Effects of different sedative drug combinations on echocardiographic, haematologic and biochemical parameters in cats

INAUGURAL – DISSERTATION
zur Erlangung des Grades
eines Doktors der Veterinärmedizin
- Doctor medicinae veterinariae -
( Dr. med. vet. )

vorgelegt von
Kirsten Biermann
München

Hannover 2009
Wissenschaftliche Betreuung: Prof. Dr. S. Kästner
Klinik für Kleintiere

1. Gutachter: Prof. Dr. S. Kästner
2. Gutachter: Prof. Dr. M. Fehr

Tag der mündlichen Prüfung: 05.11.2009

Supported by Pfizer GmbH Germany, Division Animal Health
Meinen Eltern
This study has been published in part:

3.3.6 Treatments ................................................................. ................................................. 42
3.3.7 Recovery .................................................................................................................. 42
3.3.8 Statistical analysis .................................................................................................. 43
3.4 Results ................................................................................................................................ 43
3.4.1 Echocardiographic results .............................................................................................. 43
3.4.2 Laboratory results ........................................................................................................ 45
3.4.3 Sedation ........................................................................................................................ 45
3.4.4 Recovery ........................................................................................................................ 46
3.5 Discussion ................................................................................................................................ 46
3.6 Acknowledgements .............................................................................................................. 51
3.7 References .......................................................................................................................... 52
3.8 Tables and Figures .............................................................................................................. 57
4 General Discussion ............................................................................................................... 68
4.1 Materials and Methods ...................................................................................................... 68
4.2 Results .................................................................................................................................. 69
4.3 Conclusion and outlook ....................................................................................................... 73
5 Zusammenfassung .................................................................................................................. 75
6 Summary ............................................................................................................................... 78
7 Literaturverzeichnis ................................................................................................................. 81
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Meaning</th>
</tr>
</thead>
<tbody>
<tr>
<td>2D</td>
<td>two dimensional</td>
</tr>
<tr>
<td>A</td>
<td>left ventricular area</td>
</tr>
<tr>
<td>agree</td>
<td>agreement</td>
</tr>
<tr>
<td>ALM</td>
<td>area length method</td>
</tr>
<tr>
<td>ALT</td>
<td>alanine-aminotransferase</td>
</tr>
<tr>
<td>ANOVA</td>
<td>analysis of variance</td>
</tr>
<tr>
<td>AO</td>
<td>aortic root</td>
</tr>
<tr>
<td>AST</td>
<td>aspartate aminotransferase</td>
</tr>
<tr>
<td>AUC</td>
<td>area under the curve</td>
</tr>
<tr>
<td>AV</td>
<td>atrioventricular</td>
</tr>
<tr>
<td>B</td>
<td>baseline</td>
</tr>
<tr>
<td>bpm</td>
<td>beats per minute</td>
</tr>
<tr>
<td>CBC</td>
<td>complete blood count</td>
</tr>
<tr>
<td>cm</td>
<td>centimetre</td>
</tr>
<tr>
<td>CV</td>
<td>coefficient of variation</td>
</tr>
<tr>
<td>dl</td>
<td>decilitre</td>
</tr>
<tr>
<td>ECG</td>
<td>electrocardiogram</td>
</tr>
<tr>
<td>EDD</td>
<td>enddiastolic diameter</td>
</tr>
<tr>
<td>EDV</td>
<td>enddiastolic volume</td>
</tr>
<tr>
<td>EF</td>
<td>ejection fraction</td>
</tr>
<tr>
<td>ESD</td>
<td>enddiastolic diameter</td>
</tr>
<tr>
<td>ESV</td>
<td>endsystolic volume</td>
</tr>
<tr>
<td>ET</td>
<td>ejection time</td>
</tr>
<tr>
<td>FS</td>
<td>fractional shortening</td>
</tr>
<tr>
<td>g</td>
<td>gram</td>
</tr>
<tr>
<td>GLDH</td>
<td>glutamate dehydrogenase</td>
</tr>
<tr>
<td>Hb</td>
<td>haemoglobin</td>
</tr>
<tr>
<td>HCO3</td>
<td>hydrogene carbonate</td>
</tr>
<tr>
<td>HR</td>
<td>heart rate</td>
</tr>
<tr>
<td>IM</td>
<td>intramuscularly</td>
</tr>
<tr>
<td>KD</td>
<td>Ketamine - Dexmedetomidine</td>
</tr>
<tr>
<td>kg</td>
<td>kilogram</td>
</tr>
<tr>
<td>L</td>
<td>left ventricular length</td>
</tr>
<tr>
<td>LA</td>
<td>left atrium</td>
</tr>
<tr>
<td>LA:AO</td>
<td>left atrium to aorta</td>
</tr>
<tr>
<td>LV</td>
<td>left ventricular</td>
</tr>
</tbody>
</table>
List of abbreviations

LVWd  left ventricular freewall thickness in diastole
LVWs  left ventricular freewall thickness in systole
m  metre
MB  Midazolam – Butorphanol
MBK  Midazolam – Butorphanol – Ketamine
MBK  Midazolam – Butorphanol – Dexmedetomidine
MCH  mean corpuscular haemoglobin
MCHC  mean corpuscular haemoglobin concentration
MCV  mean corpuscular volume
meth  method
MHz  mega hertz
min  minute
ml  millilitre
mmol  millimol
na  not applicable
pCO2  partial pressure of carbon dioxide
PCV  packed cell volume
PLT  platelet count
pO2  partial pressure of oxygen
PT  prothrombin time
PTT  partial thromboplastin time
Qt  cardiac output
RBC  red blood cell count
SAP  systolic arterial blood pressure
SD  standard deviation
sec  second
SED  interventricular septal thickness in diastole
SES  interventricular septal thickness in systole
SM  Simpson’s method
SV  stroke volume
T  after treatment
U  units
V  left ventricular volume
VelAo  peak velocity of the aortic flow
VelPV  peak velocity of the pulmonic flow
VTI  velocity time integral
WBC  white blood cell count
1 Introduction

Handling aggressive cats in veterinary practice can be frustrating, time consuming, and injurious for both employee and animal. Using heavy restraint or punitive measures are not advisable for many reasons, including the possibility of injury to the patient, to the staff, accelerated aggression on subsequent visits, ethical obligations, and legal liability of abuse. If certain diagnostic procedures e.g. echocardiography or blood sampling are indicated in an uncooperative cat chemical restraint is required (MOFFAT 2008). In this situation drugs or drug combinations used for sedation should fulfill the following criteria: They should be active after intramuscular (IM) administration with a rapid onset of action, should have minimal cardiovascular and haematologic effects to get representative results of diagnostic procedures and should guarantee a smooth recovery of the animal.

Midazolam is a water-soluble benzodiazepine with a rapid onset of action after IM injection. Administered alone, it causes cats to become ataxic and some to behave dysphoric making approach and restraint more difficult (ILKIW et al. 1996). In combination with ketamine midazolam induces only limited cardiopulmonary depression (AKKERDAAS et al. 2001).

Butorphanol is a synthetic morphine derivative with agonistic effects at the κ-opioid receptors and partial agonist and antagonist activity at the µ-opioid receptors (COMMISKEY et al. 2005). In cats, butorphanol provides good visceral analgesia and leads to a mild sedation when administered alone (ANSAH et al. 2002). It has only minimal effects on cardiovascular parameters.

Ketamine is a dissociative anaesthetic containing two optical isomers: S-ketamine and R-ketamine. The drug is rapidly absorbed after intramuscular administration (HANNA et al. 1988) with a rapid onset of action. As a result of sympathetic stimulation it often produces significant increases in heart rate, cardiac output and blood pressure in healthy cats (CHILD et al. 1972). However, due to its direct negative inotropic effect on the myocardium cardiovascular effects may vary depending upon the patient’s condition (CLANACHAN et al. 1976).

Dexmedetomidine, an alpha₂-adrenoceptor agonist (α₂-agonist), is the active enantiomer of racemic medetomidine (VIRTANEN et al. 1988). Dexmedetomidine
induces a dose-dependant sedation, analgesia and muscle relaxation in cats (ANSAH et al. 1998) with marked decreases in heart rate, cardiac output, and transient mild changes in blood pressure (SELMI et al. 2003).

Anaesthetics and sedatives influence stroke volume (SV) and cardiac output (Qt) dose- and drug-dependently, most of them leading to a decrease in both parameters. If echocardiography is performed in a sedated cat, the observer should be aware of influences on the heart by used drugs.

Thermodilution techniques requiring pulmonary artery catheterization are regarded as the gold standard for measuring Qt and are widely used in clinical diagnosis of circulatory diseases in human medicine (SAKKA et al. 1999). For routine examinations in veterinary medicine this method is more invasive, because placement of pulmonary catheter requires sedation or anaesthesia in most species, for example in cats.

Transthoracic echocardiography, as a non-invasive method, represents another feasible technique for measuring Qt. Several ultrasound methods have been described which can be used to evaluate this parameter, such as the Teichholz method (geometric method) (TEICHHOLZ et al. 1976), the Simpson’s method (planimetric method), the area length method (planimetric method) (SERRES et al. 2008) and the Trace method (volumetric flow across the aorta) (UEHARA et al. 1995).

The first aim of this study was to test different echocardiographic approaches to calculate cardiac output using Teichholz method, Simpson’s method, area length method and the Trace method and to evaluate the repeatability of these methods in healthy cats. Using the ultrasound method with the best repeatability in cats, the second aim of this study was to compare the effects of midazolam and butorphanol, either with ketamine or dexmedetomidine and ketamine combined with dexmedetomidine alone on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats.
Comparison of cardiac output measurement using four different ultrasound techniques in healthy cats

Kirsten Biermann, Stephan Hungerbühler, Sabine B R Kästner

Small Animal Clinic, University of Veterinary Medicine Hanover, Bischofsholer Damm 15, 30173 Hanover, Germany

Correspondence: Kirsten Biermann, Small Animal Clinic, University of Veterinary Medicine Hanover, Bischofsholer Damm 15, 30173 Hanover, Germany

Email: kirsten.biermann@tiho-hannover.de
2.1 Abstract

Background: Thermodilution method for cardiac output (Qt) measurement in cats is too complex for routine examinations. The geometric method (based on Teichholz formula), two planimetric methods (Simpson’s and area length method) and the volumetric flow method across aorta (Trace method) represent non invasive ultrasound methods for assessment of Qt developed for humans.

Objectives: To assess agreement of four ultrasound methods for measuring stroke volume (SV) and cardiac output (Qt) and to evaluate their repeatability in healthy cats.

Animals: Six healthy cats, adult, weighing 4.29 kg ± 1.00 kg (mean ± SD) [3.00 to 6.00 kg] and aged 10.3 ± 4.3 years.

Methods: Prospective, experimental study. Measurement of SV and Qt was performed by a geometric method (based on Teichholz formula), two planimetric methods (Simpson’s derived and area length method) and a volumetric flow method across aorta (Trace method) in healthy cats. The coefficient of variation (CV) was calculated to determine between-day intra-observer repeatability of the four different ultrasound methods and Bland-Altman analysis was used to assess agreement between methods.

Results: Each of the six cats was scanned on four different occasions (n = 24). The CV was acceptable (CV < 20%) for all parameters, except SV and Qt obtained by using the Simpson’s method (28.8%; 22.4%) and area length method (21.6%; 22.6%). Values of SV and Qt obtained with the Trace method were significantly higher compared to all other methods. Both planimetric methods resulted in the lowest values for SV and Qt. Tight limits of agreement were observed between both planimetric methods regarding all parameters. Comparison between planimetric methods and both other methods resulted in wider limits of agreement.

Conclusion and Clinical Importance: Cardiac output can be calculated by a geometric method based on Teichholz formula with good repeatability and obtaining Qt values comparable to previous studies in which Qt measurement was performed by using thermodilution. Both planimetric methods were not well repeatable and
seemed to underestimate SV and CO, whereas the Trace method appeared to overestimate these parameters.

Key words: Feline; Echocardiography; Cardiac output
2.2 Introduction

Measurement of cardiac output (Qt) is an important part of cardiovascular monitoring as it is the best indicator to evaluate cardiovascular function due to its determination of oxygen delivery. Furthermore, it has a very important role in understanding the pathological features of heart diseases and forms the basis for cardiovascular treatment protocols (PINSKY 2001).

Thermodilution techniques requiring pulmonary artery catheterization are regarded as the gold standard for measuring Qt and are widely used in clinical diagnosis of circulatory diseases in human medicine (SAKKA et al. 1999). For routine examinations in veterinary medicine this method is more invasive, because placement of pulmonary catheter requires sedation and anaesthesia in most species, for example in cats.

Transthoracic echocardiography, as a non-invasive method, represents another feasible technique for measuring Qt. There are several ultrasound methods existing which can be used to evaluate this parameter.

The Teichholz method, which is a geometric method, is commonly used to calculate ventricular volumes from M-mode measurements (from which Qt can be calculated). As this method does not involve direct measurement of longitudinal left ventricular (LV) dimension it may lead to either overestimation or underestimation of LV volume. Nevertheless, the Teichholz method produces the most accurate volume estimates of any of the M-mode formulas tested in humans (KRONIK et al. 1979).

The Simpson’s method (SM) and the area length method (ALM) represent two planimetric methods that use formulas to determine the ventricular volume at end diastole and end systole. These two-dimensional equations are based on the fact that the left ventricle forms an ellipse when normal. In human medicine the Simpson’s method is the most commonly used formula for echocardiographic quantification of the left ventricle as it is recommended by the European Society of Cardiology (LANG et al. 2006). It appears relatively unaffected by changes in ventricular geometry as the volume of the left ventricle is computed as the sum of individual discs. In contrast, the area length method, a simpler technique, is used if
only one plane is obtained. Both planimetric methods have already been investigated in dogs (SERRES et al. 2008) and rats (STEIN et al. 2007). Another method to calculate SV and Qt is performed by using pulsed-wave Doppler to measure the volumetric flow across the aorta (Trace method). The velocity time integral (VTI) is measured and multiplied by the cross-sectional area of the aorta which results in SV. UEHARA et al. (1995) compared Qt measurement in dogs using the Trace method and thermodilution method amongst others and obtained a high correlation ($r = 0.93$) between both methods.

To our knowledge there are no available data existing comparing the use of the different ultrasound methods for SV and Qt measurement in cats. Therefore, the aim of this study was to test different echocardiographic approaches to calculate cardiac output using Teichholz method, Simpson’s method, area length method and the Trace method and to evaluate the repeatability of these methods in healthy cats.

2.3 Materials and Methods

2.3.1 Animals

Six adult domestic short-hair cats, three neutered males and three females (one neutered), weighing 4.29 kg ± 1.00 kg (mean ± SD) [3.00 to 6.00 kg] and aged 10.3 ± 4.3 years were used. Health status was assessed by means of physical examination, echocardiographic examination, a complete blood count and serum biochemical analyses. All findings were within reference range.

The experiments were performed with the approval of the Ethical Committee of the Lower Saxony State Office for Consumer Protection and Food Safety (33.9-42502-04-08/1471).

2.3.2 Experimental design

The study was carried out as a prospective experimental trial. Cats were studied on four different days by one experienced observer to evaluate between-day intra-
observer repeatability for each echocardiographic parameter. Food but not water was withheld for 8 hours prior to the experiment as cats were sedated for another study.

2.3.3 Echocardiography

Hair was clipped on the right and left lateral thoracic wall for echocardiography. Afterwards cats were kept undisturbed in a dimly lit and quiet cage to calm down for at least 10 minutes. Transthoracic echocardiographic examinations were performed by a single operator using Vivid 7 pro BT 03 (G.E. Healthcare, Brondby, Denmark) and a 4 - 8 MHz sector transducer (G.E. Healthcare, Brondby, Denmark). Echocardiographic images were stored on the internal hard drive of the echocardiograph with a simultaneous lead II electrocardiogram (ECG) and analysed off-line using the measurement software (System Software 1.5.2; Application Software 3.6.6) in the echocardiographic recorder. All examinations were performed in accordance with the recommendations of the Echocardiography Committee of the Specialty of Cardiology, American College of Internal Medicine (THOMAS et al. 1993).

End diastole frames were considered as the frame just before the mitral valves close or the first frame that shows the QRS complex, and end systole frames as the frame just before the mitral valve opens or end of T wave. Heart rate (HR) was calculated from the preceding R-R interval on the ECG. A mean of three consecutive measurements was used for each parameter. Stroke volume and cardiac output were obtained using the following different techniques:

2.3.3.1 Teichholz method (geometric method)

To apply the Teichholz method, the following measurements were needed and obtained from two-dimensional guided M-mode on the right parasternal short axis view at the level of the papillary muscles: Left-ventricular enddiastolic (EDD) and endsystolic (ESD) diameters, interventricular septal thickness in diastole (SED) and systole (SES), left ventricular free wall thickness in systole (LVWs) and diastole (LVWd). From these measurements the enddiastolic (EDV) and endsystolic (ESV)
volume were calculated using the geometric Teichholz method (TEICHHOLZ et al. 1976):
\[
EDV = \left[\frac{7 \times (EDD)^3}{2.4 + EDD}\right]
\]
\[
ESV = \left[\frac{7 \times (ESD)^3}{2.4 + ESD}\right]
\]
Ejection Fraction (EF) was calculated by using the following formula:
\[
EF = 100 \times \frac{EDV-ESV}{EDV}
\]
Stroke volume (SV) and cardiac output (Qt) were calculated by using following formulas:
\[
SV = EDV - ESV
\]
\[
Qt = SV \times HR
\]

2.3.3.2 Simpson’s method (SM) and area length method (ALM) (planimetric methods)
The volume of the left ventricle in diastole and systole, SV and Qt were also measured from the left apical 4-chamber view using two planimetric methods: SM and ALM. Briefly, the left apical 4-chamber view was maximized in length and width and digitally stored to perform analysis off-line. After tracing along the endocardial border of the left ventricular chamber, following the mitral annulus at the heart base, computerized calculations divided the length measured from the mitral annulus to the apex of the chamber into a stack of discs. For each disc a volume was calculated and summated for the total left ventricular volume in either diastole (EDV) or systole (ESV). Subsequently, EF was calculated.
The area length method relies on the following formula:
\[
V = 0.85 \frac{A^2}{L},
\]
where V is the left ventricular volume, A is the left ventricular area and L is the left ventricular length.
For applying this equation the same end-diastolic and end-systolic left ventricular length and areas obtained from the left apical 4-chamber view were used as for the Simpson’s method.
2.3.3.3 Volumetric flow method across aorta (Trace method)

Stroke volume and cardiac output were also measured using the velocity time integral (VTI) across the aortic valves obtained by pulsed-wave Doppler from the right parasternal transverse view. Therefore, peak velocities across the aorta (Vel\textsubscript{Ao}) were stored and the VTI was evaluated by manually tracing the flow profile with the trackball and calculating the area under the curve of the peak flow velocity versus time. Stroke volume was calculated by multiplying VTI with the area of the aorta. The method was previously described by BROWN (1991) and UEHARA (1995).

2.3.4 Statistical analysis

Data are presented as mean ± SD. The same experienced echocardiographer scanned each of the six cats on four different occasions (n = 24). The mean of three measurements was recorded for each parameter. The between-day intra-observer repeatability was calculated using a two-way analysis of variance (ANOVA). From the analysis of variance table the square root of the residual mean square was calculated, which is the mean standard deviation (SD), and from this and the mean, the coefficient of variation (CV) was calculated and quoted as a percentage. Clinical acceptability was defined as a CV less than or equal to 20%.

One-way analysis of variance (ANOVA) for repeated measures was used to analyse differences between methods, followed by a Student’s t-test with Bonferroni’s correction where appropriate.

Bland-Altman analysis was used to assess agreement of four different ultrasound techniques used to measure SV and Qt (BLAND u. ALTMAN 1999).

For all comparisons, p < 0.05 was considered significant.

2.4 Results

2.4.1 Between-day variability

All six cats were scanned on four occasions (n = 24). The CV values and corresponding SD values of between-day intra-observer repeatability obtained by the
different methods are given in table 1 and 2. A CV > 20 % was observed in SV and Qt obtained by SM (28.8%; 22.4%) and ALM (21.6%; 22.6%).

2.4.2 Echocardiographic measurements
The mean values and standard deviations of each parameter measured by the different techniques including Teichholz method, SM and ALM are presented in Table 3. Parameters including EDV, ESV and EF measured by Teichholz method were statistically significantly (p<0.05) higher compared to parameters obtained with SM and ALM.

Stroke volume and cardiac output gained with the Trace method across aorta were statistically significant (p<0.05) higher compared to all other methods. Both planimetric methods resulted in the lowest values for SV and Qt (Table 4).

Table 5 shows the mean values and standard deviations of parameters obtained by pulse-wave Doppler measurement across the aorta. All findings were in line with those reported elsewhere (DEMADRON et al. 1985; RIESEN et al. 2007)

2.4.2.1 Bland-Altman analysis: Comparison between Teichholz method, Simpson’s method, area length method and Trace method
For all parameters, both planimetric methods showed tight limits of agreement (Table 6). In contrast, regarding SV and Qt, comparison between both planimetric methods and both other methods revealed wider limits of agreement. In addition, the difference between the Teichholz method and all other methods increased as the mean Qt value increased, as the Bland-Altman analysis illustrates (Fig 1A-1D).

2.5 Discussion
This study was designed to compare different echocardiographic methods for SV and Qt measurement in healthy cats and to evaluate the intraobserver variability of these techniques.

Regarding the between-day intra-observer variability of parameters obtained with M-mode and pulsed-wave Doppler, the CV was less than 8.7, demonstrating a very
good repeatability. In accordance with our findings, SIMPSON et al. (2007) also reported highly repeatable measurements using M-mode and spectral Doppler in cats by a single investigator.

The between-day variability of LV volumes, EF, SV and Qt assessed by four different echocardiographic techniques (ie, Teichholz method, SM, ALM, Trace method) were acceptable (CV < 20%), except for SV and Qt measured with SM and ALM. In contrast to our results, SERRES et al. (2008) reported a lower variability (CV < 11%) for both planimetric methods when used in dogs. A reason for the high CV in the current study might be the small size of the feline heart, which can be responsible for suboptimal images (MOISE u. FOX 1999).

Regarding the absolute SV and Qt values, we observed significant differences between the four techniques. The Trace method obtained highest values, followed by Teichholz method and both planimetric methods gained lowest values. In the current study, we did not measure SV and Qt by thermodilution or dye dilution, which are considered as the gold standards to determine these parameters in human and veterinary medicine (SAKKA et al. 1999). However, there are doubts about the use of thermodilution as the best standard for Qt determinations in cats. In these animals, the thermodilution technique with placement of a pulmonary artery catheter is a relatively invasive and complex technique, as the catheter does not float into the heart and must be positioned and verified by fluoroscopy. In dogs, horses and humans, the balloon of the catheter allows easy placement by a blind flotation technique and correct placement can be assumed from the typical pressure tracings that occur if the pulmonary catheter reaches the pulmonary artery. KUTTER et al. (2007) investigated pulmonary artery and transpulmonary thermodilution cardiac output measurement in anaesthetized cats and compared these methods with the Fick method, which is considered the absolute gold standard and most accurate method to determine Qt (MATHEWS u. SINGH 2008). In that study, a better correlation was obtained between the Fick method and the transpulmonary thermodilution method compared to the pulmonary artery thermodilution method. They state that the injection of the ice-cold bolus can cause a transient and significant slowing of the heart rate, thus leading to a decrease in Qt values.
Nevertheless, in previous studies (ALLEN et al. 1986; DYSON et al. 1987; DYSON et al. 1988; INGWERSEN et al. 1988; LAMONT et al. 2001) SV and Qt measurements were performed by thermodilution method in awake cats with similar weights to our cats and SV and Qt values ranged from 3.31 ± 0.37 ml/beat and 0.67 ± 0.037 l/min (INGWERSEN et al. 1988) to 6.8 ± 0.565 ml/beat and 1.30 ± 0.09 l/min (LAMONT et al. 2001), respectively. In comparison to our results, values gained by the Teichholz method (SV: 5.01 ± 1.43 ml/beat; Qt: 1.02 ± 0.30 l/min) showed the best convergence, whereas values obtained by the Trace method were slightly higher and both planimetric methods underestimated SV and Qt. There might be different reasons for the significantly lower SV and Qt values gained by SM and ALM in our study:

Both planimetric methods require optimal images of apical views and myocardial dropout is always a potential problem (FEIGENBAUM et al. 2005). Furthermore, for accurate applications of these methods the maximization of the left ventricle using the left apical 4-chamber view is very important. This condition can only be guaranteed if the transducer is set at the true apex and if the ultrasonic beam goes through the centre of the left ventricle. Due to the small size of the feline heart and the high heart rate it was technically demanding for the observer to optimize the left ventricle in its length. Therefore, a reason for the low values might be the fact that described conditions were not always met.

Another reason might be the shape of the feline heart, which is more slender in diameter and with the apex more pointed in shape (OWENS u. BIERY 1999). Formulas of both planimetric methods were created to measure Qt in humans and were based on the assumption that the heart forms an ellipse. So both formulas might not be optimal for measurement SV and Qt in cats.

Lastly, quantification of LV function is performed by visual estimation when tracing the endocardial border of the left ventricle. Reports have claimed high accuracy in comparison with more objective methods, at least for trained observers (VAN ROYEN et al. 1996; GUDMUNDSSON et al. 2005). However, a recent meta-analysis suggested a wide variability in this subjective assessment (MCGOWAN u. CLELAND 2003).
The experimental design of this study has certain limitations. Firstly, only six cats were used. Secondly, we did not measure SV and Qt using thermodilution thus having no true comparison to a gold standard but only agreement of echocardiographic techniques.

In conclusion, according to our results, the use of the geometric method based on the Teichholz formula is a well repeatable method for the assessment of SV and Qt in healthy cats and obtained values comparable to previous studies which have performed Qt measurement by using thermodilution. Both planimetric methods should not be recommended for the assessment of LV volume in cats, because these methods were not well repeatable and seemed to underestimate the values for SV and Qt. The Trace method was best repeatable, but appeared to overestimate SV and Qt values.

Further investigations should be carried out to compare described ultrasound methods to thermodilution or direct Fick method and to test their use in cats with heart disease.

2.6 Acknowledgements

The authors would like to thank Pfizer GmbH Germany, Division Animal Health, for support of the study.
2.7 References


KRONIK, G., J. SLANY u. H. MOSSLACHER (1979):
Comparative value of eight M-mode echocardiographic formulas for determining left ventricular stroke volume. A correlative study with thermodilution and left ventricular single-plane cineangiography.
Circulation 60, 1308-1316.

Comparison of pulmonary artery and transpulmonary thermodilution cardiac output measurement in anaesthetized cats.
In: AVA/ECVAA Meeting Leipzig, 47.

Cardiopulmonary evaluation of the use of medetomidine hydrochloride in cats.

Recommendations for chamber quantification.

Cardiac output monitoring.
Ann Card Anaesth 11, 56-68.

Reliability of reporting left ventricular systolic function by echocardiography: a systematic review of 3 methods.
Am Heart J 146, 388-397.

MOISE, N. S. u. P. R. FOX (1999):
Echocardiography and Doppler imaging.
In: P. R. FOX, D. SISSON und N. S. MOISE
Textbook of Canine and Feline Cardiology, Principles and Clinical Practice

Radiographic Interpretation for the Small Animal Clinician.
second, Williams & Wilkins, Baltimore.

A rose by any other name: cardiac output.
Comparison of Doppler-derived aortic velocities obtained from various transducer sites in healthy dogs and cats.
Vet Radiol Ultrasound 48, 570-573.

Comparison of pulmonary artery and arterial thermodilution cardiac output in critically ill patients.

Comparison of 3 ultrasound methods for quantifying left ventricular systolic function: correlation with disease severity and prognostic value in dogs with mitral valve disease.

Assessment of the repeatability of feline echocardiography using conventional echocardiography and spectral pulse-wave Doppler tissue imaging techniques.
Vet Radiol Ultrasound 48, 58-68.

Effects of anesthesia on echocardiographic assessment of left ventricular structure and function in rats.
Basic Res Cardiol 102, 28-41.

TEICHHOLZ, L. E., T. KREULEN, M. V. HERMAN u. R. GORLIN (1976):
Problems in echocardiographic volume determinations: echocardiographic-angiographic correlations in the presence of absence of asynergy.
Am J Cardiol 37, 7-11.

Recommendations for standards in transthoracic two-dimensional echocardiography in the dog and cat. Echocardiography Committee of the Specialty of Cardiology, American College of Veterinary Internal Medicine.

Determination of cardiac output by echocardiography.
2.8 Tables and Figures

**Table 1:** Between-day intra-observer standard deviations (SD) and coefficients of variation (CV) for measurement of echocardiographic parameters in six healthy cats on four different days

<table>
<thead>
<tr>
<th>Independent Variable</th>
<th>Method of Measurement</th>
<th>SD</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDV (ml)</td>
<td>Teichholz formula method</td>
<td>0.62</td>
<td>10.9</td>
</tr>
<tr>
<td></td>
<td>Simpson's derived method</td>
<td>0.42</td>
<td>13.7</td>
</tr>
<tr>
<td></td>
<td>area length method</td>
<td>0.44</td>
<td>13.6</td>
</tr>
<tr>
<td>ESV (ml)</td>
<td>Teichholz formula method</td>
<td>0.12</td>
<td>18.8</td>
</tr>
<tr>
<td></td>
<td>Simpson's derived method</td>
<td>0.14</td>
<td>16.5</td>
</tr>
<tr>
<td></td>
<td>area length method</td>
<td>0.15</td>
<td>17.3</td>
</tr>
<tr>
<td>EF (%)</td>
<td>Teichholz formula method</td>
<td>2.48</td>
<td>2.8</td>
</tr>
<tr>
<td></td>
<td>Simpson's derived method</td>
<td>7.48</td>
<td>10.5</td>
</tr>
<tr>
<td></td>
<td>area length method</td>
<td>7.35</td>
<td>10.2</td>
</tr>
<tr>
<td>SV (ml)</td>
<td>Teichholz formula method</td>
<td>0.64</td>
<td>12.8</td>
</tr>
<tr>
<td></td>
<td>Simpson's derived method</td>
<td>0.61</td>
<td><strong>28.9</strong></td>
</tr>
<tr>
<td></td>
<td>area length method</td>
<td>0.50</td>
<td><strong>21.6</strong></td>
</tr>
<tr>
<td></td>
<td>Trace method</td>
<td>0.57</td>
<td>8.7</td>
</tr>
<tr>
<td>Qt (ml)</td>
<td>Teichholz formula method</td>
<td>0.20</td>
<td>19.5</td>
</tr>
<tr>
<td></td>
<td>Simpson's derived method</td>
<td>0.10</td>
<td><strong>22.4</strong></td>
</tr>
<tr>
<td></td>
<td>area length method</td>
<td>0.11</td>
<td><strong>22.6</strong></td>
</tr>
<tr>
<td></td>
<td>Trace method</td>
<td>0.12</td>
<td>9.0</td>
</tr>
</tbody>
</table>

EDV: end-diastolic volume; ESV: end-systolic volume; EF: ejection fraction; SV: stroke volume; Qt: cardiac output
Table 2: Between-day intra-observer standard deviations (SD) and coefficients of variation (CV) for measurement of echocardiographic parameters in six healthy cats on four different days

<table>
<thead>
<tr>
<th>Parameter</th>
<th>SD</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDD (ml)</td>
<td>0.06</td>
<td>4.3</td>
</tr>
<tr>
<td>ESD (ml)</td>
<td>0.04</td>
<td>6.8</td>
</tr>
<tr>
<td>FS (%)</td>
<td>3.58</td>
<td>6.4</td>
</tr>
<tr>
<td>SED (cm)</td>
<td>0.04</td>
<td>8.7</td>
</tr>
<tr>
<td>SES (cm)</td>
<td>0.06</td>
<td>8.4</td>
</tr>
<tr>
<td>LVWd (cm)</td>
<td>0.03</td>
<td>8.1</td>
</tr>
<tr>
<td>LVWs (cm)</td>
<td>0.04</td>
<td>4.9</td>
</tr>
</tbody>
</table>
| Vel
| 0.03 | 3.3    |
| VTI
| 0.49 | 4.9    |
| ET
| 8.83 | 6.0    |
| Area
| 0.05 | 7.8    |

EDD: end-diastolic diameter; ESD: end-systolic diameter; FS: fractional shortening; SED: interventricular septal thickness in diastole; SES: interventricular septal thickness in systole; LVWd: left ventricular free wall thickness in diastole; LVWs: left ventricular free wall thickness in systole; Vel: peak velocity; VTI: velocity time integral; ET: ejection time; Ao: Aorta
Table 3: Mean ± SD values of four repeated measurements of echocardiographic parameters in six cats by one observer

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Teichholz method</th>
<th>Simpson's method</th>
<th>Area length method</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDV (ml)</td>
<td>5.66 ± 1.52 a</td>
<td>3.04 ± 0.86 b</td>
<td>3.22 ± 0.92 b</td>
</tr>
<tr>
<td>ESV (ml)</td>
<td>0.65 ± 0.30 a</td>
<td>0.84 ± 0.21 b</td>
<td>0.85 ± 0.23 b</td>
</tr>
<tr>
<td>EF (%)</td>
<td>88.31 ± 5.09 a</td>
<td>71.29 ± 8.22 b</td>
<td>72.24 ± 8.01 b</td>
</tr>
<tr>
<td>EDD (cm)</td>
<td>1.45 ± 0.14</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>ESD (cm)</td>
<td>0.64 ± 0.11</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>FS (%)</td>
<td>55.66 ± 7.13</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>SED (cm)</td>
<td>0.41 ± 0.06</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>SES (cm)</td>
<td>0.71 ± 0.09</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>LVWd (cm)</td>
<td>0.42 ± 0.05</td>
<td>na</td>
<td>na</td>
</tr>
<tr>
<td>LVWs (cm)</td>
<td>0.74 ± 0.11</td>
<td>na</td>
<td>na</td>
</tr>
</tbody>
</table>

HR: heart rate; EDV: end-diastolic volume; ESV: end-systolic volume; EF: ejection fraction; EDD: end-diastolic diameter; ESD: end-systolic diameter; FS: fractional shortening; SED: interventricular septal thickness in diastole; SES: interventricular septal thickness in systole; LVWd: left ventricular free wall thickness in diastole; LVWs: left ventricular free wall thickness in systole; na: not applicable

a, b groups with different superscripts in the same row differ significantly (p<0.05)
Table 4: Mean ± SD values of four repeated measurements of stroke volume and cardiac output obtained by four different techniques in six cats by one observer

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>SV (ml)</td>
<td>5.01 ± 1.43 a</td>
<td>2.13 ± 0.80 b</td>
<td>2.32 ± 0.85 b</td>
<td>6.54 ± 1.51 c</td>
</tr>
<tr>
<td></td>
<td>Qt (l min⁻¹)</td>
<td>1.02 ± 0.30 a</td>
<td>0.45 ± 0.17 b</td>
<td>0.49 ± 0.18 b</td>
<td>1.34 ± 0.33 c</td>
</tr>
</tbody>
</table>

SV: stroke volume; Qt: cardiac output; meth: method

abc groups with different superscripts in the same row differ significantly (p<0.05)
**Table 5:** Mean ± SD values of four repeated measurements of echocardiographic parameters in six cats by one observer

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Aorta</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Vel (m s(^{-1}))</td>
<td>1.05 ± 0.10</td>
<td></td>
<td></td>
</tr>
<tr>
<td>VTI</td>
<td>9.98 ± 1.57</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ET</td>
<td>148.08 ± 16.30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Area (m(^2))</td>
<td>0.66 ± 0.12</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Vel: peak velocity; VTI: velocity time integral; ET: ejection time
Figure 1: Bland-Altman analysis illustrating the difference between Qt values assessed by 2 methods against the average of these 2 values. The mean differences are indicated as horizontal continuous grey lines, and limits of agreement (95% CI) are indicated as horizontal dashed lines. (A) Comparison of Teichholz method and Simpson's method. (B) Comparison of Teichholz method and area length method. (C) Comparison of Simpson's method and area length method. (D) Comparison of Teichholz method and Trace method.
Table 6: Bias, lower and upper agreement analysed by Bland-Altman comparing four different ultrasound methods in cats. Parameters including left ventricular volumes, EF, SV and Qt are presented.

<table>
<thead>
<tr>
<th></th>
<th>Teichholz vs Simpson</th>
<th>Teichholz vs Area length</th>
<th>Simpson vs Area Length</th>
<th>Teichholz vs Trace</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDV Bias (ml)</td>
<td>2.62</td>
<td>2.45</td>
<td>-0.17</td>
<td>na</td>
</tr>
<tr>
<td>Lower limit of agree</td>
<td>-0.08</td>
<td>-0.29</td>
<td>-0.33</td>
<td>na</td>
</tr>
<tr>
<td>Upper limit of agree</td>
<td>5.32</td>
<td>5.18</td>
<td>-0.02</td>
<td>na</td>
</tr>
<tr>
<td>ESV Bias (ml)</td>
<td>-0.18</td>
<td>-0.20</td>
<td>-0.02</td>
<td>na</td>
</tr>
<tr>
<td>Lower limit of agree</td>
<td>-0.83</td>
<td>-0.87</td>
<td>-0.09</td>
<td>na</td>
</tr>
<tr>
<td>Upper limit of agree</td>
<td>0.46</td>
<td>0.46</td>
<td>0.05</td>
<td>na</td>
</tr>
<tr>
<td>EF Bias (%)</td>
<td>17.02</td>
<td>16.07</td>
<td>-0.95</td>
<td>na</td>
</tr>
<tr>
<td>Lower limit of agree</td>
<td>-1.04</td>
<td>-1.78</td>
<td>-3.18</td>
<td>na</td>
</tr>
<tr>
<td>Upper limit of agree</td>
<td>35.07</td>
<td>33.92</td>
<td>1.28</td>
<td>na</td>
</tr>
<tr>
<td>SV Bias (ml)</td>
<td>2.87</td>
<td>2.66</td>
<td>-0.21</td>
<td>-1.54</td>
</tr>
<tr>
<td>Lower limit of agree</td>
<td>0.24</td>
<td>0.07</td>
<td>-0.96</td>
<td>-4.89</td>
</tr>
<tr>
<td>Upper limit of agree</td>
<td>5.50</td>
<td>5.26</td>
<td>0.55</td>
<td>1.81</td>
</tr>
<tr>
<td>Qt Bias (l/min)</td>
<td>0.58</td>
<td>0.54</td>
<td>-0.04</td>
<td>-0.33</td>
</tr>
<tr>
<td>Lower limit of agree</td>
<td>0.01</td>
<td>-0.01</td>
<td>-0.15</td>
<td>-1.05</td>
</tr>
<tr>
<td>Upper limit of agree</td>
<td>1.14</td>
<td>1.08</td>
<td>0.07</td>
<td>0.39</td>
</tr>
</tbody>
</table>

EDV: enddiastolic volume; ESV: endsystolic volume; EF: ejection fraction; SV: stroke volume; Qt: cardiac output; agree: agreement; na: not applicable
3 Manuscript II

Sedative, cardiovascular, haematologic and biochemical effects of four different drug combinations administered intramuscularly in cats

Kirsten Biermann, Stephan Hungerbühler, Reinhard Mischke, Sabine B R Kästner

Small Animal Clinic, University of Veterinary Medicine Hanover, Bischofsholer Damm 15, 30173 Hanover, Germany

Correspondence: Kirsten Biermann, Small Animal Clinic, University of Veterinary Medicine Hanover, Bischofsholer Damm 15, 30173 Hanover, Germany

Email: kirsten.biermann@tiho-hannover.de
3.1 Abstract

Objective To compare the effects of four different combinations of butorphanol, midazolam, ketamine and dexmedetomidine on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats.

Study design Prospective, randomized experimental cross-over trial

Animals Six healthy cats, adult, weighing 4.29 kg ± 1.00 kg (mean ± SD) [3.00 to 6.00 kg] and aged 10.3 ± 4.3 years.

Methods Each cat received each of the following treatments intramuscularly (IM) in a randomised cross-over design with at least 8 days wash-out: midazolam 0.4 mg kg\(^{-1}\) with butorphanol 0.4 mg kg\(^{-1}\) (MB), combined with ketamine 3 mg kg\(^{-1}\) (MBK) or dexmedetomidine 5 µg kg\(^{-1}\) (MBD) or ketamine 3 mg kg\(^{-1}\) with dexmedetomidine 5 µg kg\(^{-1}\) (KD) alone. Sedation was scored on a scale from 0 = no sedation to 5 = lateral recumbency, no response, as the response to tactile and auditory stimulation and was recorded 2, 4, 6, 8 and 10 minutes after injection. Echocardiography, systolic arterial blood pressure (SAP) measurement and blood sampling were performed at baseline (B) and 10 minutes after treatment (T). Recovery was videotaped and quality was scored by the same anaesthetist, blinded to the protocol, using a numeric descriptive scale. Times to sternal position and standing were recorded. Data are presented as mean ± SD and were analysed by ANOVA for repeated measures and paired t-tests. Sedation score was calculated over 10 minutes by area under the curve (AUC). Alpha was set at 5%.

Results Baseline values did not differ among treatments. The lowest sedation score was obtained by MB (12.33 ± 5.13) and the highest by KD (39.00 ± 4.00). Quality of recovery was best with KD (0.67 ± 0.8) and worst with MB (7.33 ± 2.3). Evaluation of recovery times was only possible for MBD and KD. Times to sternal recumbency and standing were faster after treatment with KD (46 ± 8 min; 56 ± 16 min) compared to MBD (77 ± 33 min; 87 ± 36 min).

All protocols caused a decrease in SAP of 17%, 25%, 13%, 5% in MB, MBK, MBD and KD, respectively, which was statistically significant (p<0.05) with MBD. Heart rate statistically significantly (p<0.05) increased after sedation with MB (5%) and statistically significantly (p<0.05) decreased after MBD (44%) and KD (34%). All
treatments statistically significantly (p<0.05) (except MBK) decreased stroke volume of 24%, 21%, 24%, 36% in MB, MBK, MBD and KD, and cardiac output of 23%, 34%, 54%, 53%, respectively. All treatments but MB produced a statistically significant (p<0.05) decrease in packed cell volume of 20%, 31%, 29% in MBK, MBD and KD, respectively. There were minimal effects on electrolytes and serum chemistry. Glucose concentration was increased after treatment with MBD (31%) and KD (52%) and lactate concentration was statistically significantly (p<0.05) decreased after MBK (58%), MBD (72%) and KD (65%).

Conclusion and clinical relevance
Midazolam with butorphanol failed to produce adequate sedation in healthy cats and caused them to behave dysphoric and aggressive. Cardiovascular and haematological changes were only minimal with this combination. Protocol MBK led to an acceptable sedation and minimal cardiovascular changes. Both treatments with dexmedetomidine produced excellent sedation and recovery but induced more cardiovascular depression and haematologic changes.

Keywords Sedation, cats, echocardiography, midazolam, butorphanol, ketamine, dexmedetomidine
3.2 Introduction

In veterinary practice cats are generally more difficult to handle than dogs due to their species-specific behaviour. In case these animals behave uncooperative or even aggressive sedation is required for certain diagnostic procedures e.g. echocardiography or blood sampling (MOFFAT 2008). In order to get representative diagnostic results drugs should only have minimal echocardiographic and haematologic effects.

Midazolam is a water-soluble benzodiazepine with a rapid onset of action after IM injection. Administered alone, it causes cats to become ataxic and some to behave dysphoric making approach and restraint more difficult (ILKIW et al. 1996). In combination with ketamine midazolam induces only limited cardiopulmonary depression (AKKERDAAS et al. 2001).

Butorphanol is a synthetic morphine derivative with agonistic effects at the \( \kappa \)-opioid receptors and partial agonist and antagonist activity at the \( \mu \)-opioid receptors (COMMISKEY et al. 2005). In cats, butorphanol provides good visceral analgesia and leads to a mild sedation when administered alone (ANSAH et al. 2002). It has only minimal effects on cardiovascular parameters. Administered intravenously under isoflurane anaesthesia butorphanol in combination with midazolam leads to a slight decrease in heart rate, blood pressure and respiratory rate (GROSS et al. 1993).

Ketamine is a dissociative anaesthetic containing two optical isomers: S-ketamine and R-ketamine. The drug is rapidly absorbed after intramuscular administration (HANNA et al. 1988) with a rapid onset of action. As a result of sympathetic stimulation it often produces significant increases in heart rate, cardiac output and blood pressure in healthy cats (CHILD et al. 1972). However, due to its direct negative inotropic effect on the myocardium cardiovascular effects may vary depending upon the patients condition (CLANACHAN et al. 1976).

Dexmedetomidine, an alpha\(_{2}\)-adrenoceptor agonist (\( \alpha_2 \)-agonist), is the active enantiomer of racemic medetomidine (VIRTANEN et al. 1988). Dexmedetomidine induces a dose-dependant sedation, analgesia and muscle relaxation in cats (ANSAH et al. 1998) with marked decreases in heart rate, cardiac output, and transient mild changes in blood pressure (SELMI et al. 2003). Furthermore,
dexmedetomidine causes a decrease in respiratory rate and body temperature (GRANHOLM et al. 2006).

Many intramuscular drug combinations have already been investigated in cats previously but to our knowledge effects on sedation, echocardiographic, haematologic and biochemical parameters and recovery have not been evaluated. Therefore, the aim of this study was to compare the effects of midazolam and butorphanol, either with ketamine or dexmedetomidine and ketamine combined with dexmedetomidine alone on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats.

3.3 Materials and Methods

3.3.1 Animals

Six adult domestic short-hair cats, three neutered males and three females (one neutered), weighing 4.29 kg ± 1.00 kg (mean ± SD) [3.00 to 6.00 kg] and aged 10.3 ± 4.3 years were used. Animals were university-owned, group-housed and were given free access to water and provided with commercially available cat food (Feline Health Nutrition, Royal Canin Tiernahrung GmbH & Co. KG, Cologne, Germany). Health status was assessed by means of physical examination, echocardiographic examination, a complete blood count and serum biochemical analyses. All findings were within reference range.

The experiments were performed with the approval of the Ethical Committee of the Lower Saxony State Office for Consumer Protection and Food Safety (33.9-42502-04-08/1471).

3.3.2 Experimental design

The study was carried out as a blind and experimental trial. Each of the six cats received each treatment in a randomised cross-over design. A period of 8 days was allowed between treatments to avoid residual drug effects. Food but not water was
withheld for 8 hours prior to treatment. On the day of experiment each cat was weighed and clinically examined.

The chronological order of further experimental procedures is demonstrated in Figure 1. Performance of blood pressure measurement took round about 5 minutes, echocardiography 20 minutes and blood sampling required approximately 5 minutes.

3.3.3 Blood pressure measurement

Systolic arterial blood pressure (SAP) was measured by use of a Doppler ultrasonic flow detector (Ultrasonic Doppler Flow Detector, Model 811-B, Parks Medical Electronics, Inc., Aloha, Oregon, USA) placed over the palmar digital artery and an occluding cuff (40% of the leg circumference) placed just above the carpus attached to a sphygmomanometer. Five consecutive measurements were taken in order to calculate the mean.

3.3.4 Echocardiography

Hair was clipped on the right and left lateral thoracic wall for echocardiography. Afterwards cats were kept undisturbed in a dimly lit and quiet cage to calm down for at least 10 minutes.

Complete routine transthoracic echocardiographic examinations were performed by a single experienced operator using a Vivid 7 pro BT 03 (G.E. Healthcare, Brondby, Denmark) and a 4 - 8 MHz sector transducer (G.E. Healthcare, Brondby, Denmark). Echocardiographic images were stored on the internal hard drive of the echocardiograph with a simultaneous lead II electrocardiogram (ECG) and analysed off-line using the measurement software (System Software 1.5.2; Application Software 3.6.6) in the echocardiographic recorder. All echocardiographic examinations were performed in accordance with the recommendations of the Echocardiography Committee of the Specialty of Cardiology, American College of Veterinary Internal Medicine (THOMAS et al. 1993).

The following measurements were obtained from two-dimensional (2D) guided M-mode on the right parasternal short axis view at the level of the papillary muscles:
Left-ventricular enddiastolic (EDD) and endsystolic (ESD) diameters, interventricular septal thickness in diastole (SED) and systole (SES), left ventricular free wall thickness in systole (LVWs) and diastole (LVWd). From these measurements the enddiastolic (EDV) and endsystolic (ESV) volume were calculated using the Teichholz method (TEICHHOLZ et al. 1976):

$$\text{EDV} = \frac{7 \times (\text{EDD})^3}{(2.4 + \text{EDD})}$$

$$\text{ESV} = \frac{7 \times (\text{ESD})^3}{(2.4 + \text{ESD})}$$

Ejection Fraction (EF) and Fractional Shortening (FS) were calculated by using following formulas:

$$\text{FS} = \left(\frac{\text{EDD}-\text{ESD}}{\text{EDD}}\right) \times 100$$

$$\text{EF} = 100 \times \frac{\text{EDV}-\text{ESV}}{\text{EDV}}$$

The size of the left atrium (LA) and aortic root (AO) were taken by 2D images obtained from the right parasternal short axis view and the left atrium-to-aorta ratio was calculated (LA:AO) as previously described (HANSSON et al. 2002).

From the left apical four chamber view (aortic) and right short axis (pulmonic) Doppler studies were performed. Peak velocities of the aortic (VelAo) and pulmonic (VelPV) flow were measured using pulsed-wave Doppler. Heart rate (HR) was calculated from the preceding R-R interval on the ECG.

Stroke volume (SV) and cardiac output (Qt) were calculated by using following formulas:

$$\text{SV} = \text{EDV} - \text{ESV}$$

$$\text{Qt} = \text{SV} \times \text{HR}$$

A mean of three consecutive measurements was used for each parameter.

3.3.5 Blood sampling and analysis

Whole blood samples were collected from the jugular or femoral veins using a 20-gauge disposable needle into tubes containing different anticoagulants (EDTA, Lithium-heparinate, sodium fluoride, sodium citrate). Acid base balance and blood gases (pH, pO2, pCO2, HCO3), electrolytes (sodium, potassium, calcium, chloride) in lithium heparin blood and a complete blood count (CBC) in EDTA blood were measured immediately after sampling. Blood gases were corrected for body
temperature and were analysed in combination with electrolytes using the automate blood gas and electrolytes analyser Rapidlab 860 (Siemens Healthcare, Eschborn, Germany). Measurement of CBC was performed by using the Advia 120 hematology system (Siemens Healthcare, Eschborn, Germany) including the following parameters: red blood cell count (RBC), haemoglobin (hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), white blood cell count (WBC), percentage of neutrophils, lymphocytes, monocytes, eosinophils and basophils and platelet count (PLT). Visual platelet counts were performed in addition if the counter report indicated presence of a high number (at least double plus) platelet clumps.

Lithium heparinate, sodium fluoride and sodium citrate blood samples were centrifuged for 2 minutes at 10000 \( g \), the plasma samples frozen at -28°C and analysed in series at the end of the study.

Liver enzyme activity (ALT: alanine aminotransferase; AST: aspartate aminotransferase [method according to International Federation of Clinical Chemistry and Laboratory Medicine, IFCC], GLDH: glutamate dehydrogenase [optimised method of the German Society for Clinical Chemistry]), plasma total protein (Biuret method) were measured in lithium heparin plasma and plasma blood glucose (enzymatic colorimetric test, hexokinase method) and plasma lactate (enzymatic colorimetric test) in sodium fluoride plasma using an automatic analyser (Hitachi 912; Roche Diagnostics GmbH, Mannheim, Germany). A universal calibrator (calibrator for automated systems, Roche Diagnostics) was used as a standard and a human control plasma (Precinorm U, Roche Diagnostics) as quality control.

Blood coagulation tests were measured automatically by the coagulation analyzer Amax Destiny plus™ (Trinity Biotech, Lemgo, Germany) using the clotting technique (ball coagulometer). Prothrombin time (PT) was measured with the reagent Thromborel (Siemens Healthcare) following a modified test: 25 µl of diluted citrated plasma (1:20 dilution) was incubated with 25 µl fibrinogen for two minutes at 37°C and the coagulation was induced by addition of 25 µl Thromborel S (MISCHKE et al. 1996). Results of PT were expressed automatically in percent activity of a normal
feline pool plasma (n = 6; activity = 100 %), by using a calibration curve based on
different dilution of the feline pool plasma. Activated partial thromboplastin time (PTT)
was measured using APTT Reagent (Roche Diagnostics) according the
manufacturers’ recommendations.

3.3.6 Treatments
Treatments for sedation were as follows: 0.4 mg kg\textsuperscript{-1} midazolam (Midazolam;
Ratiopharm GmbH, Ulm, Germany) with 0.4 mg kg\textsuperscript{-1} butorphanol (Dolorex, Intervet
Deutschland GmbH, Unterschleissheim, Germany) (MB), combined with 3 mg kg\textsuperscript{-1}
ketamine (Ketamin 5%, Selectavet Dr. Otto Fischer GmbH, Weyarn-Holzolling,
Germany) (MBK) or 5 µg kg\textsuperscript{-1} dexmedetomidine (Dexdomitor; Pfizer GmbH
Tiergesundheit, Berlin, Germany) (MBD) or 3 mg kg\textsuperscript{-1} ketamine with 5 µg kg\textsuperscript{-1}
dexmedetomidine (KD) alone. Immediately before administration drugs were mixed
together in the same syringe and injected as one shot in the triceps brachii muscle of
the right or left forelimb.
Sedation was scored (Table 1) on a scale from 0 to 5 and recorded 2, 4, 6, 8 and 10
minutes after injection. Cats were stimulated tactile by touching the body and
clamping one hind limb with two fingers as well as auditory by clapping hands next to
the cats ears.

3.3.7 Recovery
At the end of all procedures cats were put into a cage and recovery was videotaped
by a digital camcorder (Model DCR-VX 2000E, Sony, Tokyo, Japan). The time of
treatment injection was taken as the reference start point for recording recovery
times to sternal position and to standing. Quality of recovery was assessed from the
videotapes by one anaesthetist, blinded to the protocol, and was scored using a
numerical rating scale (Table 2). Spontaneous observations of any adverse events
were recorded during the study.
3.3.8 Statistical analysis

Parametric data are presented as mean ± SD, non parametric data as median [min – max]. The Kolmogorov-Smirnov test was used to assess the distribution of the variables. Baseline and sedation values for echocardiographic, haematologic, biochemical and blood pressure data were compared by paired t-tests. One-way analysis of variance (ANOVA) for repeated measures was used to analyse differences between protocols. If differences were significant (p < 0.05), paired t-test was used for comparisons between protocols. For each cat and treatment, the area under the sedation score curve (AUC) over 10 minutes was calculated with the trapezoidal method. A paired t-test was used to compare AUC between treatments. Recovery times for MBD and KD were compared by use of Student t-tests. Score of recovery quality was analysed using ANOVA for repeated measures. Significance was considered to be p < 0.05.

3.4 Results

3.4.1 Echocardiographic results

For all echocardiographic variables, baseline values did not differ among treatments. All findings were within reference range (PIPERS et al. 1979; VOLLMAR 1991). Systolic arterial blood pressure statistically significantly (p<0.05) decreased from baseline after MBD (Table 3). No statistically significant (p<0.05) differences were observed between treatments.

There was a statistically significant (p<0.05) decrease in HR from baseline after MBD (44% decrease) and KD (34% decrease) and a statistically significant (p<0.05) increase after MB (5% increase) (Table 3). Heart rate was statistically significantly (p<0.05) lower after MBD and KD compared to MB and MBK (Table 3).

A statistically significant (p<0.05) decrease in ESD and ESV from baseline was observed after MB was injected and a statistically significant (p<0.05) increase after MBD and KD (Table 3). Endsystolic diameter (ESD) and volume (ESV) were
statistically significantly (p<0.05) higher after MBD and KD compared to MB and MBK except for ESV between MBK and KD (Table 3).

In comparison with baseline a statistically significant (p<0.05) decrease in FS and EF was seen after treatment with MBD and KD. Comparisons between treatments demonstrated a statistically significantly (p<0.05) lower FS and EF after MBD and KD were administered than after MB and MBK (Table 3).

Interventricular septal thickness in systole (SES) was statistically significantly (p<0.05) smaller after treatment with KD compared to baseline. After MBD treatment SES was statistically significantly (p<0.05) lower than after MB and MBK (Table 3).

There was a statistically significant (p<0.05) decrease in LVWs after MBD. Compared with other treatments LVWs was statistically significantly (p<0.05) lower after MBD (Table 3). A statistically significant (p<0.05) decrease in EDV from baseline was observed after MB, but there were no statistically significant (p<0.05) differences between treatments (Table 3).

Stroke volume was statistically significantly (p<0.05) decreased after MB (24% decrease), MBD (24% decrease) and KD (36% decrease) compared with baseline without statistically significant (p<0.05) differences between treatments (Table 3).

All treatments statistically significantly (p<0.05) decreased cardiac output. Treatment MBD led to a statistically significant (p<0.05) lower Qt compared to MB and MBK and Qt was statistically significantly (p<0.05) lower after KD compared to MB (Table 3).

Peak Velocity of flow across the pulmonic valves decreased statistically significantly (p<0.05) from baseline after injection of MBD and KD (Table 4). The Vel\_{PV} was statistically significantly (p<0.05) lower after MBD and KD compared to MB and MBK.

All treatments decreased statistically significantly (p<0.05) peak velocity of flow across the aortic valves. MBD and KD lead to a statistically significantly (p<0.05) lower Vel\_{Ao} compared with MB and MBK. The Vel\_{Ao} was statistically significantly (p<0.05) slower after MBK than after MB (Table 4).
3.4.2 Laboratory results

Treated cats had a statistically significantly (p<0.05) decreased RBC, PCV, haemoglobin and MCV (Table 5) after MBK, MBD and KD compared with baseline. Packed cell volume was statistically significantly (p<0.05) lower after MBD and KD were injected compared with MB. Haemoglobin was statistically significantly (p<0.05) lower after MBD compared to MB (Table 5). Analysis of thrombocytes revealed a statistically significant (p<0.05) decrease of PLT after treatment with KD compared with baseline (Table 5).

The white blood count statistically significantly (p<0.05) decreased from baseline after treatment with MBD (Table 5). In all protocols there were no statistically significant (p<0.05) differences in the differential haemogram between baseline and after treatment (data not shown).

Analysis of venous blood gases (Table 6) revealed a statistically significant (p<0.05) increase in HCO₃⁻ and temperature corrected pH after treatment with KD in comparison with baseline. Base excess decreased statistically significantly (p<0.05) from baseline after MBD and KD were injected, as well as pO₂ after KD.

Treatments had minimal effects on electrolytes and serum chemistry except glucose and lactate concentrations (Table 7 and 8). All treatments but MB led to a statistically significant (p<0.05) decrease in plasma lactate concentration without any statistically significant (p<0.05) differences between treatments. Plasma glucose concentration decreased after MB and MBK, and increased after MBD and KD. Glucose concentration changes were statistically significant (p<0.05) after MBK and KD.

Used treatments had minimal effect on liver enzymes and coagulation parameters (Table 8). There was a decrease in plasma total protein with all treatments, which was statistically significant (p<0.05) with MBD and KD (Table 8).

3.4.3 Sedation

The lowest sedation score was gained with MB (Figure 2), which was statistically significantly (p<0.05) lower to all other treatments. The highest sedation score was
reached with KD. Both treatments with dexmedetomidine (MBD, KD) had statistically significantly (p<0.05) higher sedation scores compared to MB and MBK (Figure 2). All cats were uneasy and unwilling to be restraint after MB and showed more defence movement than prior to treatment. Two cats behaved even aggressive and were hissing after MB was injected.

3.4.4 Recovery

Times to sternal recumbancy and standing were not measurable in all cats treated with MB and in two cats treated with MBK because animals were already able to stand after the second blood sampling (approximately 40 minutes after treatment) was performed. In four cats given MBK, time to sternal recumbancy was 38 ± 6 minutes (min.) and time to standing was 49 ± 19 min. Comparison was only made between treatments MBD and KD. Recovery times were faster for cats after KD than with MBD for both times, to sternal recumbancy (MBD: 77 ± 33; KD: 46 ± 8) and to standing (MBD: 87 ± 36; KD: 56 ± 16). The difference between MBD and KD was statistically significant (p<0.05) regarding the time to sternal recumbancy. Quality of recovery was best with KD (0.67 ± 0.8) and worst with MB (7.33 ± 2.3) (Figure 3). Four of six cats vomited after treatment with KD. No vomitus was observed with other treatments. One cat showed transient atrioventricular (AV) dissociation after MB and transient first-degree atrioventricular block after MBD.

3.5 Discussion

The aim of this study was to evaluate and assess the influence of four different intramuscular drug combinations on sedation, recovery and on echocardiographic, haematologic and biochemical parameters in healthy cats, without any attempts to control pre- and afterload conditions as we tried to mimic clinical situation. Midazolam combined with butorphanol failed to produce adequate sedation and caused cats to behave in a dysphoric or aggressive manner. ILKIW et al. (1996) reported similar effects on cat’s behaviour when administering midazolam.
intramuscularly alone. Butorphanol in a dose of 0.4 mg kg\(^{-1}\) may not be able to reduce the adverse behavioural effects of midazolam in healthy cats or might even cause central stimulation.

Due to ketamines depressant effects on the central nervous system, especially the antagonism of the N-methyl-D-aspartate receptor (Kohrs u. Durieux 1998), the combination of midazolam, butorphanol and ketamine led to a deeper sedation compared to MB.

However, deepest and longest sedation was achieved with treatments including dexmedetomidine. The \(\alpha_2\)-agonists are well known as potent sedatives (Murrell u. Hellebrekers 2005). Ketamine combined with dexmedetomidine led to a higher sedation score, meaning a faster and deeper sedation, compared with MBD. One may conclude from this that ketamine in a dose of 3 mg kg\(^{-1}\) has more sedative properties than midazolam combined with butorphanol, both in a dose of 0.4 mg kg\(^{-1}\). However, Selmi et al. (2003) evaluated sedative effects of dexmedetomidine (10 µg kg\(^{-1}\)) alone and in combination with butorphanol (0.2 mg kg\(^{-1}\)) and ketamine (5 mg kg\(^{-1}\)) in cats and reported similar results among treatments regarding time to lateral recumbency, duration of lateral recumbency and time to recovery from sedation. Recovery times were faster with MBK than with both treatments with dexmedetomidine. Especially in two cats after treatment with MBK recovery times were not measurable as both animals were already standing after the procedures (approximately 40 minutes after treatment injection).

Best quality of recovery was obtained with ketamine combined with dexmedetomidine. This combination caused cats to recover very smooth, well coordinated and with less activity in the cage. Other studies as well reported satisfactory recoveries after anaesthesia with ketamine in combination with \(\alpha_2\)-agonists in cats (Cullen u. Jones 1977). Due to sympathetic tone cats have a higher heart rate in hospital than at home and even higher under restraint during echocardiography (Abbott 2005). Heart rates during echocardiography obtained in this study were similar to rates recorded by others (Hamlin 1989; Abbott 2005). A significant increase in HR was observed after treatment with MB making echocardiography even more difficult (Santilli u. S. 47
BUSSADORI 1998). KANDA and HIKASA (2008a) observed a slight increased HR in cats after intramuscular midazolam (0.5 mg kg$^{-1}$) administration as well. The decreases in ESD, EDV and ESV after MB administration resulted in a significantly decreased stroke volume leading to a decrease in cardiac output (23% decrease). The decreased SV might be the consequence of an impaired left ventricular filling due to an increased HR. Nevertheless, cardiac output was influenced by MB to a lower extent compared to other treatments used in this study. Similar to our findings, KOJIMA et al. (1999) observed only small changes in blood pressure and cardiac output in dogs sedated with midazolam 0.1 mg kg$^{-1}$ and butorphanol 0.2 mg kg$^{-1}$ intramuscularly.

Although heart rate and stroke volume were not significantly decreased after MBK, this treatment produced a significant decrease in cardiac output (34% decrease), but not as severe as MBD (54% decrease) and KD (53% decrease). Both treatments with dexmedetomidine, MBD and KD, had more depressant effects on cardiovascular parameters in comparison to the other treatments. After their administration heart rate, fractional shortening, stroke volume and cardiac output were significantly decreased from baseline due to central and peripheral $\alpha_2$-adrenoceptor activation. As a result of a decrease in central sympathetic action and an indirect baroreceptor-mediated increase in vagal tone bradycardia occurs after dexmedetomidine administration (VIRTANEN 1989). In our study ESV significantly increased after MBD and KD. Reasons for this increase may be the reduced contractility of the heart on the one hand and the $\alpha_2$-agonist-mediated increase in afterload caused by vasoconstriction on the other hand. The higher ESV resulted in a significant decrease in stroke volume which in addition to the decreased HR led to a decreased cardiac output. MURELL et al. (2005) state that the precise mechanism of dexmedetomidine to decrease cardiac output is unknown but three mechanisms are considered: direct myocardial effect, decreased function in response to $\alpha_2$-agonist-mediated increase in afterload and myocardial hypoxia and dysfunction in response to coronary vasoconstriction. SCHMELING et al. (1991) and DEMORAIS and MUIR (1995) found that the decrease in cardiac output after medetomidine administration is not a direct depression of myocardial contractility but primarily caused by the decrease in heart
rate and increase in vascular resistance. Results of a study by FLACKE et al. (1992), in which dexmedetomidine was investigated in isolated dog hearts, support these findings, as no myocardial depressant effect was found. So far, in myocardium no $\alpha_2$-adrenoceptor evidence has been provided. The marked reduction of cardiac output (approximately 50% decrease) in our study is comparable to data obtained from ALLEN et al. (1986) who measured Qt by thermodilution after administration of a xylazine (1.0 mg kg$^{-1}$) - ketamine (10 mg kg$^{-1}$) combination in cats.

In the current study, all treatments decreased systolic arterial blood pressure. $\alpha_2$-agonists have a biphasic dose-related (KALLIO et al. 1989) effect on blood pressure. Firstly, stimulation of peripheral $\alpha_2$-agonist receptors leads to vasoconstriction which mediates a transitory increase in blood pressure (DOCHERTY u. MCGRATH 1980). Secondly, prolonged hypotension occurs due to a central hypotensive effect and a decrease in sympathetic nervous tone. Due to the facts that we used dexmedetomidine in a low dose of 5 $\mu$g kg$^{-1}$ and performed blood pressure measurement only approximately 30 minutes after treatment the initial hypertensive phase could not be detected and SAP was decreased after treatment.

Red blood cell count, Hb and PCV significantly decreased after treatment with MBK, MBD and KD. This phenomenon may be explained by pooling of the circulating blood cells in the spleen or other reservoirs due to the decreased sympathetic activity. However, WILSON et al. (2004) investigated the influence of four different anaesthetic protocols on splenic size in dogs and found out that the administration of medetomidine, ketamine and diazepam leads to a greater splenic volume compared with acepromazine and propofol but that there is a lack of correlation between the packed cell volume and splenic size. They suggested sequestration of red blood cells in nonsplenic sites. Another reason might be the shifting of fluid from extravascular compartment to intravascular compartment (WAGNER et al. 1991) in order to maintain normal cardiac output in the animal.

In this study, glucose concentration had a marked increase after treatment with MBD and KD. This well known effect is in accordance with the study from KANDA et al. (2008b) who investigated the effects of medetomidine and xylazine on plasma concentration of glucose in cats. Tested doses ranged from 20 - 320 $\mu$g kg$^{-1}$
medetomidine IM and 0.5 - 8 µg kg\(^{-1}\) xylazine IM and led to a remarkable dose-dependant hyperglycaemia. The responsible mechanism behind the phenomenon is the inhibition of insulin secretion through the binding of α\(_2\)-agonists on α\(_2\)β-receptors in the β cells of the pancreas (HILLAIRE-BUYS et al. 1985).

In our study plasma lactate concentrations at baseline were quiet high but still within the reference range (1.0-3.5 mmol l\(^{-1}\)). Catecholamine release and muscle activation due to stress during blood sampling may be reasons for higher lactate values. In the current study cats were quiet excited after echocardiography at baseline when the first blood sampling was performed. All treatments except MB led to a significant decrease in plasma lactate concentrations. Responsible reasons may be muscle relaxation, stress reduction and redistribution during sedation. LÖBERT (2003) investigated the influence of anaesthesia on plasma lactate and pyruvate levels in dogs and they observed an obvious decrease in plasma lactate during anaesthesia mainly in animals that were agitated at baseline. There are concerns about the adequate tissue perfusion after dexmedetomidine due to its depressant effect on cardiac output and increased vascular resistance. Similar to our findings, UILENREEF et al. (2008), who investigated changes in plasma lactate concentrations during dexmedetomidine continuous rate infusion (1-3 µg kg\(^{-1}\) hour\(^{-1}\)) in dogs, reported values under 2 mmol l\(^{-1}\). In our study, lactate concentrations even decreased after treatments indicating that there was no appearance of inadequate tissue oxygen supply. As plasma total protein concentrations decreased as well, another reason for lower lactate concentrations after treatment might be a dilutional effect.

Emesis was the most common adverse effect in our study but was only observed after sedation with KD. Previous studies have described same emetic effects after administration of a α\(_2\)-agonist in cats (GRANHOLM et al. 2006; BELDA et al. 2009). However, MBD did not cause cats to vomit in our study although opioids are considered to stimulate the chemoreceptor trigger zone for emesis in the medulla. LASCELLES et al. (2004) observed vomitus after IM administration of hydromorphon in a dose of 0.1 mg kg\(^{-1}\) but not in combination with butorphanol in a dose of 0.4 mg
kg⁻¹ or with butorphanol alone in cats. Butorphanol in a dose of 0.4 mg kg⁻¹ IM seems not to be emetic in cats.

One cat showed transient first degree atrioventricular block with MBD. Alpha₂-agonists are known to cause AV blockade secondary to the initial increase in blood pressure, and reflex (baroreceptors) increase in vagal tone. The same cat had transient atrioventricular dissociation after administration of MB. We cannot explain this side effect as midazolam and butorphanol are not known to cause arrhythmias, especially not in doses used in this study. Maybe this cat has an individual sensitivity to arrhythmia.

The experimental design of this study has some limitations. Pre- and afterload conditions alter the majority of the echocardiographic parameters assessed in this study. No attempts were made to keep loading conditions constant because we tried to mimic clinical situation. However, we used healthy cats in this study and baseline values were not different among the different protocols. Therefore, equal baseline loading conditions can be assumed.

In conclusion, this study showed that midazolam with butorphanol failed to produce adequate sedation in healthy cats and caused them to behave dysphoric and aggressive. Cardiovascular and haematological changes were only minimal with this combination. Treatment with MBK led to acceptable sedation and mild cardiovascular changes. Both treatments with dexmedetomidine produced excellent sedation and recovery but induced more cardiovascular depression and haematological changes.

3.6 Acknowledgements

The authors would like to thank Pfizer GmbH Germany, Division Animal Health, for support of the study.
3.7 References


CULLEN, L. K. u. R. S. JONES (1977):
Clinical Observations on Xylazine-Ketamine Anesthesia in Cat.

The Effects of Medetomidine on Cardiac Contractility in Autonomically Blocked Dogs.

DOCHERTY, J. R. u. J. C. MCGRATH (1980):
A comparison of pre- and post-junctional potencies of several alpha-adrenoceptor agonists in the cardiovascular system and anococcygeus muscle of the rat. Evidence for two types of post-junctional alpha-adrenoceptor.
Naunyn Schmiedebergs Arch Pharmacol 312, 107-116.

Effect of dexmedetomidine, an alpha 2-adrenergic agonist, in the isolated heart.
J Cardiothorac Vasc Anesth 6, 418-423.

Evaluation of the clinical efficacy and safety of dexmedetomidine or medetomidine in cats and their reversal with atipamezole.
Vet Anaesth Analg 33, 214-223.

Cardiorespiratory effects of combined midazolam and butorphanol in isoflurane-anesthetized cats.
Vet Surg 22, 159-162.

Heart-Rate of the Cat.

Pharmacokinetics of ketamine HCl and metabolite I in the cat: a comparison of i.v., i.m., and rectal administration.
J Vet Pharmacol Ther 11, 84-93.

Left atrial to aortic root indices using two-dimensional and M-mode echocardiography in cavalier King Charles spaniels with and without left atrial enlargement.
Vet Radiol Ultrasound 43, 568-575.
Effects of alpha-adrenoceptor agonists and antagonists on insulin secreting cells and pancreatic blood vessels: comparative study.
Eur J Pharmacol 117, 253-257.

ILKIW, J. E., C. M. SUTER, T. B. FARVER, D. MCNEAL u. E. P. STEFFEY (1996):
The behaviour of healthy awake cats following intravenous and intramuscular administration of midazolam.
J Vet Pharmacol Ther 19, 205-216.

Effects of dexmedetomidine, a selective alpha 2-adrenoceptor agonist, on hemodynamic control mechanisms.
 Clin Pharmacol Ther 46, 33-42.

KANDA, T. u. Y. HIKASA (2008a):
Effects of medetomidine and midazolam alone or in combination on the metabolic and neurohormonal responses in healthy cats.

KANDA, T. u. Y. HIKASA (2008b):
Neurohormonal and metabolic effects of medetomidine compared with xylazine in healthy cats.

Ketamine: teaching an old drug new tricks.
Anesth Analg 87, 1186-1193.

Comparison of cardiopulmonary effects of medetomidine-midazolam, acepromazine-butorphanol and midazolam-butorphanol in dogs.
Zentralbl Veterinarmed A 46, 353-359.

LASCELLES, B. D. u. S. A. ROBERTSON (2004):
Antinociceptive effects of hydromorphone, butorphanol, or the combination in cats.

Establishment of lactate- and pyruvate measurement in plasma and liquor cerebrospinalis for diagnosis of mitochondrial malfunctions in dogs.
Hannover, Small animal clinic, Inaugural-Dissertation.


Dexmedetomidine continuous rate infusion during isoflurane anaesthesia in canine surgical patients. 
Vet Anaesth Analg 35, 1-12.

Pharmacological profiles of medetomidine and its antagonist, atipamezole. 

Characterization of the selectivity, specificity and potency of medetomidine as an alpha 2-adrenoceptor agonist.
Eur J Pharmacol 150, 9-14.

Ultrasonic Examination of the Heart in Dog and Cat by the 2-Dimensional (2d) and M-Mode-Method and Their Consequences for the Therapy of Cardiopathies. 
Kleintierpraxis 36, 459-468.

Cardiovascular effects of xylazine and detomidine in horses. 

The effect of four anesthetic protocols on splenic size in dogs. 
Vet Anaesth Analg 31, 102-108.
3.8 Tables and Figures

**Figure 1:** Chronological order of procedures

BP: Blood pressure measurement  
Echo: Echocardiography  
BS: Blood sampling  
Inj: Injection of treatment
<table>
<thead>
<tr>
<th>Description</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>No sedation, normal movement</td>
<td>0</td>
</tr>
<tr>
<td>Slight ataxia, able to stand</td>
<td>1</td>
</tr>
<tr>
<td>Strong ataxia, sternal recumbency</td>
<td>2</td>
</tr>
<tr>
<td>Lateral recumbency, strong reaction to stimulation</td>
<td>3</td>
</tr>
<tr>
<td>Lateral recumbency, slight reaction to stimulation</td>
<td>4</td>
</tr>
<tr>
<td>Lateral recumbency, no response to stimulation</td>
<td>5</td>
</tr>
</tbody>
</table>
**Table 2:** Recovery score evaluated by one anaesthetist, blinded to the treatment, using a numeric rating scale

<table>
<thead>
<tr>
<th>Observation</th>
<th>score</th>
<th>Patient criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Comfort</td>
<td>0</td>
<td>smooth</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>mild agitated</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>moderate agitated</td>
</tr>
<tr>
<td>Coordination</td>
<td>0</td>
<td>no ataxia</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>mild ataxia</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>moderate ataxia</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>severe ataxia</td>
</tr>
<tr>
<td>Vocalization</td>
<td>0</td>
<td>no vocalization</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>some vocalization</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>severe vocalization</td>
</tr>
<tr>
<td>Movement during sternal recumbency</td>
<td>0</td>
<td>normal</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>occasional position changes</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>frequent position changes</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>trashing</td>
</tr>
<tr>
<td>Locomotor activity</td>
<td>0</td>
<td>no locomotor activity</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>some</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>moderate</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>severe</td>
</tr>
<tr>
<td>Scratching and grooming</td>
<td>0</td>
<td>no scratching and grooming</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>scratching and grooming</td>
</tr>
</tbody>
</table>

**Total of recovery score** 0 - 14
**Table 3:** M-mode parameters at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB</th>
<th>MK</th>
<th>MBD</th>
<th>KD</th>
</tr>
</thead>
<tbody>
<tr>
<td>SAP (mmHg)</td>
<td>B 151 ± 8</td>
<td>149 ± 12</td>
<td>142 ± 12</td>
<td>152 ± 9</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>T 126 ± 28</td>
<td>112 ± 41</td>
<td>123 ± 20*</td>
<td>145 ± 19</td>
</tr>
<tr>
<td>EDD (cm)</td>
<td>B 206 ± 30</td>
<td>213 ± 17</td>
<td>202 ± 23</td>
<td>205 ± 16</td>
</tr>
<tr>
<td>ESD (cm)</td>
<td>B 0.66 ± 0.08</td>
<td>0.62 ± 0.16</td>
<td>0.65 ± 0.10</td>
<td>0.64 ± 0.09</td>
</tr>
<tr>
<td>FS (%)</td>
<td>T 52.03 ± 10.87*</td>
<td>46.07 ± 5.75a</td>
<td>36.58 ± 5.15* b</td>
<td>37.23 ± 5.25* b</td>
</tr>
<tr>
<td>SED (cm)</td>
<td>B 0.44 ± 0.04</td>
<td>0.38 ± 0.02</td>
<td>0.39 ± 0.07</td>
<td>0.43 ± 0.04</td>
</tr>
<tr>
<td>SES (cm)</td>
<td>T 0.74 ± 0.08a</td>
<td>0.71 ± 0.10</td>
<td>0.66 ± 0.07b</td>
<td>0.77 ± 0.05</td>
</tr>
<tr>
<td>LVWd (cm)</td>
<td>B 0.42 ± 0.04</td>
<td>0.40 ± 0.02</td>
<td>0.40 ± 0.07</td>
<td>0.45 ± 0.04</td>
</tr>
<tr>
<td>LVWs (cm)</td>
<td>B 0.74 ± 0.11</td>
<td>0.72 ± 0.10</td>
<td>0.70 ± 0.12</td>
<td>0.80 ± 0.08</td>
</tr>
<tr>
<td>EDV (ml)</td>
<td>B 5.52 ± 1.46</td>
<td>5.21 ± 0.79</td>
<td>5.54 ± 0.73</td>
<td>6.38 ± 2.35</td>
</tr>
<tr>
<td>ESV (ml)</td>
<td>B 4.22 ± 1.50*</td>
<td>4.43 ± 0.89</td>
<td>5.32 ± 1.28</td>
<td>5.25 ± 1.60</td>
</tr>
<tr>
<td>EF (%)</td>
<td>T 85.00 ± 10.11*</td>
<td>80.69 ± 5.68a</td>
<td>70.02 ± 6.71* b</td>
<td>70.94 ± 6.75* b</td>
</tr>
<tr>
<td>SV (ml)</td>
<td>B 4.83 ± 1.37</td>
<td>4.58 ± 0.69</td>
<td>4.86 ± 0.58</td>
<td>5.75 ± 2.17</td>
</tr>
<tr>
<td>Qt (l)</td>
<td>B 0.98 ± 0.31</td>
<td>1.05 ± 0.21</td>
<td>0.98 ± 0.16</td>
<td>1.09 ± 0.35</td>
</tr>
<tr>
<td>LA (cm)</td>
<td>B 1.19 ± 0.12</td>
<td>1.22 ± 0.09</td>
<td>1.18 ± 0.11</td>
<td>1.21 ± 0.07</td>
</tr>
<tr>
<td>AO (cm)</td>
<td>B 0.90 ± 0.11</td>
<td>0.87 ± 0.09</td>
<td>0.87 ± 0.08</td>
<td>0.89 ± 0.06</td>
</tr>
<tr>
<td>LA:AO</td>
<td>B 1.34 ± 0.12</td>
<td>1.40 ± 0.11</td>
<td>1.37 ± 0.04</td>
<td>1.37 ± 0.08</td>
</tr>
</tbody>
</table>

SAP: systolic arterial blood pressure; HR: heart rate; EDD: end-diastolic diameter; ESD: end-systolic diameter; FS: fractional shortening; SED: interventricular septal thickness in diastole; SES: interventricular septal thickness in systole; LVWd: left ventricular free wall thickness in diastole; LVWs: left ventricular free wall thickness in systole; EDV: end-diastolic volume; ESV: end-systolic volume; EF: ejection fraction; SV: stroke volume; Qt: cardiac output; LA: left atrium; AO: aortic diameter; LA:AO: Left atrium-to-aorta ratio

* significantly (p<0.05) different from baseline within groups

abdef groups with different superscripts in the same row differ significantly (p<0.05)
**Table 4:** Doppler-derived parameters at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB</th>
<th>MBK</th>
<th>MBD</th>
<th>KD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vel&lt;sub&gt;PV&lt;/sub&gt;</td>
<td>B 0.94 ± 0.25</td>
<td>1.00 ± 0.16</td>
<td>0.94 ± 0.21</td>
<td>0.94 ± 0.13</td>
</tr>
<tr>
<td></td>
<td>T 0.80 ± 0.20&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.75 ± 0.20&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.46 ± 0.07&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.46 ± 0.08&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Vel&lt;sub&gt;Ao&lt;/sub&gt;</td>
<td>B 1.10 ± 0.12</td>
<td>1.02 ± 0.05</td>
<td>1.05 ± 0.11</td>
<td>1.01 ± 0.10</td>
</tr>
<tr>
<td></td>
<td>T 0.95 ± 0.10&lt;sup*a&lt;/sup&gt;</td>
<td>0.80 ± 0.13&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.57 ± 0.06&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.60 ± 0.04&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Vel<sub>PV</sub>: peak velocity of the pulmonic flow; Vel<sub>Ao</sub>: peak velocity of the aortic flow

* significantly (p<0.05) different from baseline within groups

<sup>a</sup><sup>b</sup><sup>c</sup> groups with different superscripts in the same row differ significantly (p<0.05)
Table 5: Haematological parameters at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB</th>
<th>MBK</th>
<th>MBD</th>
<th>KD</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBC (10^3 µl⁻¹)</td>
<td>B 9.37 ± 1.31</td>
<td>9.71 ± 0.63</td>
<td>9.94 ± 0.78</td>
<td>9.78 ± 0.78</td>
</tr>
<tr>
<td>Hb (g dl⁻¹)</td>
<td>B 13.58 ± 0.94</td>
<td>13.62 ± 1.00</td>
<td>13.65 ± 0.68</td>
<td>13.93 ± 1.01</td>
</tr>
<tr>
<td>PCV (%)</td>
<td>B 40.85 ± 2.57</td>
<td>39.70 ± 2.19</td>
<td>39.85 ± 2.09</td>
<td>40.85 ± 4.53</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>B 44.10 ± 4.44</td>
<td>40.92 ± 1.33</td>
<td>40.18 ± 2.18</td>
<td>41.85 ± 4.30</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>B 14.65 ± 1.33</td>
<td>14.03 ± 0.35</td>
<td>13.78 ± 1.03</td>
<td>14.27 ± 1.13</td>
</tr>
<tr>
<td>MCHC (g dl⁻¹)</td>
<td>B 33.27 ± 0.62</td>
<td>34.27 ± 0.90</td>
<td>34.28 ± 1.66</td>
<td>34.23 ± 1.64</td>
</tr>
<tr>
<td>PLT (10^3 µl⁻¹)</td>
<td>B 350.83 ± 158.84</td>
<td>327.17 ± 145.82</td>
<td>323.67 ± 136.32</td>
<td>289.16 ± 122.35</td>
</tr>
</tbody>
</table>

RBC: red blood count; PCV: packed cell volume; MCV: mean corpuscular volume; MCH: mean corpuscular haemoglobin; MCHC: mean corpuscular haemoglobin concentration; PLT: platelets; MPV: mean platelet volume; WBC: white blood count

* significantly (p<0.05) different from baseline within groups
ab groups with different superscripts in the same row differ significantly (p<0.05)
Table 6: Venous blood gases at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB</th>
<th>MBK</th>
<th>MBD</th>
<th>KD</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.33 ± 0.03</td>
<td>7.36 ± 0.04</td>
<td>7.35 ± 0.06</td>
<td>7.35 ± 0.03</td>
</tr>
<tr>
<td></td>
<td>7.33 ± 0.02</td>
<td>7.35 ± 0.04</td>
<td>7.35 ± 0.04</td>
<td>7.38 ± 0.03*</td>
</tr>
<tr>
<td>pCO₂ (mmHg)</td>
<td>37.37 ± 3.58</td>
<td>36.72 ± 5.96</td>
<td>38.33 ± 6.60</td>
<td>38.40 ± 2.99</td>
</tr>
<tr>
<td></td>
<td>38.93 ± 2.55</td>
<td>40.13 ± 3.11</td>
<td>42.05 ± 2.31</td>
<td>39.37 ± 2.33</td>
</tr>
<tr>
<td>pO₂ (mmHg)</td>
<td>51.05 ± 6.39</td>
<td>48.08 ± 6.68</td>
<td>48.13 ± 9.81</td>
<td>50.30 ± 9.59</td>
</tr>
<tr>
<td></td>
<td>48.35 ± 5.79</td>
<td>45.10 ± 5.05</td>
<td>41.50 ± 4.60</td>
<td>40.67 ± 7.62*</td>
</tr>
<tr>
<td>HCO₃⁻ (mmol l⁻¹)</td>
<td>19.07 ± 0.98</td>
<td>20.35 ± 1.65</td>
<td>20.42 ± 2.02</td>
<td>20.47 ± 1.07</td>
</tr>
<tr>
<td></td>
<td>19.82 ± 0.95</td>
<td>21.72 ± 0.69*</td>
<td>22.78 ± 1.87</td>
<td>22.52 ± 1.92*</td>
</tr>
</tbody>
</table>

pCO₂: partial pressure of carbon dioxide; pO₂: partial pressure of oxygen; HCO₃⁻: hydrogen carbonate
* significantly (p<0.05) different from baseline within groups
Table 7: Electrolytes at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB</th>
<th>MBK</th>
<th>MBD</th>
<th>KD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium (mmol l⁻¹)</td>
<td>150.13 ± 1.92</td>
<td>152.13 ± 2.72</td>
<td>151.02 ± 2.42</td>
<td>152.03 ± 1.91</td>
</tr>
<tr>
<td>Potassium (mmol l⁻¹)</td>
<td>3.94 ± 0.10</td>
<td>3.91 ± 0.19</td>
<td>3.79 ± 0.39</td>
<td>4.10 ± 0.47</td>
</tr>
<tr>
<td>Calcium ion (mmol l⁻¹)</td>
<td>1.20 ± 0.07</td>
<td>1.17 ± 0.06</td>
<td>1.14 ± 0.04</td>
<td>1.22 ± 0.13</td>
</tr>
<tr>
<td>Chloride (mmol l⁻¹)</td>
<td>112.00 ± 3.10</td>
<td>111.83 ± 2.64</td>
<td>113.33 ± 2.50</td>
<td>110.83 ± 3.19</td>
</tr>
</tbody>
</table>

* ion: ionized
* significantly (p<0.05) different from baseline within groups
Table 8: Serum profile and at baseline (B) and 10 minutes after (T) intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MB B</th>
<th>MBK T</th>
<th>MBD T</th>
<th>KD T</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactate (mmol l⁻¹)</td>
<td>2.91 ± 1.10</td>
<td>3.14 ± 1.04</td>
<td>2.97 ± 0.62</td>
<td>3.31 ± 0.74</td>
</tr>
<tr>
<td>Glucose (mmol l⁻¹)</td>
<td>6.25 ± 0.95</td>
<td>5.90 ± 0.68</td>
<td>6.54 ± 2.09</td>
<td>5.98 ± 0.66</td>
</tr>
<tr>
<td>ALT (U l⁻¹)</td>
<td>36.43 ± 7.28</td>
<td>30.87 ± 10.91</td>
<td>37.04 ± 28.48</td>
<td>34.87 ± 7.98</td>
</tr>
<tr>
<td>GLDH (U l⁻¹)</td>
<td>1.48 ± 0.55</td>
<td>2.37 ± 0.55</td>
<td>1.90 ± 0.52</td>
<td>1.69 ± 0.37</td>
</tr>
<tr>
<td>TP (g dl⁻¹)</td>
<td>101.35 ± 18.89</td>
<td>93.52 ± 13.08</td>
<td>97.70 ± 19.23</td>
<td>108.30 ± 19.72</td>
</tr>
<tr>
<td>PT (%)</td>
<td>97.28 ± 21.35</td>
<td>89.87 ± 8.85</td>
<td>95.77 ± 14.35</td>
<td>93.58 ± 12.55</td>
</tr>
<tr>
<td>PTT (sec)</td>
<td>12.17 ± 0.67</td>
<td>12.65 ± 0.80</td>
<td>12.60 ± 0.77</td>
<td>11.71 ± 1.71</td>
</tr>
</tbody>
</table>

ALT: alanine aminotransferase; GLDH: glutamate dehydrogenase; AST: aspartate aminotransferase; TP: total protein; PT: prothrombin time; PTT: partial thromboplastin time

* significantly (p<0.05) different from baseline within groups

abc groups with different superscripts in the same row differ significantly (p<0.05)
Figure 2: Sedation score after intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD)

\[ a,b,c \] protocols with different superscripts differ significantly (p<0.05)
Figure 3: Recovery score after intramuscular injection of four different drug combinations: midazolam + butorphanol (MB), midazolam + butorphanol + ketamine (MBK), midazolam + butorphanol + dexmedetomidine (MBD), ketamine + dexmedetomidine (KD) in six cats (mean ± SD).

a,b,c protocols with different superscripts differ significantly (p<0.05)
4 General Discussion

4.1 Materials and Methods

The main aim of the thesis was to compare the effects of midazolam and butorphanol, either with ketamine or dexmedetomidine and ketamine combined with dexmedetomidine alone on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats.

Cats received the different treatments in a randomized cross-over design. This design has the advantage of eliminating individual subject differences from the overall treatment effect, thus enhancing statistical power. Health status of cats was assessed by means of physical examination, echocardiographic examination, a complete blood count and serum biochemical analyses. All findings were within reference range. Due to their physical health and to the fact that they were all domestic short hair cats, we could eliminate individual breed differences and disease effects as well.

Systolic arterial blood pressure measurement was performed using Doppler ultrasonographic flowmeter. As we tried to mimic clinical situation direct arterial blood pressure measurement was not carried out as this method is too complex to perform in routine examinations. However, BINNS et al. (1995) compared three different indirect blood pressure measurements and reported the highest overall accuracy of Doppler when compared with direct arterial blood pressure measurement.

In both studies, echocardiography was performed by a single experienced observer to avoid interobserver differences. SIMPSON et al. (2007) demonstrated discrepancies between observers for certain echocardiographic parameters and state that when possible the same clinician should perform repeated measurements in any one individual patient.

The anaesthetist, who evaluated the videotaped recovery and the echocardiographer, both were blinded to the treatments cats received. So it was guaranteed that they were not prejudiced against drug effects.
4.2 Results

As we wanted to compare the echocardiographic effects of four different drug combinations we initially tested four ultrasound methods to evaluate stroke volume and cardiac output. Thus, the best repeatable ultrasound method obtaining the most representative results was chosen to compare the four treatments.

Regarding the between-day intra-observer variability of parameters obtained with M-mode and pulsed-wave Doppler, the coefficient of variation was less than 8.7, demonstrating a very good repeatability. In accordance with our findings, SIMPSON et al. (2007) also reported highly repeatable measurements using M-mode and spectral Doppler in cats by a single investigator.

The between-day variability of LV volumes, EF, SV and Qt assessed by four different echocardiographic techniques (ie, Teichholz method, SM, ALM, Trace method) were acceptable (CV < 20%), except for SV and Qt measured with SM and ALM. In contrast to our results, SERRES et al. (2008) reported a lower variability (CV < 11%) for both planimetric methods when used in dogs. A reason for the high CV in the current study might be the small size of the feline heart, which can be responsible for suboptimal images (MOISE u. FOX 1999).

Regarding the absolute SV and Qt values, we observed significant differences between the four techniques. The volumetric flow method obtained highest values, followed by Teichholz method and both planimetric methods gained lowest values. In the current study we did not measure SV and Qt by thermodilution, the gold standard to determine these parameters. However, values obtained by the Teichholz method (SV: 5.01 ± 1.43 ml/beat; Qt: 1.02 ± 0.30 l/min) showed the best convergence in comparison with SV and Qt values obtained by thermodilution method in previous studies in awake cats with similar weights (ALLEN et al. 1986; DYSON et al. 1987; DYSON et al. 1988; INGWERSEN et al. 1988; LAMONT et al. 2001), whereas values obtained by the Trace method were slightly higher and both planimetric methods underestimated SV and Qt.

The Teichholz method seemed to be the best suitable technique for echocardiographic measurements in the cat. So this method was used in the main study in which effects of midazolam and butorphanol, either with ketamine or
dexmedetomidine and ketamine combined with dexmedetomidine alone on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats were compared.

Midazolam combined with butorphanol failed to produce adequate sedation and caused cats to behave in a dysphoric or aggressive manner. ILKIW et al. (1996) reported similar effects on cat’s behaviour when administering midazolam intramuscularly alone. Butorphanol in a dose of 0.4 mg kg\textsuperscript{-1} may not be able to reduce the adverse behavioural effects of midazolam in healthy cats or might even cause central stimulation. Deepest and longest sedation was achieved with treatments including dexmedetomidine. The α\textsubscript{2}-agonists are well known as potent sedatives (MURRELL u. HELLEBREKERS 2005). Ketamine combined with dexmedetomidine led to a higher sedation score, meaning a faster and deeper sedation, compared with MBD. One may conclude from this that ketamine in a dose of 3 mg kg\textsuperscript{-1} has more sedative properties than midazolam combined with butorphanol, both in a dose of 0.4 mg kg\textsuperscript{-1}. Recovery times were faster with MBK than with both treatments with dexmedetomidine. Especially in two cats after treatment with MBK recovery times were not measurable as both animals were already standing after the procedures (approximately 40 minutes after treatment injection). Best quality of recovery was obtained with ketamine combined with dexmedetomidine. This combination caused cats to recover very smooth, well coordinated and with less activity in the cage. Other studies as well reported satisfactory recoveries after anaesthesia with ketamine in combination with α\textsubscript{2}-agonists in cats (CULLEN u. JONES 1977).

Due to sympathetic tone cats have a higher heart rate in hospital than at home and even higher under restraint during echocardiography (ABBOTT 2005). Heart rates during echocardiography obtained in this study were similar to rates recorded by others (HAMLIN 1989; ABBOTT 2005). A statistically significant (p<0.05) increase in HR was observed after treatment with MB making echocardiography even more difficult (SANTILLI u. BUSSADORI 1998). The decreases in ESD, EDV and ESV after MB administration resulted in a statistically significantly (p<0.05) decreased stroke volume leading to a decrease in cardiac output (23% decrease). The
decreased SV might be the consequence of an impaired left ventricular filling due to an increased HR. Nevertheless, cardiac output was influenced by MB to a lower extent compared to other treatments used in this study. Similar to our findings, KOJIMA et al. (1999) observed only small changes in blood pressure and cardiac output in dogs sedated with midazolam 0.1 mg kg\(^{-1}\) and butorphanol 0.2 mg kg\(^{-1}\) intramuscularly.

Although heart rate and stroke volume were not statistically significantly (p<0.05) decreased after MBK, this treatment produced a statistically significant (p<0.05) decrease in cardiac output (34% decrease), but not as severe as MBD (54% decrease) and KD (53% decrease).

Both treatments with dexmedetomidine, MBD and KD, had more depressant effects on cardiovascular parameters in comparison to the other treatments. After their administration heart rate, fractional shortening, stroke volume and cardiac output were statistically significantly (p<0.05) decreased from baseline due to central and peripheral \(\alpha_2\)-adrenoceptor activation. As a result of a decrease in central sympathetic action and an indirect baroreceptor-mediated increase in vagal tone bradycardia occurs after dexmedetomidine administration (VIRTANEN 1989). In our study ESV significantly increased after MBD and KD. Reasons for this increase may be the reduced contractility of the heart on the one hand and the \(\alpha_2\)-agonist-mediated increase in afterload caused by vasoconstriction on the other hand. The higher ESV resulted in a statistically significant (p<0.05) decrease in stroke volume which in addition to the decreased HR led to a decreased cardiac output. SCHMELING et al. (1991) and DEMORAIS and MUIR (1995) found that the decrease in cardiac output after medetomidine administration is not a direct depression of myocardial contractility but primarily caused by the decrease in heart rate and increase in vascular resistance. Results of a study by FLACKE et al. (1992), in which dexmedetomidine was investigated in isolated dog hearts, support these findings, as no myocardial depressant effect was found. So far, in myocardium no \(\alpha_2\) -adrenoceptor evidence has been provided. The marked reduction of cardiac output (approximately 50% decrease) in our study is comparable to data obtained from
ALLEN et al. (1986) who measured Qt by thermodilution after administration of a xylazine (1.0 mg kg\(^{-1}\)) - ketamine (10 mg kg\(^{-1}\)) combination in cats. 

In the current study, all treatments decreased systolic arterial blood pressure. Alpha\(_2\)-agonists have a biphasic dose-related (KALLIO et al. 1989) effect on blood pressure. Firstly, stimulation of peripheral \(\alpha_2\)-agonist receptors leads to vasoconstriction which mediates a transitory increase in blood pressure (DOCHERTY u. MCGRATH 1980). Secondly, prolonged hypotension occurs due to a central hypotensive effect and a decrease in sympathetic nervous tone. Due to the facts that we used dexmedetomidine in a low dose of 5 µg kg\(^{-1}\) and performed blood pressure measurement only approximately 30 minutes after treatment and the initial hypertensive phase could not be detected SAP was decreased after treatment.

Red blood cell count, Hb and PCV statistically significantly (p<0.05) decreased after treatment with MBK, MBD and KD. This phenomenon may be explained by pooling of the circulating blood cells in the spleen or other reservoirs due to the decreased sympathetic activity. However, WILSON et al. (2004) investigated the influence of four different anaesthetic protocols on splenic size in dogs and found out that the administration of medetomidine, ketamine and diazepam leads to a greater splenic volume compared with acepromazine and propofol but that there is a lack of correlation between the packed cell volume and splenic size. They suggested sequestration of red blood cells in nonsplenic sites. Another reason might be the shifting of fluid from extravascular compartment to intravascular compartment (WAGNER et al. 1991) in order to maintain normal cardiac output in the animal.

In this study, glucose concentration had a marked increase after treatment with MBD and KD. The responsible mechanism behind the phenomenon is the inhibition of insulin secretion through the binding of \(\alpha_2\)-agonists on \(\alpha_2\beta\)-receptors in the \(\beta\) cells of the pancreas (HILLAIRE-BUYS et al. 1985).

In our study plasma lactate concentrations at baseline were quiet high but still within the reference range (1.0-3.5 mmol l\(^{-1}\)). Catecholamine release and muscle activation due to stress during blood sampling may be reasons for higher lactate values. In the current study cats were quiet excited after echocardiography at baseline when the first blood sampling was performed. All treatments except MB led to a statistically
significant (p<0.05) decrease in plasma lactate concentrations. Responsible reasons may be muscle relaxation, stress reduction and redistribution during sedation. There are concerns about the adequate tissue perfusion after dexmedetomidine due to its depressant effect on cardiac output and increased vascular resistance. Similar to our findings, UILENREEF et al. (2008), who investigated changes in plasma lactate concentrations during dexmedetomidine continuous rate infusion (1-3 µg kg\(^{-1}\) hour\(^{-1}\)) in dogs, reported values under 2 mmol l\(^{-1}\). In our study, lactate concentrations even decreased after treatments indicating that there was no appearance of inadequate tissue oxygen supply. As plasma total protein concentrations decreased as well, another reason for lower lactate concentrations after treatment might be a dilutional effect.

4.3 Conclusion and outlook

In conclusion, according to our results of the initial study, the use of the geometric method based on the Teichholz formula is a well repeatable method for the assessment of SV and Qt in healthy cats and obtained values comparable to previous studies which have performed Qt measurement by using thermodilution. Both planimetric methods should not be recommended for the assessment of LV volume in cats, because these methods were not well repeatable and seemed to underestimate the values for SV and Qt. The Trace method was best repeatable, but appeared to overestimate SV and Qt values. Further investigations should be carried out to compare described ultrasound methods to thermodilution or direct Fick method and to test their use in cats with heart disease.

The main study showed that midazolam with butorphanol failed to produce adequate sedation in healthy cats and caused them to behave dysphoric and aggressive. Cardiovascular and haematological changes were only minimal with this combination. Treatment with MBK led to acceptable sedation and mild cardiovascular changes. Both treatments with dexmedetomidine produced excellent sedations and recoveries but induced more cardiovascular depression and haematological changes.
Further studies investigating the four treatments in cats with heart disease would be interesting, as well as the use of dexmedetomidine in a lower dose, for example of 3 \( \mu g \ kg^{-1} \), to ease the cardiovascular depression but may be to maintain excellent sedation and recovery.
Einfluss verschiedener Sedationsprotokolle auf echokardiographische, hämatologische und biochemische Parameter bei der Katze


In einem kompletten Cross over Design wurden 6 gesunde Katzen für die Studie untersucht. Jede Katze erhielt jedes der folgenden vier Sedationsprotokolle in zufälliger Reihenfolge mit einem Intervall von mindestens acht Tagen: Midazolam 0,4 mg/kg mit Butorphanol 0,4 mg/kg (MB), kombiniert sowohl mit Ketamin 3 mg/kg (MBK), als auch mit Dexmedetomidin 5 µg/kg (MBD) und Ketamin 3 mg/kg mit Dexmedetomidin 5 µg/kg (KD) alleine. Die verschiedenen Substanzen wurden in einer Mischspritze intramuskulär verabreicht. Der Sedationsgrad wurde 2, 4, 6, 8 und 10 Minuten nach Injektion mittels eines Punktesystems (0-5) bestimmt. Getestet wurden das Verhalten und die Reaktion auf eine Stimulation, die sowohl auditorisch als auch takttil erfolgte. Jeweils vor (B) und nach (T) Sedation wurde eine echokardiographische Untersuchung, eine Blutdruckmessung und Blutentnahme für eine hämatologische und blutchemische Untersuchung durchgeführt. Die echokardiographische Untersuchung beinhaltete neben Routineuntersuchungen die Messung des Schlagvolumens und des Herzauswurfs mittels vier verschiedener Methoden: Teichholz Methode (geometrische Methode), Simpsons Methode und
Zusammenfassung

area length Methode (planimetrische Methoden) und Trace Methode (volumetrische Flußmethode).
Am Ende der Untersuchungen wurden die Tiere in einen Käfig gelegt, die Aufwachphase auf Video aufgenommen und die Zeit bis zum Aufrichten der Katze in Brustlager und bis zum Hinstellen gemessen. Die Qualität der Aufwachphase wurde von einem Anästhesisten mittels einer Numerisch Deskriptiven Skala (NDS) evaluiert, der das jeweilig applizierte Protokoll nicht kannte.
Der p-Wert wurde auf 0,05 festgelegt.
Die Ausgangswerte der echokardiographischen Untersuchung unterschieden sich nicht zwischen den Protokollen. Alle Werte befanden sich innerhalb der Referenzbereiche. Der VK für die Messungen von Schlagvolumen und Herzauswurf der Teichholz und Trace Methode lag < 20%, der der Simpsons und area length Methode > 20%. Die Trace Methode lieferte statistisch signifikant (p<0,05) höhere Werte für das Schlagvolumen und den Herzauswurf verglichen mit allen anderen Methoden, wohingegen die Simpsons und area length Methode statistisch signifikant (p<0,05) niedrigere Werte erzielten. Die Bland-Altman Analyse ergab die beste Übereinstimmung zwischen beiden planimetrischen Methoden. Alle Protokolle führten zu einem Abfall des systolisch arteriellen Blutdrucks, welcher mit MBD (von 142 ± 12 auf 123 ± 20 mmHg) statistisch signifikant (p<0,05) war. Die Herzfrequenz fiel statistisch signifikant (p<0,05) nach Sedation mit MBD (44% Abfall) und KD (34% Abfall) und stieg statistisch signifikant (p<0,05) nach Sedation mit MB (5% Anstieg) an. Alle Protokolle außer MBK führten zu einem statistisch signifikanten (p<0,05) Abfall des Schlagvolumens ohne statistisch signifikante Unterschiede zwischen den
Protokollen. Der Herzauswurf wurde durch alle Protokolle statistisch signifikant (p<0,05) gesenkt. Beide Protokolle mit Dexmedetomidin erzielten die niedrigsten Herzauswürfe (MBD: 0,45 ± 0,11 l/min; KD: 0,51 ± 0,15 l/min). Die Protokolle MBK, MBD und KD senkten den Hämatokrit statistisch signifikant (p<0,05), wohingegen der Einfluss aller Protokolle auf die Elektrolyte und Serumchemie gering war. Die Glukosekonzentration im Plasma stieg nach Injektion von MBD und KD statistisch signifikant (p<0,05) an. Der niedrigste Sedationsgrad wurde mit MB (12,33 ± 5,13) erreicht, der höchste mit KD (39,00 ± 4,00). Nach Gabe von MB verhielten sich vier Katzen abwehrbereit, teilweise sogar aggressiv. Die Qualität der Aufwachphase war am besten mit KD (0,67 ± 0,8) und am schlechtesten mit MB (7,33 ± 2,3).

6 Summary

Kirsten Biermann

Effects of different sedative drug combinations on echocardiographic, haematologic and biochemical parameters in cats

One aim of this study was to test different echocardiographic approaches to calculate cardiac output using Teichholz method, Simpson’s method, area length method and the Trace method and to evaluate the repeatability and agreement of these methods in healthy cats. Using the ultrasound method with best repeatability, the second aim of this study was to compare the effects of midazolam and butorphanol, either with ketamine or dexmedetomidine and ketamine combined with dexmedetomidine alone on sedation, echocardiographic, haematologic and biochemical parameters and recovery in cats.

Six healthy cats were included into the study. Each cat received each of the following treatments intramuscularly (IM) in a randomised cross-over design with at least 8 days wash-out: midazolam 0.4 mg kg\(^{-1}\) with butorphanol 0.4 mg kg\(^{-1}\) (MB), combined with ketamine 3 mg kg\(^{-1}\) (MBK) or dexmedetomidine 5 µg kg\(^{-1}\) (MBD) or ketamine 3 mg kg\(^{-1}\) with dexmedetomidine 5 µg kg\(^{-1}\) (KD) alone. Sedation was scored on a scale from 0 = no sedation to 5 = lateral recumbency, no response, as the response to tactile and auditory stimulation and was recorded 2, 4, 6, 8 and 10 minutes after injection. Echocardiography, systolic arterial blood pressure (SAP) measurement and blood sampling were performed at baseline and 10 minutes after treatment. Echocardiography included measurement of standard parameters as well as stroke volume (SV) and cardiac output (Qt) measurements obtained by four different methods: Teichholz method (geometric method), Simpson’s method and area length method (planimetric methods) and trace method (volumetric flow method).

Recovery was videotaped and quality was scored by the same anaesthetist, blinded to the protocol, using a numeric descriptive scale (NDS). Times to sternal position and standing were recorded. Data are presented as mean ± SD and were analysed
Summary

by ANOVA for repeated measures and paired t-tests. Sedation score was calculated over 10 minutes by area under the curve (AUC). The coefficient of variation (CV) was calculated to evaluate between-day intra-observer repeatability of different cardiac output measurements and Bland-Altman analysis was used to assess agreement between methods. Alpha was set at 5%.

Baseline values did not differ among treatments. The CV was acceptable (CV < 20%) for all parameters, except SV and Qt obtained by using the Simpson’s method (28.8%; 22.4%) and area length method (21.6%; 22.6%). Values of SV and Qt obtained with the Trace method were significantly higher compared with all other methods. Both planimetric methods resulted in the lowest values for SV and Qt. Tight limits of agreement were observed between both planimetric methods regarding all parameters. Comparison between planimetric methods and both other methods resulted in wider limits of agreement.

All treatments caused a decrease in SAP of 17%, 25%, 13%, 5% in MB, MBK, MBD and KD, respectively, which was statistically significant (p<0.05) with MBD. Heart rate statistically significantly (p<0.05) increased after MB (5%) and statistically significantly (p<0.05) decreased after MBD (44%) and KD (34%). All treatments statistically significantly (p<0.05) (except MBK) decreased SV by 24%, 21%, 24%, 36% in MB, MBK, MBD and KD, and Qt by 23%, 34%, 54%, 53%, respectively. All treatments but MB produced a statistically significant (p<0.05) decrease in packed cell volume of 20%, 31%, 29% in MBK, MBD and KD, respectively. There were minimal effects on electrolytes and serum chemistry. Glucose concentration was increased after treatment with MBD (31%) and KD (52%) and lactate concentration was statistically significantly (p<0.05) decreased after MBK (58%), MBD (72%) and KD (65%). The lowest sedation score was obtained with MB (12.33 ± 5.13) and the highest with KD (39.00 ± 4.00). Quality of recovery was best with KD (0.67 ± 0.8) and worst with MB (7.33 ± 2.3). Evaluation of recovery times was only possible for MBD and KD. Times to sternal recumbency and standing were faster after treatment with KD (46 ± 8 min; 56 ± 16 min) compared to MBD (77 ± 33 min; 87 ± 36 min).

In conclusion, Qt can be calculated by a geometric method based on Teichholz formula with good repeatability and obtaining Qt values comparable to previous
Studies in which Qt measurement was performed by using thermodilution. Both planimetric methods were not well repeatable and seemed to underestimate SV and CO, whereas the Trace method appeared to overestimate these parameters. Midazolam with butorphanol failed to produce adequate sedation in healthy cats and caused them to behave dysphoric and aggressive. Cardiovascular and haematological changes were only minimal with this combination. Protocol MBK led to an acceptable sedation and minimal cardiovascular changes. Both treatments with dexmedetomidine produced excellent sedation and recovery but induced more cardiovascular depression and haematologic changes.
7 Literaturverzeichnis


CHILD, K. J., B. DAVIS, M. G. DODDS u. D. J. TWISSELL (1972):  
Anaesthetic, cardiovascular and respiratory effects of a new steroidal agent CT 1341:  
a comparison with other intravenous anaesthetic drugs in the unrestrained cat.  
Br J Pharmacol 46, 189-200.

Cardiovascular effects of ketamine in the pithed rat, rabbit and cat.  
Br J Anaesth 48, 935-939.

Butorphanol: effects of a prototypical agonist-antagonist analgesic on kappa-opioid receptors. 

CULLEN, L. K. u. R. S. JONES (1977):  
Clinical Observations on Xylazine-Ketamine Anesthesia in Cat.  
Ve Rec 101, 115-116.

Two-Dimensional Echocardiography in the Normal Cat.  
Vet Radiol 26, 149-158.

The Effects of Medetomidine on Cardiac Contractility in Autonomically Blocked Dogs.  

DOCHERTY, J. R. u. J. C. MCGRATH (1980):  
A comparison of pre- and post-junctional potencies of several alpha-adrenoceptor agonists in the cardiovascular system and anococcygeus muscle of the rat. Evidence for two types of post-junctional alpha-adrenoceptor. 
Naunyn Schmiedebergs Arch Pharmacol 312, 107-116.

evaluation of acepromazine/meperidine/atropine premedication followed by thiopental anesthesia in the cat.  
Can J Vet Res 52, 419-422.

Effects of saffan on cardiopulmonary function in healthy cats.  

Feigenbaum’s Echocardiography.  
6th, Lippincott Williams & Wilkins, Philadelphia.
Effect of dexmedetomidine, an alpha 2-adrenergic agonist, in the isolated heart.
J Cardiothorac Vasc Anesth 6, 418-423.

Evaluation of the clinical efficacy and safety of dexmedetomidine or medetomidine in cats and their reversal with atipamezole.
Vet Anaesth Analg 33, 214-223.

Cardiorespiratory effects of combined midazolam and butorphanol in isoflurane-anesthetized cats.
Vet Surg 22, 159-162.

Visually estimated left ventricular ejection fraction by echocardiography is closely correlated with formal quantitative methods.
Int J Cardiol 101, 209-212.

Heart-Rate of the Cat.

Pharmacokinetics of ketamine HCl and metabolite I in the cat: a comparison of i.v., i.m., and rectal administration.
J Vet Pharmacol Ther 11, 84-93.

Left atrial to aortic root indices using two-dimensional and M-mode echocardiography in cavalier King Charles spaniels with and without left atrial enlargement.
Vet Radiol Ultrasound 43, 568-575.

Effects of alpha-adrenoceptor agonists and antagonists on insulin secreting cells and pancreatic blood vessels: comparative study.
Eur J Pharmacol 117, 253-257.

ILKIW, J. E., C. M. SUTER, T. B. FARVER, D. MCNEAL u. E. P. STEFFEY (1996):
The behaviour of healthy awake cats following intravenous and intramuscular administration of midazolam.
J Vet Pharmacol Ther 19, 205-216.
Cardiopulmonary effects of a ketamine hydrochloride/acepromazine combination in healthy cats.

Effects of dexmedetomidine, a selective alpha 2-adrenoceptor agonist, on hemodynamic control mechanisms.
Clin Pharmacol Ther 46, 33-42.

KANDA, T. u. Y. HIKASA (2008a):
Effects of medetomidine and midazolam alone or in combination on the metabolic and neurohormonal responses in healthy cats.

KANDA, T. u. Y. HIKASA (2008b):
Neurohormonal and metabolic effects of medetomidine compared with xylazine in healthy cats.

Ketamine: teaching an old drug new tricks.
Anesth Analg 87, 1186-1193.

Comparison of cardiopulmonary effects of medetomidine-midazolam, acepromazine-butorphanol and midazolam-butorphanol in dogs.
Zentralbl Veterinarmed A 46, 353-359.

KRONIK, G., J. SLANY u. H. MOSSLACHER (1979):
Comparative value of eight M-mode echocardiographic formulas for determining left ventricular stroke volume. A correlative study with thermodilution and left ventricular single-plane cineangiography.
Circulation 60, 1308-1316.

Comparison of pulmonary artery and transpulmonary thermodilution cardiac output measurement in anaesthetized cats.
In: AVA/ECVAA Meeting Leipzig, 47.


Radiographic Interpretation for the Small Animal Clinician.
second, Williams & Wilkins, Baltimore.

A rose by any other name: cardiac output.

Echocardiography in the domestic cat.

Comparison of Doppler-derived aortic velocities obtained from various transducer sites in healthy dogs and cats.
Vet Radiol Ultrasound 48, 570-573.

Comparison of pulmonary artery and arterial thermodilution cardiac output in critically ill patients.

Doppler echocardiographic study of left ventricular diastole in non-anaesthetized healthy cats.

The Effects of the Stereoisomers of the Alpha-2-Adrenergic Agonist Medetomidine on Systemic and Coronary Hemodynamics in Conscious Dogs.
Anesthesiology 75, 499-511.

Evaluation of the sedative and cardiorespiratory effects of dexametomidine, dexametomidine-butorphanol, and dexametomidine-ketamine in cats.

Comparison of 3 ultrasound methods for quantifying left ventricular systolic function: correlation with disease severity and prognostic value in dogs with mitral valve disease.
Assessment of the repeatability of feline echocardiography using conventional echocardiography and spectral pulse-wave Doppler tissue imaging techniques. Vet Radiol Ultrasound 48, 58-68.

Effects of anesthesia on echocardiographic assessment of left ventricular structure and function in rats. Basic Res Cardiol 102, 28-41.

TEICHHOLZ, L. E., T. KREULEN, M. V. HERMAN u. R. GORLIN (1976):
Problems in echocardiographic volume determinations: echocardiographic-angiographic correlations in the presence of absence of asynergy. Am J Cardiol 37, 7-11.


Comparison and reproducibility of visual echocardiographic and quantitative radionuclide left ventricular ejection fractions. Am J Cardiol 77, 843-850.


Danksagung

Mein herzlicher Dank gilt Frau Prof. Dr. Sabine Kästner für die Überlassung des interessanten Themas, die jederzeit gewährte Unterstützung und hervorragende Betreuung beim Anfertigen dieser Arbeit!

Ganz besonders danke ich Herrn Dr. Stephan Hungerbühler für die Durchführung der Herzsonographien, für die fachlich sehr hilfreiche Unterstützung sowie für die witzige Zeit im Ultraschall!

Vielen Dank an Herrn Prof. Dr. Reinhard Mischke für die konstruktiven Hilfestellungen bezüglich der Laboruntersuchungen.

Herrn Prof. Dr. Ingo Nolte danke ich für die Bereitstellung der Geräte sowie für das großzügige Entgegenkommen hinsichtlich der Behandlung drei erkrankter Katzen.

Herzlich bedanke ich mich bei Vike Marth-Begovic und Dirk Menzel für die Bestimmung der Laborparameter sowie bei Andreas Köppen und Olaf de la Roi für die Hilfe bezüglich jeglicher technischer Schwierigkeiten inklusive der Formatierung der Arbeit.

Ein großer Dank gilt allen Tierpflegern des Instituts für Tierernährung sowie allen Mitarbeitern der Klinik für Kleintiere für ihre tatkräftige Einsatzbereitschaft! Besonders danke ich Julia für ihren moralischen Beistand und die stets kompetente Hilfe!

Von Herzen danke ich Tilman für seine großartige Unterstützung und liebevolle Fürsorge sowie für die Hilfestellungen bei der Formatierung der Arbeit und die Korrekturen!

Der größte Dank gebührt meiner Familie, insbesondere meinen Eltern, die mich immer unterstützt haben und mir jederzeit mit Rat und Tat zur Seite standen. Ohne Euch wäre diese Arbeit nicht zustande gekommen!