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Effects of vatinoxan on cardiorespiratory function, fecal output and plasma drug concentrations in horses anesthetized with isoflurane and infusion of medetomidine

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ABSTRACT

A constant rate infusion (CRI) of medetomidine is used to balance equine inhalation anesthesia, but its cardiovascular side effects are a concern. This experimental crossover study aimed to evaluate the effects of vatinoxan (a peripheral α2-adrenoceptor antagonist) on cardiorespiratory and gastrointestinal function in anesthetized healthy horses. Six horses received medetomidine hydrochloride 7 μg/kg IV alone (MED) or with vatinoxan hydrochloride 140 μg/kg IV (MED + V). Anesthesia was induced with midazolam and ketamine and maintained with isoflurane and medetomidine CRI for 60 min. Heart rate, carotid and pulmonary arterial pressures, central venous pressure, cardiac output and arterial and mixed venous blood gases were measured. Selected cardiopulmonary parameters were calculated. Plasma drug concentrations were determined. Fecal output was measured over 24 h. For statistical comparisons, repeated measures analysis of covariance and paired *t*-tests were applied.

Heart rate decreased slightly from baseline in the MED group. Arterial blood pressures decreased with both treatments, but significantly more dobutamine was needed to maintain normotension with MED + V ($P = 0.018$). Cardiac index (CI) and oxygen delivery index (DO_2I) decreased significantly more with MED, with the largest difference observed at 20 min: CI was 39 ± 2 and 73 ± 18 ($P = 0.009$) and DO_2I 7.4 ± 1.2 and 15.3 ± 4.8 ($P = 0.014$) mL/min/kg with MED and MED + V, respectively. Fecal output or plasma concentrations of dexmedetomidine did not differ between the treatments. In conclusion, premedication with vatinoxan induced hypotension, thus its use in anesthetized horses warrants further studies. Even though heart rate and arterial blood pressures remained clinically acceptable with MED, cardiac performance and oxygen delivery were lower than with MED + V.

Introduction

Medetomidine and its active enantiomer dexmedetomidine are potent α2-adrenoceptor agonists (α2-agonists) with a short half-life in horses (Bettschart-Wolfensberger et al., 1999; Ranheim et al., 2015). Thus, they are interesting for use as constant rate infusion (CRI) in equine anesthesia, albeit they currently only have Market Authorisation for dogs and cats. A medetomidine CRI reduces the minimal alveolar concentration of inhalation agents (Bettschart-Wolfensberger et al., 2001; Ringer et al., 2007) and improves quality of recovery from general anesthesia in horses (Ringer et al., 2007; Marcilla et al., 2012). The typical, α2-agonist-induced cardiovascular side effects, such as increased systemic vascular resistance (SVR), bradycardia and decreased cardiac index (CI; Flacke et al., 1990; Yamashita et al., 2000), are a concern for its use (Risberg et al., 2016). Furthermore, in conscious horses, medetomidine reduces gastrointestinal motility (Rezende et al., 2015), which may already be decreased by general anesthesia, predisposing the animals to post-anesthetic colic.

Vatinoxan (previously MK-467 or L-659,066) mainly blocks α2-adrenoceptors located outside of the central nervous system (Clineschmidt et al., 1988). In standing horses, vatinoxan alleviated detomidine- and romifidine-induced vasoconstriction, the related bradycardia, and gastrointestinal hypomotility

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without a significant effect on sedation (Vainionpää et al., 2013; de Vries et al., 2016). Intravenous (IV) dosing of vatinoxan in combination with detomidine for premedication before isoflurane anesthesia, however, induced severe hypotension in horses (Pakkanen et al., 2015). In another study, IV vatinoxan reversed the α2-agonist-induced vasoconstriction and improved CI during general anesthesia (maintained with isoflurane and xylazine or dexmedetomidine CRI) in horses, but also resulted in hypotension and reduced tissue perfusion (Wittenberg-Voges et al., 2018). Both studies concluded that the optimal dose of vatinoxan for premedication for general anesthesia needs to be determined in horses (Pakkanen et al., 2015; Wittenberg-Voges et al., 2018).

This study aimed firstly to evaluate the effects of vatinoxan on cardiorespiratory function during general anesthesia maintained with isoflurane and medetomidine CRI; secondly, the effects of vatinoxan on post-anesthetic gastrointestinal function in horses was investigated. Because vatinoxan has been shown to alter the pharmacokinetics of some drugs, including α2-agonists in dogs (Honkavaara et al., 2012) and horses (Vainionpää et al., 2013; Pakkanen et al., 2015; de Vries et al., 2016), plasma concentrations of medetomidine enantiomers, vatinoxan and induction drugs were also analysed.

Materials and methods

Horses

The study was approved by the National Animal Experiment Board of Finland (License number ESAVI/4789/04.10.07/2014; Approval date 19 June 2014).

Six horses (four Standardbreds, two Warmbloods; three mares and three geldings) with a median age of 10 years (range 5–19 years) and a median bodyweight (bwt) of 508 kg (range 451–614 kg) were studied. The horses were considered healthy on the basis of results of physical examination and routine hematology tests. At least 2 months before the start of this study, the right common carotid arteries of the horses were relocated subcutaneously (Tapio et al., 2017). Exclusion criteria included signs of systemic illness and abnormal findings in cardiovascular status. The sample size was chosen based on results from previous comparable studies (Vainionpää et al., 2013; Tapio et al., 2018) indicating that a sample size of six horses would enable detection of clinically relevant differences in cardiorespiratory parameters.

To determine the baseline values, the mean daily fecal output for each horse was measured by collecting and weighing feces over a 3-day period. The median of three measurements, obtained on separate days before the anesthesia for another study (Tapio et al., 2018), was used as the baseline value for each horse for heart rate (HR), rectal temperature, cardiac output ($\dot{Q}t$), systolic, mean and diastolic (SAP, MAP and DAP, respectively) arterial blood pressure, systolic, mean and diastolic pulmonary arterial blood pressure (SPAP, MPAP and DPAP, respectively), mean central venous pressure and arterial and venous blood gas analysis.

The horses had unlimited access to water and were fed hay. Feed, but not water, was withheld for 12 h before anesthesia and 6 h after recovery.

Study design

Each horse received two treatments in a randomized order separated by a minimum of 14 days. Simple randomization was performed by assigning the treatment order to each horse using a masked method, starting alternately with each treatment.

Instrumentation

Catheter sites were subcutaneously infiltrated with a small amount of mepivacaine (Scandicain 2%, AstraZeneca). Intravenous jugular catheters (Intraflon, Laboratoires Pharmaceutiques Vygon), central venous catheters (Cavafix Certo, B. Braun) and pulmonary arterial catheters (Arrow-Berman angiographic catheter, Arrow International) were aseptically placed before each anesthesia and before administration of premedication. The correct locations of the central venous catheter and pulmonary arterial catheter were verified by characteristic pressure wave forms on the monitor screen (S/5 compact Critical Care Monitor, Datex Ohmeda). Transducer accuracy was verified against a mercury manometer in the measurement range prior to use. An arterial catheter (BD arterial cannula, Becton-Dickinson Critical Care Systems) was placed into the right common carotid artery after the induction of anesthesia.

Anesthesia

Horses received an IV bolus of medetomidine hydrochloride (Dorbene, Vetcare) 7 μg/kg alone (MED) or with (MED + V) vatinoxan hydrochloride 140 μg/kg. Medications were diluted in saline (Natriumklorid 0.9%, B. Braun) to achieve a total volume of 20 mL, and the bolus was given over 15 s. Anesthesia was induced 10 min later, at T0, with midazolam (Midazolam Accord, Accord Healthcare Ltd.) 0.06 mg/kg and ketamine (Ketador, Richter Pharma AG) 2.2 mg/kg given as an IV bolus over 15 s. After endotracheal intubation, the horse was positioned in dorsal recumbency on a padded surgical table. Anesthesia was maintained with isoflurane (IsoFlo, Zoetis) in 100% oxygen, initiated immediately after connecting the horse to the breathing circuit (Tafonius, Hallowell EMC), and medetomidine CRI at 3.5 μg/kg/h, initiated 10 min after induction (T10). The expiratory isoflurane was set at 1.2 vol% and adjusted in the presence of spontaneous nystagmus or absence of palpebral reflex by 0.1%. All horses were mechanically ventilated with an initial rate of 8 breaths/min and a tidal volume of 12 mL/kg. Ventilation was adjusted to maintain target expiratory CO_2 (P_ECO_2) at 35–55 mmHg. Dobutamine infusion (Dobuject, Primex Pharmaceuticals) was started at T10 at a rate based on the first MAP reading (Table 1). The dose rate was then doubled every 5 min until MAP was in the target range of 70–80 mmHg by a blinded investigator (MRR). When MAP was >90 mmHg, the dobutamine infusion was stopped. Ringer's solution (Fresenius Kabi AB) was administered at a rate of 3 mL/kg/h. The medetomidine CRI was continued for 60 min (until T70). The horse was then transferred to the recovery box and extubated once swallowing. The recovery was unassisted.

Data collection

Sedation was scored prior to premedication (baseline) and 1 min prior to induction of anesthesia (T-1) by a blinded investigator (MRR) with a range of possible scores from 0 (no sedation) to 10 (heavy standing sedation) (Rohrbach et al., 2009). Time from administration of induction drugs to lateral recumbency was recorded, and quality of induction was scored (Rossetti et al., 2008) from 0 (excellent) to 5 (poor). During anesthesia the following parameters were recorded every 5 min, from T10 through to T70: HR, SAP, MAP, DAP, Systolic Pulmonary Arterial Pressure (SPAP), Mean Pulmonary Arterial Pressure (MPAP), Diastolic Pulmonary Arterial Pressure (DPAP), inspired oxygen fraction and P_ECO_2. Central venous pressure, $\dot{Q}t$ and arterial and mixed venous blood gas analysis were recorded at T20, T40 and T70. Cardiac output was determined by lithium dilution method (LiDCO Plus, LiDCO) as previously described (Hallowell and Corley, 2005). The initially entered standard values of 6.2 mmol/L of hemoglobin and 140 mmol/L of sodium were replaced by the measured values for the calculations. Arterial and mixed venous blood was anerobically collected, placed in iced water and analysed (ABL800 Flex, Radiometer Medical) within 10 min of sample collection. The values were temperature-corrected on the basis of concurrently recorded rectal temperature.

The following variables were calculated afterwards: cardiac index (CI), stroke volume index (SVI), systemic resistance index (SVRI), pulmonary vascular resistance index (PVRI), left ventricular workload (LVW), arterial (CaO_2) and mixed venous ($C\bar{v}O_2$) oxygen contents, venous admixture, arterial (SaO_2) and mixed venous ($S\bar{v}O_2$) blood oxygen saturation, oxygen delivery index (DO_2I), oxygen consumption (VO_2I), oxygen extraction (O_2ER) and alveolar dead space ventilation (VD_{alv}:V_T; Boyd et al., 1991; Clerbaux et al., 1993; Hardman and Aitkenhead, 2003; Haskins et al., 2005; see Supplement, Formulae for the calculated parameters).

The total amount of dobutamine administered was recorded. Recovery was observed and the times from termination of anesthesia to extubation, sternal recumbency and standing were recorded. Horses were observed at least every other hour and the fecal output was determined for 24 h after recovery. In addition, bronchoalveolar lavage samples related to another study were collected without medication from the same horses for 156 h after anesthesia; these results will be published elsewhere.

Arterial blood samples (10 mL) for analysis of plasma concentrations dexmedetomidine, levomedetomidine, vatinoxan, midazolam and ketamine were collected into EDTA-containing tubes at T-1, T20, T40 and T70 (see Supplement).

Statistical analysis

Statistical analyses were performed with commercial software (SAS, SAS Institute Inc., and SPSS, IBM).

Table 1
Starting rate of dobutamine infusion in relation to the initial mean arterial pressure value obtained from the arterial catheter placed in the right common carotid artery.

Mean arterial pressure (mmHg)	Rate of dobutamine infusion (μg/kg/min)
60–69	0.5
50–59	1
40–49	2
<40	4

Change from baseline was used as the response with all other cardiorespiratory variables, except for those measured only during anesthesia (venous admixture and $VD_{alv}:V_T$), SaO_2 and $S\bar{v}O_2$. The differences between treatments were assessed with repeated measures analysis of covariance. The model consisted of a baseline covariate, the main effects of treatment, period, sequence and time point of measurement and two-way interactions of period*time point, sequence*time point and treatment*time point as fixed effects and the main effect of horse and two-way interaction terms of period*horse and time point*horse as a random effect. For venous admixture and $VD_{alv}:V_T$, the same model as described above, excluding the baseline covariate and using the actual values instead of change as the response, was used. The normality assumptions were checked using Kolmogorov–Smirnov tests. For SaO_2, $S\bar{v}O_2$, SPAP, MPAP and DPAP, the normality could not be verified, thus non-parametric Wilcoxon signed-rank sum testing by time point was performed. Estimates of treatment effect and within treatment changes were calculated from the fitted models over time and by time point using model contrasts.

For the amount of dobutamine, plasma drug concentrations, total fecal output, induction and recovery times, the normality assumptions were confirmed with Shapiro–Wilk test, and paired *t*-test was then applied to investigate differences between the treatments. For the sedation and induction scores, Wilcoxon signed-rank sum test was applied to investigate differences between the treatments.

Statistical significance was set at $P < 0.05$.

Results

There were no adverse events during the experiment, and none of the horses were excluded from the study.

All horses were sedated after medetomidine (Table 2). Induction was significantly faster with MED + V than with MED alone (Table 2).

Compared to baseline, HR was significantly higher at T10 with MED + V and significantly lower at T20, 40 and 60 with MED. These changes from baseline were significantly different between the two treatments only at T10 (Table 3). Overall, arterial blood pressures significantly decreased from baseline within both treatments, but significantly more dobutamine was administered with MED + V than with MED to maintain MAP in the target range (Table 2). The range of the dobutamine CRI rate was 0–1 μg/kg/min with MED and 0–4 μg/kg/min with MED + V. Carotid arterial pressures decreased significantly more from baseline with MED + V than with MED at their first measurement at T10 (Table 3). Thereafter (i.e. after initiation of dobutamine CRI), SAP tended to be higher and DAP lower with MED + V than with MED but the change from baseline differed significantly between the treatments only at T30, 40 and 70 for SAP and T40 for DAP. Pulmonary arterial pressures differed between treatments only at T30 and T40; SPAP decreased significantly more from baseline with MED than with MED + V (Table 3). There were no significant changes in PVRI overall (Table 3). Central venous pressure significantly decreased from baseline with both treatments. Furthermore, significant decreases from baseline were observed in CI, SVI and DO_2I and significant increases in SVRI and O_2ER with MED, while with MED + V only CI decreased from baseline at T70 (Table 4). Of these parameters, the changes from baseline differed between the treatments only with CI, DO_2I and O_2ER. Left ventricular workload decreased significantly more from baseline with MED than with MED + V (Table 4). Conversely, no changes in Hb, $P\bar{v}O_2$, $C\bar{v}O_2$ and CaO_2 were observed with MED, whereas they transiently increased from baseline with MED + V (Table 4). No differences were detected between the treatments in the increase of PaO_2 or the decrease of VO_2I from baseline during anesthesia or in $VD_{alv}:V_T$ or venous admixture (Table 4). The arterial blood O_2 saturation was ≥98% in all horses and there was no significant difference between the treatments (data not shown); $S\bar{v}O_2$ was significantly higher with MED + V than with MED (Table 4).

All recoveries were uneventful, and there were no differences in recovery times between the treatments (Table 1). Fecal output was markedly decreased after anesthesia without any difference between the treatments (Table 1). None of the horses developed any clinical signs of abdominal discomfort.

Of the measured plasma drug concentrations, only ketamine concentrations differed significantly between treatments; they were lower in the MED + V group (see Supplement, Plasma drug concentration analysis).

Table 2
Median (minimum, maximum) sedation and induction scores and mean (± standard deviation) induction and recovery times, amount of dobutamine administered and total fecal output of six adult horses. The horses were administered as premedication medetomidine 7 μg/kg alone (MED) or with vatinoxan 140 μg/kg (MED + V) and sedation was scored 1 min before induction (T-1). Anesthesia was induced with midazolam 0.06 mg/kg and ketamine 2.2 mg/kg 10 min after premedication and maintained with isoflurane and medetomidine CRI 3.5 μg/kg/h for 1 h. Dobutamine was administered to maintain normotension according to a previously defined intervention plan. Times for recovery were recorded from the cessation of CRI. Total fecal output was recorded over 24 h after anesthesia.

	Baseline	MED	MED + V	*P*
Sedation score at T-1	1 ± 0.3	6.5 (6, 7)[a]	6.5 (5, 7)[a]	0.414
Induction score	–	1 (1, 1)	2 ± (1, 3)	0.063
Induction time (s)	–	83 ± 11	63 ± 10	0.008
Time of extubation (min)	–	20 ± 7	25 ± 4	0.305
Time of sternal (min)	–	34 ± 8	34 ± 7	0.973
Time of standing (min)	–	39 ± 5	39 ± 15	0.876
Total amount of dobutamine (μg/kg)	–	27 ± 16	67 ± 25	0.018
Total fecal output (kg)	23 ± 4	13 ± 4[a]	11 ± 3[a]	0.423
Number of piles of feces	14 ± 2	9 ± 2[a]	10 ± 2[a]	0.542

[a] Significant difference ($P < 0.05$) within treatment with respect to baseline. *P*-values refer to statistical difference between the treatments.

Discussion

In this study, adding vatinoxan to the premedication regimen for isoflurane anesthesia balanced with medetomidine CRI resulted in significant hypotension during anesthesia in healthy horses. Hypotension responded to dobutamine treatment and, consequently, tissue oxygen delivery was improved compared to MED alone. Isoflurane has hypotensive effects in horses (Grosenbaugh and Muir, 1998). Furthermore, dorsal recumbency can exacerbate hypotension by decreasing venous return, as suggested by our finding of decreased CVP. Medetomidine counteracts these effects by causing vasoconstriction, thus maintaining higher MAP and SVR during isoflurane anesthesia (Ringer et al., 2007; Risberg et al., 2016). Vatinoxan apparently antagonized the vasoconstriction, which consequently led to hypotension, as suggested in earlier reports (Pakkanen et al., 2015; Wittenberg-Voges et al., 2018) and also by our finding that SVRI tended to be higher with MED. Despite the dose of vatinoxan used in the present study being smaller (140 μg/kg) than the ones used in earlier equine reports (200–250 μg/kg; Pakkanen et al., 2015; Wittenberg-Voges et al., 2018) hypotension was still observed. Conversely, in standing horses vatinoxan alone did not induce changes in arterial blood pressure (de Vries et al., 2016). The effect of vatinoxan on the cardiovascular changes induced by dexmedetomidine was dose-dependent in dogs (Honkavaara et al., 2011). Thus, further dose-response studies of vatinoxan are warranted to limit its hypotensive effect in anesthetized horses, especially when vatinoxan is combined with medetomidine that suppresses cardiovascular function in horses to a lesser extent than other α2-agonists (Bettschart-Wolfensberger et al., 1999; Yamashita et al., 2000).

Significantly more dobutamine was needed to maintain normotension with MED + V than with MED. As a positive inotropic agent, dobutamine can confound the interpretation of the cardiovascular observations. Indeed, we observed improved cardiac performance with MED + V relative to MED, but it is not possible to differentiate the effects of vatinoxan and dobutamine. The study design is limited by lack of cardiorespiratory measurements during the anesthesia prior to administration of

Table 3
Mean (± standard deviation) heart rate, carotid and pulmonary arterial pressures of six horses before (baseline) and during general anesthesia maintained with isoflurane and medetomidine CRI 3.5 μg/kg/h from 10 min after induction (T10) to the end of general anesthesia at 70 min (T70). The horses were administered as premedication medetomidine 7 μg/kg alone (MED) or with vatinoxan 140 μg/kg (MED + V) 10 min before induction with midazolam 0.06 mg/kg and ketamine 2.2 mg/kg. Baseline values were obtained prior to general anesthesia as a mean of three separate measurements in the same standing unmedicated horses.

Variable	Baseline	Treatment	T10	T20	T30	T40	T50	T60	T70
HR (beats/min)	37 ± 4.5	MED	34 ± 3.6	29 ± 3.9[b]	31 ± 4.8	31 ± 3.7[b]	32 ± 5.0	30 ± 4.5[b]	31 ± 5.3
		MED + V	42 ± 13.4[a,b]	33 ± 4.6	35 ± 7.9	34 ± 4.4	35 ± 6.3	33 ± 5.0	36 ± 5.4
SAP (mmHg)	152 ± 12.2	MED	102 ± 21.9[b]	102 ± 11.7[b]	95 ± 15.1[b]	98 ± 6.8[b]	95 ± 11.2[b]	106 ± 12.9[b]	92 ± 8.0[b]
		MED + V	76 ± 15.7[a,b]	112 ± 11.7[b]	123 ± 21.0[a,b]	118 ± 27.4[a,b]	111 ± 13.7[b]	120 ± 20.7[b]	111 ± 20.5[a,b]
MAP (mmHg)	123 ± 10.5	MED	77 ± 21.0[b]	80 ± 8.0[b]	68 ± 15.7[b]	74 ± 10.1[b]	71 ± 6.4[b]	82 ± 5.2[b]	75 ± 6.1[b]
		MED + V	52 ± 12.1[a,b]	83 ± 21.9[b]	76 ± 9.9[b]	80 ± 14.1[b]	71 ± 11.1[b]	79 ± 3.9[b]	73 ± 4.6[b]
DAP (mmHg)	101 ± 9.6	MED	68 ± 23.9[b]	72 ± 9.8[b]	67 ± 10.0[b]	74 ± 6.9[b]	60 ± 6.6[b]	73 ± 5.7[b]	64 ± 4.9[b]
		MED + V	41 ± 12.4[a,b]	66 ± 23.0[b]	60 ± 8.1[b]	59 ± 8.9[a,b]	58 ± 13.7[b]	62 ± 4.7[b]	62 ± 6.4[b]
SPAP (mmHg)	43 ± 5.9	MED	0 ± 5.6	6 ± 4.7	6 ± 5.1	8 ± 3.4	7 ± 6.1	9 ± 5.3	8 ± 5.3
		MED + V	11 ± 12.6	25 ± 25.2	20 ± 11.1[a]	18 ± 10.5[a]	17 ± 5.6	17 ± 10.6	14 ± 9.6
MPAP (mmHg)	24 ± 5.2	MED	−4 ± 6.1	0 ± 3.1	−1 ± 1.6	1 ± 3.7	−1 ± 3.9	1 ± 4.7	0 ± 4.8
		MED + V	3 ± 12.9	11 ± 26.0	3 ± 3.3	5 ± 5.5	1 ± 4.9	2 ± 4.2	2 ± 4.7
DPAP (mmHg)	7 ± 5.0	MED	−9 ± 6.6	−8 ± 5.7	−9 ± 3.1	−8 ± 5.1	−8 ± 6.3	−7 ± 6.4	−8 ± 5.9
		MED + V	−6 ± 16.2	3 ± 27.4	−6 ± 4.6	−3 ± 7.6	−8 ± 4.1	−7 ± 5.1	−7 ± 4.3

HR, heart rate; SAP, systolic arterial pressure; MAP, mean arterial pressure; DAP, diastolic arterial pressure; SPAP, systolic pulmonary arterial pressure; MPAP, mean pulmonary arterial pressure; DPAP, diastolic pulmonary arterial pressure.
[a] Significant differences ($P < 0.05$) between treatments (compared to baseline).
[b] Within treatment differences ($P < 0.05$) compared to baseline at a given time point.

Table 4
Mean (± standard deviation) cardiorespiratory variables and blood gas analysis results of six horses before (baseline) and during general anesthesia maintained with isoflurane and medetomidine CRI 3.5 μg/kg/h from 10 min after induction (T10) to the end of general anesthesia at 70 min (T70). The horses were administered as premedication medetomidine 7 μg/kg alone (MED) or with vatinoxan 140 μg/kg (MED + V) 10 min before induction with midazolam 0.06 mg/kg and ketamine 2.2 mg/kg. Baseline values were obtained prior to general anesthesia as a mean of three separate measurements in the same standing unmedicated horses.

Variable	Baseline	Treatment	T20	T40	T70
Cardiac index (mL/min/kg)	79 ± 19	MED	39 ± 2[b]	47 ± 10[b]	46 ± 7[b]
		MED + V	73 ± 18[a]	69 ± 15[a]	62 ± 26[b]
Hemoglobin (g/L)	136 ± 12	MED	128 ± 12	133 ± 15	129 ± 13
		MED + V	143 ± 14	153 ± 13[b]	147 ± 19
Mean central venous pressure (mmHg)	10 ± 3.4	MED	−9 ± 2.7[b]	−8 ± 3.5[b]	−8 ± 4.7[b]
		MED + V	−4 ± 9.6[b]	−7 ± 4.7[b]	−6 ± 7.7[b]
Left ventricular workload (kg•m/min)	71 ± 23	MED	21 ± 2[b]	24 ± 7[b]	23 ± 2[b]
		MED + V	42 ± 13[a,b]	38 ± 10[a,b]	31 ± 11[b]
Stroke volume index (mL/beat/kg)	2.1 ± 0.5	MED	1.4 ± 0.2[b]	1.5 ± 0.3[b]	1.6 ± 0.3[b]
		MED + V	2.4 ± 0.6	2.1 ± 0.3	1.8 ± 0.8
Systemic vascular resistance index (mmHg/mL/min/kg)	1.4 ± 0.4	MED	2.2 ± 0.3[b]	1.8 ± 0.3	1.8 ± 0.4
		MED + V	1.3 ± 0.6	1.3 ± 0.4	1.6 ± 0.9
Pulmonary vascular resistance index (mmHg/mL/min/kg)	0.2 ± 0.1	MED	0.2 ± 0.1	0.2 ± 0.1	0.2 ± 0.1
		MED + V	0.1 ± 0.1	0.1 ± 0.1	0.1 ± 0.04
Arterial oxygen content (mL/dL)	19 ± 1.6	MED	19 ± 1.6	19 ± 2.2	19 ± 1.8
		MED + V	21 ± 1.7	22 ± 2.0[a,b]	21 ± 2.8[b]
Mixed venous oxygen content (mL/dL)	15 ± 1.7	MED	13 ± 1.7	14 ± 2.5	13 ± 2.6
		MED + V	17 ± 1.7[a]	18 ± 2.9[a,b]	17 ± 3.6[a]
Arterial O_2 tension (mmHg)	106 ± 2.7	MED	287 ± 100[b]	278 ± 93[b]	250 ± 112[b]
		MED + V	284 ± 109[b]	283 ± 142[b]	260 ± 118[b]
Mixed venous O_2 tension (mmHg)	39 ± 3.0	MED	33 ± 3.1	35 ± 4.1	35 ± 6.4
		MED + V	42 ± 1.7[a]	47 ± 8.2[a,b]	46 ± 10[a,b]
Mixed venous blood O_2 saturation (%)	79 ± 0.0	MED	71 ± 0.1	74 ± 0.1	73 ± 0.1
		MED + V	83 ± 0.0[a]	85 ± 0.1[a]	84 ± 0.1[a]
Arterial CO_2 tension (mmHg)	40 ± 2.1	MED	43 ± 3.3	45 ± 4.7	47 ± 5.3
		MED + V	46 ± 3.7	49 ± 4.3	50 ± 5.5
Oxygen delivery index (mL/min/kg)	15.1 ± 4.3	MED	7.4 ± 1.2[b]	9.1 ± 2.7[b]	8.3 ± 1.4[b]
		MED + V	15.3 ± 4.8[a]	15.4 ± 4.1[a]	13.2 ± 7.0
Oxygen consumption index (mL/min/kg)	3.2 ± 0.7	MED	2.4 ± 0.1[b]	2.5 ± 0.3[b]	2.2 ± 0.2[b]
		MED + V	3.0 ± 0.9	2.4 ± 0.3[b]	2.1 ± 0.6[b]
Oxygen extraction ratio (%)	20 ± 3	MED	33 ± 5[b]	29 ± 6[b]	28 ± 6[b]
		MED + V	20 ± 2[a]	17 ± 6[a]	19 ± 8[a]
Venous admixture (%)	–	MED	14 ± 4.5	17 ± 4.8	18 ± 6.5
		MED + V	19 ± 6.9	22 ± 9.8	20 ± 10.9
Alveolar dead space ventilation (%)	–	MED	0.16 ± 0.03	0.16 ± 0.06	0.17 ± 0.05
		MED + V	0.17 ± 0.05	0.17 ± 0.03	0.20 ± 0.02

[a] Significant differences ($P < 0.05$) between treatments (change from baseline was compared between the treatments for all parameters other than mixed venous blood O_2 saturation, venous admixture and alveolar dead space ventilation which were compared by time points).
[b] Within treatment differences ($P < 0.05$) compared to baseline at a given time point.

dobutamine. Dobutamine increases MAP by increasing myocardial contractility, and thus, $\dot{Q}t$, although with low doses (0.5 μg/kg/min) increases in $\dot{Q}t$ have not been consistently detected in horses (Raisis et al., 2000; Loughran et al., 2017). Our observations of higher LVW and SVI with MED + V compared to MED likely reflect increased myocardial contractility, an effect of dobutamine.

A medetomidine CRI decreases CI and increases SVR in anesthetized horses (Ringer et al., 2007; Risberg et al., 2016),

which was observed in the present study, whereas with MED + V, SVRI and CI remained mainly at baseline levels. Vatinoxan is known to alleviate the peripheral cardiovascular effects of α2-agonists, mainly by antagonizing vasoconstriction, and consequently, a normal SVR is maintained and a reflex bradycardia is prevented (Bryant et al., 1998; Pagel et al., 1998). In the present study, the more stable SVRI and CI likely contributed to improved DO_2I and $O_2ER\%$ with MED + V compared to MED, suggesting improved tissue oxygen delivery with MED + V. The increase in $O_2ER\%$ likely reflects the increasing oxygen cleavage from blood by tissues in face of decreasing DO_2I. However, to confirm whether oxygen delivery is improved in specific tissues with vatinoxan, perfusion and local blood flow should be measured directly (Raisis et al., 2000; Wittenberg-Voges et al., 2018). It is noteworthy that our results (significant decreases in CI and DO_2I and increases in $O_2ER\%$ with MED) indicate that medetomidine CRI during general anesthesia can compromise tissue oxygen delivery and local perfusion, even though commonly observed cardiovascular parameters (i.e. MAP and HR) may remain clinically acceptable. This should be considered especially when anesthetizing horses with cardiovascular compromise.

Certain drugs, particularly α2-agonists and ketamine, have been shown to interact with LiDCO lithium sensors in vitro, potentially causing bias in $\dot{Q}t$ measurements (Ambrisko et al., 2013). However, the plasma concentrations of these drugs measured in the present study were lower than those causing significant alterations in sensor voltage in vitro, and therefore, significant biases in $\dot{Q}t$ measurements, and parameters calculated thereof, would be unlikely.

Hemoglobin and CaO_2 concentration increased significantly with MED + V at the same time point (T40). Concurrent increases in Hb and CaO_2 have been reported before with dobutamine in anesthetized horses and attributed to splenic contraction, thus increasing Hb concentrations and the oxygen-carrying capacity of blood (Loughran et al., 2017). The increases in mixed venous blood oxygenation parameters with MED + V may partly be related to increased Hb concentrations or, furthermore, to improved hemodynamics, as previously reported in horses receiving vatinoxan and other α2-agonists (Pakkanen et al., 2015; de Vries et al., 2016; Tapio et al., 2018). Based on the PaO_2 results, oxygenation of blood, however, was adequate with both treatments.

Fecal output decreased after general anesthesia in all horses, but no horses showed signs of gastrointestinal distress. Vatinoxan did not influence the decrease, despite reports of it improving gastrointestinal borborygmi in standing horses sedated with detomidine (Vainionpää et al., 2013; Tapio et al., 2018) or romifidine (de Vries et al., 2016). It is possible that the gastrointestinal suppression associated with general anesthesia lasted longer than the effects of a single dose of vatinoxan administered before the anesthesia.

No difference was detected between the treatments in sedation scores after premedication, nor in plasma drug concentrations of medetomidine, in contrast to previous reports with other α2-agonists (Vainionpää et al., 2013; Pakkanen et al., 2015; de Vries et al., 2016). The small number of sampling time points for the determination of plasma drug concentrations did not allow for further pharmacokinetic analysis. However, enhanced circulation with MED + V may have resulted in faster induction because the distribution of midazolam and ketamine to the brain was facilitated.

This study design reflects clinical practice, as balancing inhalation anesthesia with a medetomidine CRI has become popular in horses (Gozalo-Marcilla et al., 2017). It does not, however, allow comparisons with isoflurane anesthesia without any adjunctive CRI. Recently, further concerns about the effect of medetomidine CRI on cardiovascular function in horses have been reported (Risberg et al., 2016). Whether the protocols investigated in our study have advantages over sole isoflurane anesthesia with regard to cardiovascular function remains to be determined.

Conclusions

Combining vatinoxan with medetomidine for premedication resulted in hypotension that responded to dobutamine infusion, and cardiac performance and oxygen delivery were maintained. However, further research is warranted to limit the hypotensive effect of vatinoxan in anesthetized horses. Although heart rate and arterial blood pressures remained clinically acceptable with MED, cardiac performance and tissue oxygen delivery were lower than with MED + V.

Conflict of interest statement

Financial support was provided by Vetcare Ltd. (Mäntsälä, Finland) for the study expenses; materials and drugs. Vetcare Ltd. did not have any involvement in the study design, data analysis and interpretation, or writing and publication of the manuscript. None of the authors has any other financial or personal relationships that could inappropriately influence or bias the content of the paper.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.tvjl.2019.105345.

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