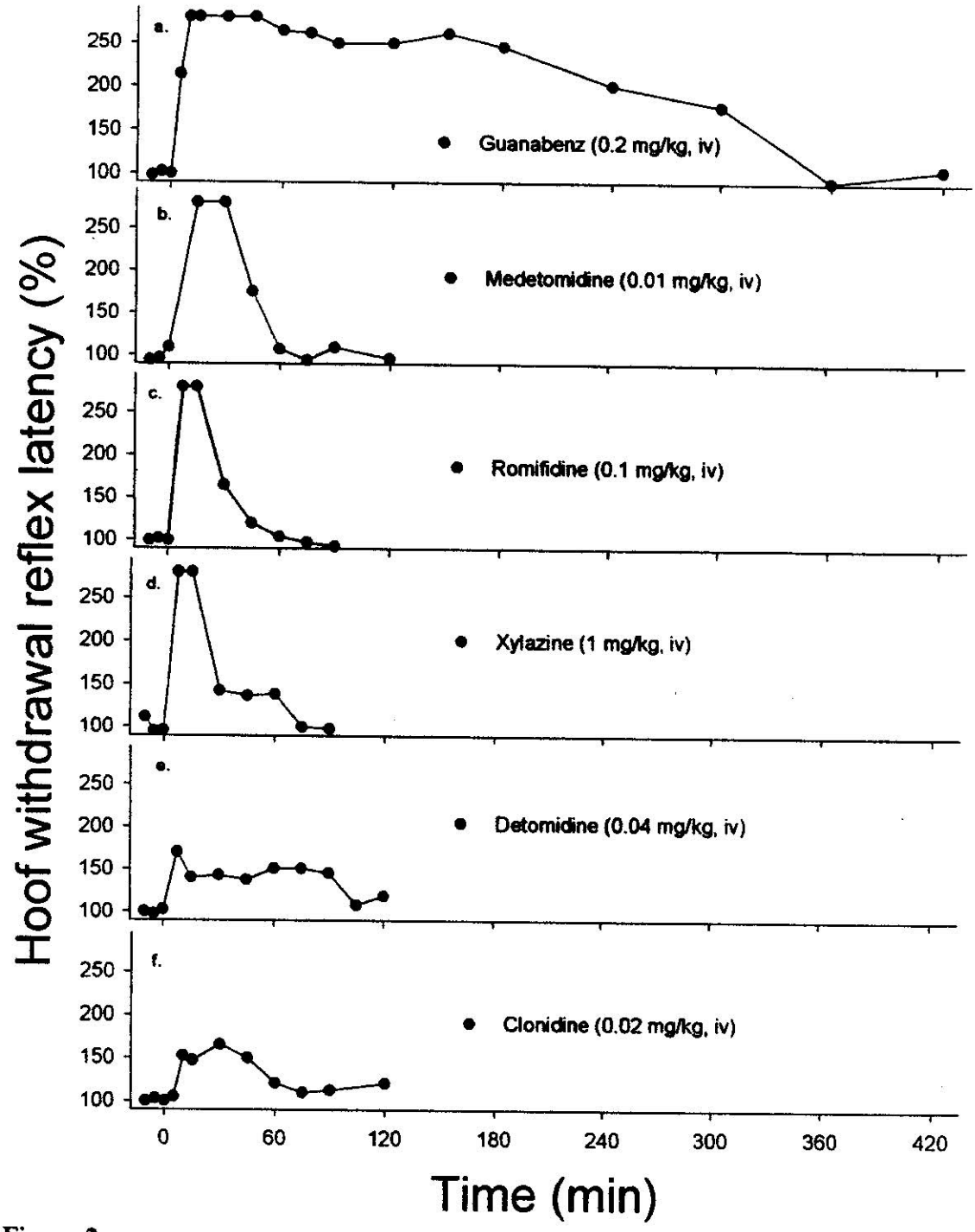
Hoof withdrawal reflex latency (%)
a.
Guanabenz (0.2 mg/kg, iv)
b.
Medetomidine (0.01 mg/kg, iv)
c.
Romifidine (0.1 mg/kg, iv)
d.
Xylazine (1 mg/kg, iv)
e.
Detomidine (0.04 mg/kg, iv)
f.
Clonidine (0.02 mg/kg, iv)
250
200
150
100
0
60
120
180
240
300
360
420
Time (min)

Figure 2

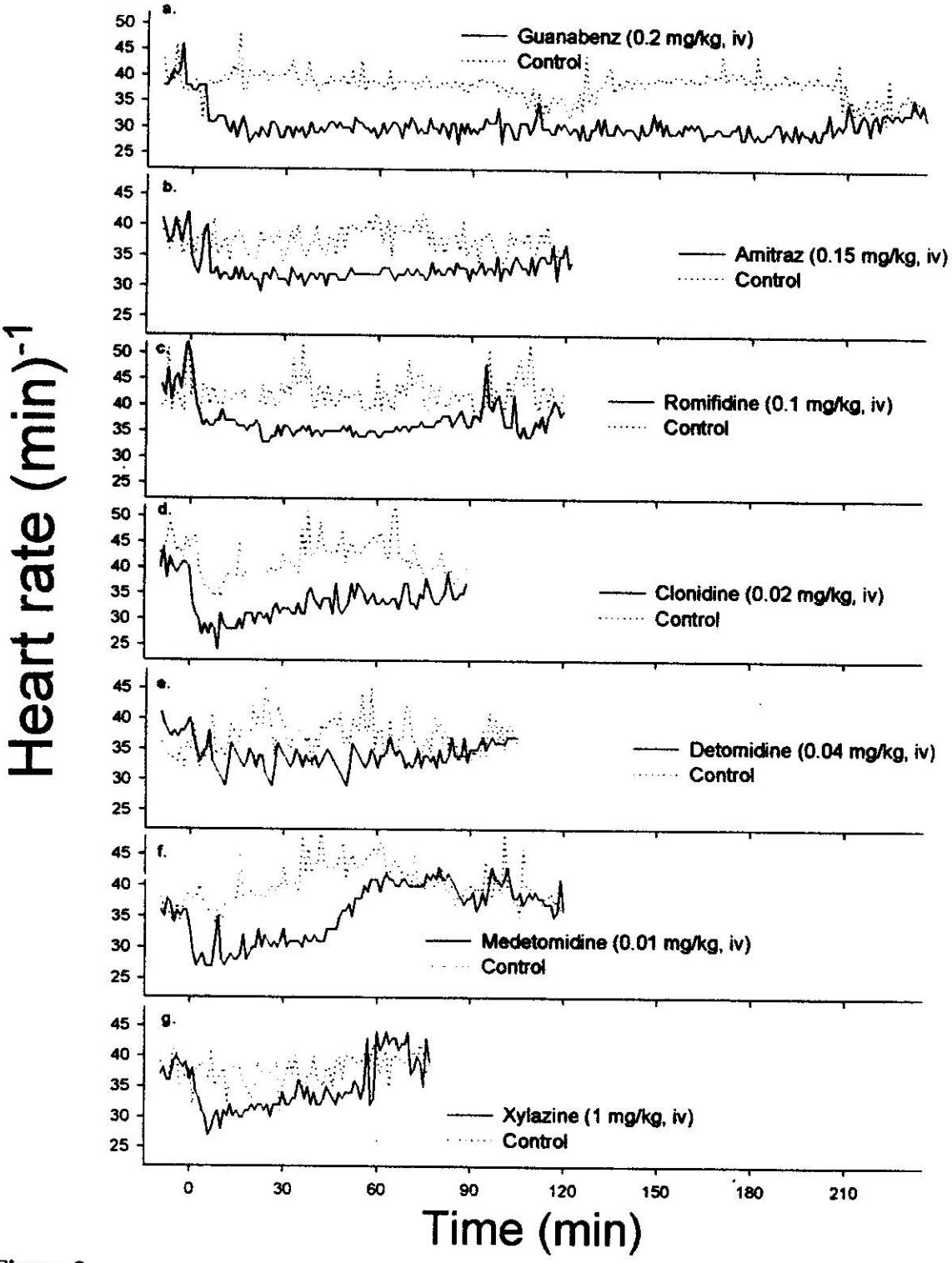
a.
Guanabenz (0.2 mg/kg, iv)
Control
b.
Amitraz (0.15 mg/kg, iv)
Control
c.
Romifidine (0.1 mg/kg, iv)
Control
d.
Clonidine (0.02 mg/kg, iv)
Control
e.
Detomidine (0.04 mg/kg, iv)
Control
f.
Medetomidine (0.01 mg/kg, iv)
Control
g.
Xylazine (1 mg/kg, iv)
Control
Heart rate (min)-1
Time (min)
0
30
60
90
120
150
180
210

Figure 3

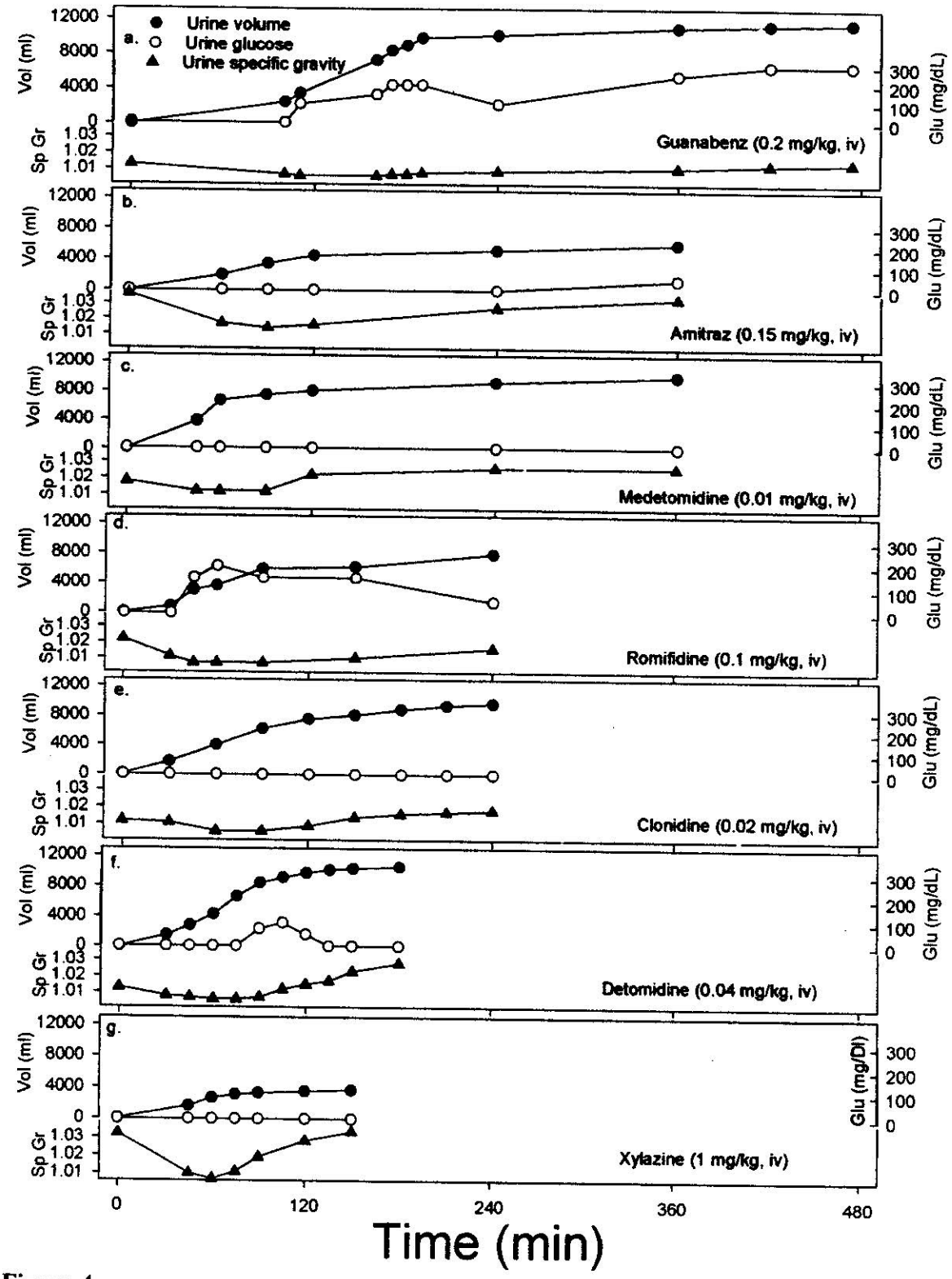
a.
Urine volume
Urine glucose
Urine specific gravity
Guanabenz (0.2 mg/kg, iv)
b.
Amitraz (0.15 mg/kg, iv)
c.
Medetomidine (0.01 mg/kg, iv)
d.
Romifidine (0.1 mg/kg, iv)
e.
Clonidine (0.02 mg/kg, iv)
f.
Detomidine (0.04 mg/kg, iv)
g.
Xylazine (1 mg/kg, iv)
Vol (ml)
Sp Gr
Glu (mg/dL)
0
120
240
360
480
Time (min)

Figure 4