PERIOPERATIVE FLUID ADMINISTRATION TO OPTIMISE HAEMODYNAMICS WITHOUT FLUID OVERLOAD IN ANAESTHETISED DOGS

A thesis presented in partial fulfilment

of the requirements for the degree of

Doctor of Philosophy

in

Veterinary Science

at Massey University, Manawatu

New Zealand

HIROKI SANO

2021

Dedication:

To my colleagues at the veterinary teaching hospital, Massey University,

Thank you for helping my research projects,

Thank you for your support and patience,

I really appreciate your invaluable assistance in this work.

ABSTRACT

Perioperative fluid therapy is the mainstay of anaesthetic management. Fluid administration improves haemodynamics during anaesthesia as it increases preload and thus cardiac output and blood pressure. However, excessive fluid administration can cause detrimental adverse effects, such as haemodulution and oedema, resulting in prolonged hospital stay and increased morbidity and mortality in people. Therefore, fluid administration should be restricted to those who are able to increase stroke volume or cardiac output in response to the fluid administration (responders) and should not be given to those who are unable to do so (non-responders) based on the famous "Frank–Starling law of the heart"

Previously static parameters such as central venous pressure were believed to be a clinical gold standard to estimate preload and fluid responsiveness. Over the last decade, dynamic parameters such as pulse pressure variation and pleth variability index have been shown to be reliable predictors for fluid responsiveness in people. This study found that pulse pressure variation and pleth variability index were more accurate than central venous pressure for predicting fluid responsiveness in dogs.

Mini-fluid challenge is another technique that is currently available and can be reliably used to determine fluid responsiveness in human medicine. Mini-fluid challenge is an administration of a small amount of fluid to increase preload. Thus, fluid responsiveness can be assessed based on whether stroke volume increases following mini-fluid challenge according to the Frank-Starling curve. The change in stroke volume of a heart at the steep portion of the Frank-Starling curve will be greater than at the plateau portion after mini-fluid challenge. The studies revealed a percentage change in pulse wave transit time (a

surrogate parameter of stroke volume, which was also one of results in this thesis) following mini-fluid challenge could predict fluid responsiveness in mechanically ventilated anaesthetised dogs under an experimental condition, and spontaneously breathing anaesthetised dogs undergoing stifle surgery in clinical setting.

Lastly, these methods are still of limited use in veterinary clinical practice because of availability of equipment, difficulty of their interpretation and a cumbersome process. The main purpose of this thesis was to obtain evidence on how to optimise haemodynamics in anaesthetised dogs and prevent excessive fluid administration. The time when most practitioners administer a bolus of fluid during anaesthesia is when hypotension is encountered because of anaesthesia. Thus, prevention of hypotension could avoid excessive fluid administration. Therefore, the study found that prophylactic noradrenaline administration, which counteracts some of the cardiovascular adverse effects of anaesthesia, was able to prevent hypotension, and thus minimise fluid administration in anaesthetised dogs.

Although all of these methods tested in this thesis have pros and cons in clinical veterinary practice, they were showen to be able to optimise haemodynamics without fluid overload in anaesthetised dogs.

ACKNOWLEDGEMENT

I have been fortunate to have many people assist me throughout this research project. Firstly, to my supervisors; Associate Professor Paul Chambers, Professor Craig Johnson, and Associate professor Nick Cave, the guidance, consultant, routine chat, phone calls, and assistance have been greatly appreciated. Working with excellent mentors has been an inspiration. I particularly thank you for the mentoring and friendship from Paul, who has always been supportive, throughout this journey, despite the challenging environment, including intensive clinical duties, teaching responsibility, administrative works and other ongoing research projects. I thank you, to Craig, for many chats and discussion about many research ideas. I thank you, to Nick for mentoring the writing scientific articles, and making study design.

I thank you to all anaesthesia nurses and intern at the teaching hospital, who helped with assisting my experiments and collecting clinical cases, even though they had a lot of other duties during my research project.

To my colleagues and friends, Drs Mike Gieseg, Paul Wightman, Janis Bridges, Paola Giordano, and Vicki Walsh, who have been supportive and offered advice throughout the part time PhD and work adventure, thank you for all your advice. To my dear friends Drs. Kerrie Lewis and Joana Chagas, who provided all supports throughout this adventure.

To my research collaborators, Drs. Yoshihiro Sugo and Mitsuyoshi Furukawa from Nihon Nihon Kohden, who provided a special multiparameter monitor for this project, thank you for all supports and unique ideas.

Finally, and most importantly, to my family, who without, this work would not have been possible. To my parents and grandfather, who without the lifelong support and advice, this would not be possible, we only wish my grandfather could be here to read this.

This PhD was partially funded by Massey University Research Funds. All necessary approvals were obtained from the Massey University Animal Ethics Committee.

LIST OF PUBLICATIONS

Sano H, Seo J, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P: Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anesthetized dogs. *Journal of Veterinary Emergency and Critical Care.* 28(4):301-309, 2018. doi: 10.1111/vec.12728.

Sano H & Chambers JP: Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Veterinary Anaesthesia and Analgesia.* 44(5):1057-1067, 2017. doi: 10.1016/j.vaa.2016.11.014.

Sano H, Fujiyama M, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P: Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. *Journal of Veterinary Emergency and Critical Care.* 29(4):391-398, 2019. doi: 10.1111/vec.12860.

Sano H & Chambers P: Investigation of change in pulse wave transit time following mini-fluid challenge with colloid predicting fluid responsiveness in spontaneously breathing anaesthetised dogs undergoing orthopaedic surgery. *13th World Congress of Veterinary Anaesthesiology in Venice, Italy.* Poster session, 131, September 2018. The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

Sano H, Chambers P, Bridges J, Johnson C & McGlade K: Effects of noradrenaline infusion prior to hypotension on anaesthetic management in dogs undergoing ovariohysterectomy. *American College of Veterinary Anesthesia and Analgesia Annual Conference in Washington D.C., USA*. Oral presentation, 111, September 2019. doi: 10.1016/j.vaa.2019.08.039. The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

TABLE OF CONTENTS

CHAPTER 5: INVESTIGATION OF CHANGE IN PULSE WAVE TRANSIT TIME FOLLOWING MINI-FLUID CHALLENGE PREDICTING FLUID RESPONSIVENESS IN SPONTANEOUSLY BREATHING ANAESTHETISED DOGS

Sano H & Chambers P: Investigation of change in pulse wave transit time following mini-fluid challenge with colloid predicting fluid responsiveness in spontaneously breathing anaesthetised dogs undergoing orthopaedic surgery. *13th World Congress of Veterinary Anaesthesiology in Venice, Italy.* Poster session, 131, September 2018. The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

Chapter 6: EFFECTS OF NORADRENALINE INFUSION PRIOR TO HYPOTENSION ON ANAESTHETIC MANAGEMENT IN DOGS UNDERGOING OVARIOHYSTERECTOMY

Sano H, Chambers P, Bridges J, Johnson C & McGlade K: Effects of noradrenaline infusion prior to hypotension on anaesthetic management in dogs undergoing ovariohysterectomy. *American College of Veterinary Anesthesia and Analgesia Annual Conference in Washington D.C., USA*. Oral presentation, 111, September 2019. doi: 10.1016/j.vaa.2019.08.039. The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

LIST OF ABBREVIATIONS

ASA	American Society of Anesthesiologists
ANOVA	analysis of variance
ASA	American Society of Anesthesiologists
AUC	area under a curve
AUROC	area under the receiver operating characteristic
BG	blood glucose (mmol/L)
CDA	clinical depth of anaesthesia
CI	cardiac index (mL/kg/minute)
CO	cardiac output (mL/minute)
CVP	central venous pressure (mmHg)
DO_2	oxygen delivery (mL/minute)
ECG	electrocardiogram
esCO	CO estimated from PWTT (mL)
$esCO_{IBP}$	CO estimated from PWTT and calibrated with IBP (L/minute)
$esCO_{NIBP}$	CO estimated from PWTT and calibrated with NIBP (L/minute)
esSV	SV estimated from PWTT (L/minute)
$F_E'Iso$	end-tidal isoflurane concentration (%)
FFC	full fluid challenge
f_R	respiratory rate (breaths/minute)
Hb	haemoglobin (g/dL)
HR	heart rate (beats/minute)
IBP	invasive blood pressure (mmHg)
IM	intramuscularly
IV	intravenously

MAP	mean arterial pressure (mmHg)
MFC	mini-fluid challenge
NIBP	non-invasive blood pressure (mmHg)
PCV	packed cell volume (%)
$P_E'CO_2$	end-tidal carbon dioxide tension (mmHg)
PI	perfusion index
PIP	peak inspiratory pressure (cmH_2O)
PPV	pulse pressure variation (%)
PVI	pleth variability index (%)
PWTT	pulse wave transit time (msecond)
ΔPWTT	percentage change in PWTT (%)
$\Delta PWTT_{1,2,3,10}$	ΔPWTT after 1, 2, 3, and 10 mL/kg colloid administration (%)
$\Delta PWTT_{FFC}$	percentage change in PWTT over FFC (%)
$\Delta PWTT_{MFC}$	percentage change in PWTT over MFC (%)
ROC	receiver operator characteristic
SD	standard deviation
SE	standard error
SV	stroke volume (mL)
SVV	stroke volume variation (%)
SVR	systemic vascular resistance (dynes/second/cm^5)
ΔSV	percentage change in SV (%)
T	temperature (°C)
TEE	transoesophageal echocardiography
TDCO	CO measured by thermodilution technique (L/minute)
TDSV	SV measured by thermodilution technique (mL)

TS	total solids (g/L)
VT	tidal volume (mL/kg)
VTI	velocity time integral (cm)
ΔVTI	percentage change in VTI (%)
$\Delta VTI_{1,2,3,10}$	ΔVTI after 1, 2, 3, and 10 mL/kg colloid administration (%)
ΔVTI_{FFC}	percentage change in VTI over FFC (%)
ΔVTI_{MFC}	percentage change in VTI over MFC (%)
95% CI	95% confidence interval
ΔPWTT	percentage change in PWTT (%)
$\Delta PWTT_{FFC}$	percentage change in PWTT over FFC (%)
$\Delta PWTT_{MFC}$	percentage change in PWTT over MFC (%)
$\Delta PWTT_{1,2,3,10}$	ΔPWTT after 1, 2, 3, and 10 mL/kg colloid administration (%)
ΔSV	percentage change in SV (%)
ΔVTI	percentage change in VTI (%)
ΔVTI_{FFC}	percentage change in VTI over FFC (%)
ΔVTI_{MFC}	percentage change in VTI over MFC (%)
$\Delta VTI_{1,2,3,10}$	ΔVTI after 1, 2, 3, and 10 mL/kg colloid administration (%)

LIST OF TABLES

Chapter 2

Chapter 3

Chapter 4

mL/kg) and the fluid challenge (10 mL/kg) in responders and non-responders. Data are mean (standard deviation). HR; heat rate, MAP; mean arterial pressure, CVP; central venous pressure, VTI; velocity time integral, PWTT; pulse wave transit time. Some variables are extracted from the concurrent study (**Chapter 2**)(Sano et al., 2018). $^{*}p <$ 0.05 between responders (n = 14) and non-responders (n = 10). $^{\dagger}p < 0.05$ between baseline and either 1, 2, 3 or 10 mL/kg.

Table 4.2 Comparison of $\Delta VTI_{1,2,3,10}$ and $\Delta PWTT_{1,2,3,10}$ after the mini-fluid challenges (1, 2 and 3 mL/kg) and the fluid challenge (10 mL/kg) in responders and non-responders. Data are mean (standard deviation). $\Delta VTI_{1,2,3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 1, 2, 3, and 10 mL/kg of fluid; $\Delta PWTT_{1,2,3,10}$, percentage change in pulse wave transit time after 1, 2, 3, and 10 mL/kg of fluid. Some variables are extracted from the concurrent study (**Chapter 2**)(Sano et al., 2018). $^{*}p < 0.05$ between responders (n = 14) and non-responders (n = 10).

Table 4.3 Areas under the receiver operator characteristic (ROC) curves and cutoff values for the prediction of fluid responsiveness. AUC: Area under the curve, 95%CI: 95% confidence interval, $\Delta VTI_{1,2,3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 1, 2, 3, and 10 mL/kg of fluid; $\Delta PWTT_{1,2,3,10}$, percentage change in pulse wave transit time after 1, 2, 3, and 10 mL/kg of fluid. $^{*}p <$ 0.05 between AUC and 0.5.

Chapter 5

Table 5.1 Characteristics of 45 dogs [responders (n = 19) and non-responders (n = 26)] before anaesthesia. Data are presented as mean ± standard deviation if the data were normally distributed or median (range) if not. $^{*}p<0.05$ between responders (n = 19) and non-responders (n = 26). f_R, respiratory rate; BUN, blood urea nitrogen; TTA, Tibial tuberosity advancement; TPLO, Tibial plateau leveling osteotomy; NA, non-applicable because BUN was 5-15 mg/dL for all dogs.

Table 5.2 Cardiorespiratory variables before and after the mini-fluid challenges (MFC: 3 mL/kg) and the full fluid challenges (FFC: 6 mL/kg) in responders and non-responders. Data are presented as median (interquartile range). $*p < 0.017$ compared to baseline using Wilcoxon test with Bonferroni adjustment. $^{\dagger}p < 0.05$ between responders (n = 19) and

non-responders (n = 26). HR, heart rate; MAP, mean arterial pressure; VTI, velocity time integral; PWTT, pulse wave transit time; f_R, respiratory rate; $P_E'CO_2$, partial pressure of end-tidal carbon dioxide; F_E'Iso, end-tidal isoflurane concentration.

Table 5.3 Comparison of percentage changes in velocity time integral (VTI) and pulse wave transit time (PWTT) over the mini-fluid challenges (MFC: 3 mL/kg) and the full fluid challenges (FFC: 6 mL/kg) in responders and non-responders. Data are presented as mean ± standard deviation. $^{*}p < 0.05$ between responders (n = 19) and non-responders (n = 26). ΔHR_{MFC}, ΔMAP_{MFC}, ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage change in heart rate, mean arterial pressure, VTI and PWTT over MFC; ΔHR_{FFC}, ΔMAP_{FFC}, ΔVTI_{FFC} and $\Delta PWTT_{FFC}$, percentage change in heart rate, mean arterial pressure, VTI and PWTT over FFC; NA, non-applicable because the definition of the responders was 15% > ΔVTI_{FFC}.

Table 5.4 Ability of percentage change in velocity time integral (ΔVTI_{MFC}) and pulse wave transit time ($\Delta PWTT_{MFC}$) over the mini-fluid challenge to predict increase in stroke volume of 15%. AUROC, area under the receiver operating characteristic curve; 95%CI, 95% confidence interval, Sen; sensitivity, Spe; specificity.

Chapter 6

Table 6.1 Preoperative characteristics of dogs between noradrenaline and control groups. Breed is expressed as breed name (number of dogs). Other values are medians (range). BCS, body condition score; HR, heart rate; f_R, respiratory rate; T, temperature; PCV, packed cell volume; TS, total solids; BG, blood glucose; BUN, blood urea nitrogen. BUN was 5-15 mg/dL for all dogs.

Table 6.2 Intraoperative values from dogs undergoing ovariohysterectomy between noradrenaline and control groups. Values are medians (range). HR, heart rate; MAP, mean arterial pressure; CI, cardiac index; f_R, respiratory rate; $P_E'CO_2$, partial pressure of end-tidal carbon dioxide; F_E'Iso, End-tidal isoflurane concentration; CDA, Clinical depth of anaesthesia; T, temperature.

LIST OF FIGURES

Chapter 1

maximum pulse pressure after a positive pressure breath; PP_{Min}, minimum pulse pressure after a positive pressure breath (from Scott et al. 2001).

Figure 1.7 Pleth Variability Index (PVI) PVI, 100 × (PI_{max} - PI_{min})/PI_{max}; PI_{max}, Maximum perfusion index; PI_{min}, Minimum PI (from Masimo HP; https://www.masimo.com/pvi/)

Figure 1.8 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

Figure 1.9 Pulse Wave Transit Time (PWTT). PWTT was calculated as the time from the ECG R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

Figure 1.10 Cerebral blood flow autoregulation (Paulson et al., 1990). It typically operates between MAP of the order of 60 and 150 mmHg (normal MAP).

Figure 1.11 Renal perfusion flow autoregulation (Shipley & Study, 1951). Renal perfusion flow (RPF) drops off at 70 mmHg but the plateau really starts at about 110 mmHg. Glomerular filtration (GFR) plateaus at 120 mmHg and by the time renal artery mean blood pressure is 70 mmHg the GFR is at about 70%.

Chapter 2

Figure 2.1 The Frank–Starling curve of the heart (A), definition of Pulse pressure variation (PPV) (B) and Pleth variability index (PVI) (C). In mechanically ventilated patients, stroke volume (SV) can change greater (a) in response to the variation of preload (b) caused by positive-pressure ventilation (Responders). In contrast, an increase in SV is relatively insensitive (c) to preload changes (d) on the plateau portion of the curve

(Non-responders). PP, pulse pressure (systolic arterial pressure - diastolic arterial pressure); Average PP, (PPmax + PPmin)/2; PI, perfusion index; max, maximum; min, minimum.

Figure 2.2 Comparison of areas under the receiver operator characteristic (ROC) curve for CVP, PPV and PVI. CVP, central venous pressure; PPV, pulse pressure variation; PVI, pleth variability index.

Chapter 3

Figure 3.1 Pulse wave transit time (PWTT). PWTT was calculated as the time from the electrocardiogram R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

Figure 3.2 Timeline of the study. After the calibration, 10 measurements were taken; before drug administration (baseline1: BL1), during phenylephrine administration (0.5 μg/kg/minute: PH1; 1 μg/kg/minute: PH2), 30 minutes after cessation of PH1 (baseline2: BL2), during high isoflurane administration (2.0-2.5%: ISO1; 2.5-3.0%: ISO2), 30 minutes after cessation of ISO2 (baseline3: BL3), during dobutamine administration (1 μg/kg/minute: DO1; 2 μg/kg/minute: DO2) and 30 minutes after cessation of DO2 (baseline4: BL4).

Figure 3.3 Area under the receiver operator characteristic (ROC) curve for the percentage change in pulse wave transit time (PWTT) to detect a 15% change in stroke volume derived from the thermodilution technique (TDSV). The closer to 1.0 of an area under the ROC curve indicates, the more reliable diagnostic method. 95% CI, 95% confidence interval. The area under the ROC curve and the cut-off value of the percentage change in PWTT to detect 15% change in stroke volume were 0.91 (95% CI, 0.85e0.98; $p < 0.001$) and 2.7% (sensitivity: 86%; specificity: 81%), respectively.

Figure 3.4 Four-quadrant plot with linear regression analysis between percentage changes in PWTT and TDSV. The dotted lines limit an exclusion zone of ±15% for TDSV and 2.7% for PWTT. PWTT, pulse wave transit time; TDSV, stroke volume measured by

the thermodilution technique.

Figure 3.5 Bland-Altman plots for the difference between the methods plotted against their mean. (A) TDCO *versus* esCO with NIBP calibration, (B) TDCO *versus* esCO with IBP calibration. The solid line represents the mean bias, the two broken lines indicate the mean bias ± 1.96 standard deviation; the dotted line is the line of equality. esCO, estimated cardiac output based on pulse wave transit time; IBP, invasive blood pressure; NIBP, non-invasive blood pressure; SD, standard deviation; TDCO, cardiac output measured by the thermodilution technique.

Chapter 4

Figure 4.1 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

Figure 4.2 Correlation between $\Delta PWTT_3$ and ΔVTI_{10}, ΔVTI_3, and **$\Delta PWTT_{10}$ and ΔVTI_{10}** $\Delta VTI_{3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 3, and 10 mL/kg of fluid; $\Delta PWTT_{3,10}$, percentage change in pulse wave transit time after 3, and 10 mL/kg of fluid. $^{*}p < 0.05$

Figure 4.3 Receiver operator characteristic (ROC) curve for ΔVTI_3 and $\Delta PWTT_3$. ΔVTI_3 and $\Delta PWTT_3$, percentage changes in velocity time integral of main pulmonary arterial blood flow and pulse wave transit time after a 3 mL/kg mini-fluid challenge; AUC, area under the curve; 95%CI, 95% confidence interval.

Figure 4.3 The grey zone and the best cutoff value within the optimal cutoff values for $\Delta PWTT_3$. Distribution of the cutoffs for each bootstrapped population (1000 "optimal" values). Grey rectangle (the grey zone), 95% confidence interval for the optimal cutoffs; Black line (the best cutoff value), mean; $\Delta PWTT_3$, percentage changes in pulse wave transit time after a 3 mL/kg mini-fluid challenge.

Chapter 5

Figure 5.1 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

Figure 5.2 Pulse Wave Transit Time (PWTT). PWTT was calculated as the time from the ECG R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

Figure 5.3 Receiver operator characteristic curve for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$. ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage changes in velocity time integral of aortic blood flow and pulse wave transit time after a 3 mL/kg mini-fluid challenge.

Figure 5.4 The grey zone and the best cutoff value within the optimal values for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$. Distribution of the cutoffs for each bootstrapped population (1000 "optimal" values). Grey rectangle, grey zone; Black line (the best cutoff value); ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage changes in velocity time integral of aortic blood flow and pulse wave transit time after a 3 mL/kg of mini-fluid challenge.

Chapter 6

Figure 6.1 Mean ± SE haemodynamic parameters over the time points. T0, 5 minutes after the induction; T1, 10 minutes after the infusion started; T2 ,before start of the surgery; T3 ,time of skin incision; T4 ,time of removal of the first ovary; T5 ,time of removal of the second ovary; T6 ,time of closure of the abdominal wall; T7 ,immediately after the skin was sutured. Overall, mean arterial pressure was significantly greater ($p < 0.01$) and heart rate was significantly lower ($p < 0.01$) in the noradrenaline group than those in the control group, while cardiac index was no significant between groups ($p = 0.12$).

Figure 6.2 Mean ± SE end-tidal isoflurane concentration (Left) and frequency of clinical

depth of anaesthesia (CDA) scores (Right) over the time points. T0, 5 minutes after the induction; T1, 10 minutes after the infusion started; T2 ,before start of the surgery; T3 ,time of skin incision; T4 ,time of removal of the first ovary; T5 ,time of removal of the second ovary; T6 ,time of closure of the abdominal wall; T7 ,immediately after the skin was sutured. CDA scores of 1, 2 and 3 indicated light anaesthesia, adequate surgical anaesthesia and an excessive depth of anaesthesia, respectively. C, control group; N. norepinephrine group. End-tidal isoflurane concentration in the noradrenaline group was significantly higher than that in the control group ($p < 0.01$) although to account for repeated measures in the CDA data, there was no effect of treatment with a cumulative link mixed model ($p = 0.66$).

Figure 6.3 Mean ± SE haematology parameters before and after the surgery. Blood samples were taken from the arterial catheter immediately after the catheter was placed (Pre) and immediately after the end of anaesthesia (Post). Packed cell volume ($p < 0.001$), total solids ($p < 0.001$) and blood glucose ($p = 0.023$) were significantly lower in the control group than those in the noradrenaline group after the surgery, while there was no effect of treatment on lactate ($p = 0.116$) and creatinine between groups ($p = 0.06$).

Chapter 7

Figure 7.1 Clinical interpretation of haemodynamics. MAP can be calculated by CO and SVR, and CO can be calculated by HR and SV. SV is determined by preload, contractility and afterload. MAP and HR is only parameters that can be obtained clinically, which are not enough to comprehend haemodynamics. MAP; mean arterial pressure, CO; cardiac output, SVR; systemic vascular resistance, HR; heart rate, SV; stroke volume.

INTRODUCTION

THESIS STRUCTURE

The studies presented in this thesis are in form of manuscripts published in peer-reviewed journals and presented at major anaesthesia conferences, and formatted for the style of the journal they were published in. Consequently, there is some repetition of background information and methods in some of the chapters, and also the units (SI or American) depends on the style of the journal. All manuscripts have been standardised to one referencing style throughout the thesis. References are included at the end of each chapter. Throughout **Chapter 1-6**, figures and tables are labelled as Figure 1. Table 1 etc, in line with the published manuscript. As such, there are multiple Figure 1s etc throughout the thesis, however, where required, each figure is clearly identified as **Chapter 2, Figure 2.1** etc.

THESIS OUTLINE

Chapter 1 of the thesis is a literature review, which provides the reader with a brief overview of perioperative fluid therapy and cardiac function with fluid loading, then moves on to assessment of fluid responsiveness. An overview of measurement of cardiac output is covered, as well as perioperative haemodynamic management in dogs. The literature review reveals that there are many questions still unanswered in regard to perioperative fluid administration to optimise haemodynamics without fluid overload in anaesthetised dogs.

The aim of the study described in **Chapter 2** is to investigate whether dynamic indices (pulse pressure variation and pleth variability index) could predict fluid responsiveness more accurately than classical static index (central venous pressure) in dogs. These dynamic indices have been proved to be reliable predictors for fluid responsiveness in people but have not been investigated in dogs at the time of the study. However, the use of these in clinical veterinary practice is impractical for various reasons.

Chapter 3 evaluated the feasibility of pulse wave transit time as a surrogate of stroke volume in dogs. Particularly, the study focused on the ability of change in pulse wave transit time to detect changes in actual stroke volume. Accurate measurement of change in stroke volume is necessary for an alternative technique to assess fluid responsiveness, the mini-fluid challenge studied in **Chapter 4 and 5**.

Chapter 4 and 5 investigated whether a percentage change in pulse wave transit time (a surrogate parameter of stroke volume) following mini-fluid challenge could predict fluid responsiveness in mechanically ventilated anaesthetised dogs under experimental conditions (**Chapter 4**), and spontaneously breathing anaesthetised dogs undergoing

stifle surgery in clinical setting (**Chapter 5**).

Chapter 6 investigated whether prophylactic noradrenaline administration would be able to prevent hypotension, and thus minimise the requirement for fluid administration in anaesthetised dogs. This study was performed because all methods tested in previous chapters are still of limited use in veterinary clinical practice due to availability of equipment, difficulty of their interpretation, and a cumbersome process.

To conclude, in the final chapter of this thesis, a discussion of the key findings of this research is provided, including future direction.

Chapter 1

Literature review

---

1. OVERVIEW OF PERIOPERATIVE FLUID THERAPY

1.1 Introduction

Fluid therapy has had an important role of haemodynamic management since successful treatment of cholera patients with intravenous fluid was first reported by O'Shaughnessy and Latta in the 1830s (Latta, 1832; O'Shaughnessy, 1831). Fluid administration compensated for the luminal losses, and corrected the circulation deficit. In the 1880s, fluid therapy was established as a treatment of haemorrhage and shock, after it was shown to improve the circulation of a patient suffering from antepartum haemorrhage (Egerton Jennings, 1882; Thomas, 1898). Perioperative continuous fluid administration was described in 1924 as restoring the ongoing fluid loss, and supporting the circulation in surgical patients (Matas, 1924), and it was found to prevent pre-renal injury, resulting in improved morbidity and mortality (Coller, Dick, & Maddock, 1936). Therefore, the efficacy of perioperative fluid therapy to support patient care has been established for many years.

In the late 1900s, the concept of supranormal fluid resuscitation was proposed by Shoemaker et al (Shoemaker, Appel, Kram, Waxman, & Lee, 1988). They observed that

survivors from shock who received fluid resuscitation had supranormal levels of oxygen delivery and cardiac output compared to non-survivors who did not. The concept that early supranormal fluid resuscitation improved the outcome was supported by other (Bishop et al., 1995; Bishop et al., 1993). However, several prospective randomised clinical studies showed conflicting results (Boyd, Grounds, & Bennett, 1993; Durham, Neunaber, Mazuski, Shapiro, & Baue, 1996; Gattinoni et al., 1995). In addition, although Shoemaker et al. showed a benefit of fluid therapy in resuscitating patients with shock, they also concluded that fluid therapy was also a contributing factor to the outcome of critically injured patients. Patients who could achieve supranormal haemodynamic values were more likely to survive than those who could not, regardless of the resuscitation technique (Velmahos et al., 2000). Therefore, the need for aggressive fluid therapy to achieve supranormal resuscitation has been reconsidered.

Nowadays, perioperative fluid therapy is commonly used to provide essential fluid for maintenance of body functions, and replace fluid losses due to co-existing diseases, haemorrhage, evaporation from surgical sites, and accumulation in an unknown third space. Perioperative fluid is routinely administered to achieve supranormal haemodynamic optimisation. However, more recently, the focus has been focused on perioperative fluid restriction compared to liberal fluid therapy. Lobo et al. demonstrated that restricting perioperative intravenous fluid to less than traditionally administered, increased gastric emptying and decreased complications and length of hospital stay in 20 patients undergoing elective colonic resection (Lobo et al., 2002), compared to patients given liberal fluid therapy. Prospective randomised trials and a meta-analysis showed the benefits of fluid restriction and avoidance of fluid overload in improving clinical outcomes in different clinical settings (Brandstrup et al., 2003; McArdle et al., 2009;

Nisanevich et al., 2005; Rahbari et al., 2009). Although the aim of fluid administration is to support adequate tissue perfusion, excessive fluid administration caused tissue oedema (Holte, Jensen, & Kehlet, 2003), and impaired oxygen and nutrient delivery to tissues (Cotton, Guy, Morris, & Abumrad, 2006).

Therefore, fluid imbalance, a term to describe either too much or too little fluid, should be avoided, which is challenging in clinical practice.

1.2 Traditional perioperative fluid therapy

The purpose of perioperative fluid therapy is to maintain sufficient circulating volume to ensure perfusion of the organs and oxygen delivery to the tissues. Patients might become hypovolaemic during anaesthesia and surgery because of fasting overnight, inability to drink water due to the stressful environment such as a hospital ward, ongoing losses from unexpected urinary output due to underlying diseases, blood loss, and evaporation from surgical sites or expired gas. Thus, large volumes of crystalloid were traditionally administered to replace the fluid deficit during anaesthesia.

A fluid bolus is also commonly administered when hypotension induced by general anaesthetic drugs occurs. General anaesthesia causes vasodilatation, leading to relative hypovolaemia (Noel-Morgan & Muir, 2018).Thus, since cardiac output is dependent on filling pressure, it is considered rational to give fluids to correct the relative hypovolaemia and increase filling pressure. However, it has been shown that fluid loading has little or no influence on anaesthesia-related hypotension in people (Norberg et al., 2007) and dogs (Valverde, Gianotti, Rioja-Garcia, & Hathway, 2012). This is probably due to the negative inotropic effect of anaesthesia, which prevents an increase in cardiac output in response

to an increased filling pressure. Thus, hypotension induced by anaesthesia may be more appropriately treated using inotropic and vasopressor drugs, instead of a large volume of fluid, which may case patients to become hypervolaemic.

Furthermore, it was found that during major surgical procedures, patients experience a marked loss of extravascular fluid regardless of blood loss. Following from that, an implausible hypothesis was posted in the 1960s (Shires, Williams, & Brown, 1961), which was that body cavity exposure during surgery induced an internal redistribution of extracellular fluid to a "third space" resulting in a reduction in the circulating volume. This third space into which fluid disappeared was hypothesised to be in traumatised tissue or the gastrointestinal tract. To compensate for the loss of circulating volume due to this hypothetical third space (Jacob, Chappell, & Rehm, 2009), large volumes of crystalloid fluids were historically administered perioperatively. However, with the use of methods to measure extracellular volume with radioactive tracers, recent trials found an unchanged or even increased extracellular volume during surgery, the opposite of the previous third space concept. Thus, traditional large volumes of fluid therapy cause fluid overload, which may result in haemodilution (including dilution of clotting factors), interstitial oedema and thus impaired oxygen and nutrition delivery. Indeed, excessive fluid resuscitation has been reported to cause oedema, with increased morbidity (Arieff, 1999), impaired coagulation (Ruttmann, James, & Aronson, 1998), bacterial translocation and sepsis (Ratner, Lysenko, Paul, & Weiser, 2005) and poor wound healing (Lang, Boldt, Suttner, & Haisch, 2001). In addition, the increase in body weight from excess fluid has been shown to be correlated with postoperative morbidity and mortality (Holte et al., 2003; Lowell, Schifferdecker, Driscoll, Benotti, & Bistrian, 1990). Subsequently, a systematic review published in 2006 concluded that the concept of third space was

fundamentally incorrect (Brandstrup, Svensen, & Engquist, 2006). Therefore, restrictive perioperative fluid therapy started to be investigated.

1.3 Restrictive perioperative fluid therapy

Early randomised trials supported positive benefits with restrictive fluid therapy in abdominal surgery, with a faster return of gastrointestinal function, fewer complications and shorter hospital stay compared to the traditional, or liberal fluid therapy (Brandstrup et al., 2003; Lobo et al., 2002; Nisanevich et al., 2005). For example, in a randomised, multicentre trial comparing traditional and restrictive fluid groups in 141 patients undergoing colorectal surgery, patients in the restrictive group had a lower complication rate (anastomotic leakage, wound infection, and cardiovascular and pulmonary compromises) in the restrictive group compared to the liberal group (33% vs 51%, p = 0.02) (Brandstrup et al., 2003). However, there were also studies that have shown no benefit from a restrictive fluid protocol compared with the traditional protocol (MacKay et al., 2006; Vermeulen, Hofland, Legemate, & Ubbink, 2009). A review of postoperative outcome from restrictive *vs* traditional fluid therapy in seven randomised trials (six involving major abdominal surgery and one involving knee arthroplasty), revealed that three studies found an improved outcome (faster return of gastrointestinal function and reduced hospital length of stay) with a restrictive fluid regimen, whereas two studies found no difference, and two studies found that although the total number of complications was reduced, the number of patients with complications was not significantly reduced (Bundgaard-Nielsen, Secher, & Kehlet, 2009). However, it is difficult to form a holistic interpretation of the studies, because there was variation in the definition of liberal or restrictive protocols in clinical practice, whereby a restrictive regime in one centre was actually liberal in another. The studies also varied in design,

types of fluid administered, indications for administering additional fluid, outcomes variables, and definitions of intra- and postoperative periods (Corcoran, Rhodes, Clarke, Myles, & Ho, 2012; P. S. Myles et al., 2018; Rahbari et al., 2009). Eventually, the conclusion was that optimal fluid therapy requires the assessment of each individual's haemodynamic status, to administer what is called "individualised goal-directed fluid therapy" (Chong, Wang, Berbenetz, & McConachie, 2018; Xu et al., 2018).

Outcomes may be improved if fluid therapy is individualised based on objective feedback on the patient's individual fluid responsiveness. The "fluid responsiveness" is derived from the physiological principal of the Frank-Starling curve and determined by dynamic predictors, described later in this Chapter (see Section 3. ASSESSMENT OF FLUID RESPONSIVENESS in Chapter 1"). A meta-analysis of ninety-five randomised trails concluded that individualised goal-directed fluid therapy using dynamic predictors reduced postoperative morbidity and mortality in adult surgical patients compared to conventional fluid therapy although the quality of evidence was low (Chong et al., 2018). Another meta-analysis of eleven randomised studies showed that gastrointestinal function improved with goal-directed fluid therapy compared with conventional fluid therapy in patients undergoing colorectal surgery (Xu et al., 2018). Both meta-analyses concluded that individualised goal-directed fluid therapy reduced perioperative fluid administration compared to historical fluid therapy. Thus, more recent expert guideline/consensus statements/meta-analysis on perioperative fluid therapy have supported more restrictive fluid regimens (Jia et al., 2017; Lassen et al., 2009; P. Myles et al., 2017; Schol, Terink, Lance, & Scheepers, 2016; Self et al., 2018). Individualised goal-directed fluid therapy based on fluid responsiveness derived from the Frank-Starling curve has yet to be widely accepted in veterinary practice, but it is argued that it should be, because there is no reason

to believe there are fundamental species differences in this respect, and both hypovolaemia and hypervolaemia are known to cause increased perioperative morbidity and mortality (Doherty & Buggy, 2012; Navarro et al., 2015) (Figure 1.1).

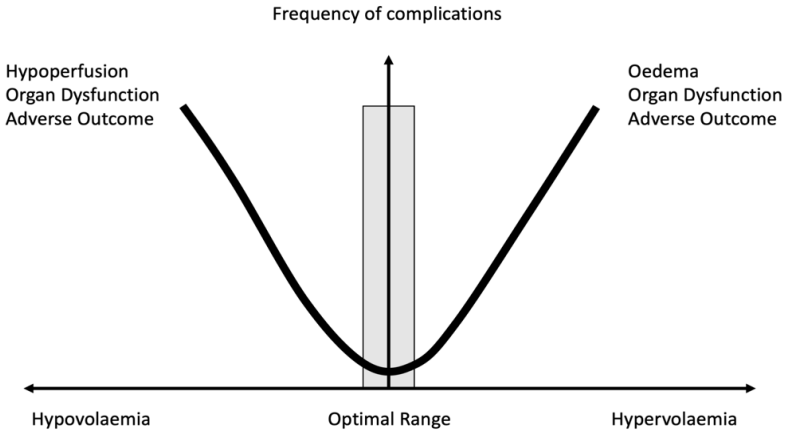

Figure 1 Avoidance of both hypo- and hypervolaemia is the aim of intraoperative fluid therapy in order to prevent adverse outcomes. Modified from Doherty & Buggy (Doherty & Buggy, 2012)

1.4 Fluid Therapy Guidelines for Dogs and Cats

The basic concept of fluid therapy for dogs may be extrapolated from the human literature because physiology of mammals is similar. American Animal Hospital Association (AAHA)/American Association of Feline Practitioners (AAFP) published fluid therapy guidelines (Davis et al., 2013), anaesthesia guidelines (Bednarski et al., 2011) and monitoring guidelines (Grubb et al., 2020) including perioperative fluid therapy for dogs. These guidelines are recommendations from AAHA/ AAFP based on expert opinion rather than controlled trials. The guidelines recommend therapy that is tailored to each

patient and constantly re-evaluated and reformulated according to changes in status. During the perioperative period, the proposed benefits of fluid therapy include replacement of normal–ongoing fluid losses and fluid losses related to surgery, and support of cardiovascular function based on the Frank-Starling curve. Current recommendations are to deliver < 10 mL/kg/hr to avoid adverse effects associated with hypervolaemia, particularly in cats due to their smaller blood volume to weight ratio (Breznock & Strack, 1982; D. C. Brodbelt, Pfeiffer, Young, & Wood, 2007; Tang, Wu, & Peng, 2011). In the absence of evidence-based perioperative fluid rates for animals, the guidelines suggest initially starting at 3 mL/kg/hour in cats and 5 mL/kg/hour in dogs, based on clinical experience.

The primary risk of providing excessive intravenous fluids in healthy patients is of vascular overload, which may actually lead to worsened outcomes, including lung water; decreased pulmonary function; coagulation deficits; reduced gut motility; reduced tissue oxygenation; increased infection rate; and positive fluid balance, with decreases in packed cell volume, total protein concentration, and body temperature (Chappell, Jacob, Hofmann-Kiefer, Conzen, & Rehm, 2008; Voldby & Brandstrup, 2016). Supranormal fluid resuscitation with 10-30 mL/kg/hour of crystalloid to isoflurane-anaesthetised dogs did not improve either urine production or oxygen delivery to tissues (calculated from cardiac output measured by thermodilution and oxygen content derived from arterial blood gas analysis) , as estimated using cardiac output measured by thermodilution, and oxygen content derived from arterial blood gas analysis (Muir, Kijtawornrat, Ueyama, Radecki, & Hamlin, 2011).

2. CARDIAC FUNCTION AND FLUID LOADING

2.1 The heart functions

The main function of the heart is to distribute sufficient oxygenated blood to the entire body. First, deoxygenated venous blood returns through the vena cavae to the right atrium which then pushes the blood into the right ventricle. Second, the right ventricle pumps the blood through the pulmonary artery and then the pulmonary capillaries in the lungs where gas exchange takes place. Third, the oxygenated blood moves from the lungs through the pulmonary veins to the left atrium, which pushes the blood into the left ventricle. Forth, the left ventricle pumps oxygenated blood through the aorta to the organs and peripheral tissues. The volume of blood that is pumped from the heart on each contraction cycle is called the stroke volume (SV). Cardiac output (CO) is defined as the volume of blood that the heart pumps to the systemic circulation per minute, and is dependent on heart rate (HR) and SV (CO = HR × SV). SV is influenced by preload, contractility, and afterload independently.

The preload is the amount of myocardial stretch prior to each contraction, and clinically represents the blood volume in the ventricle or the venous return at the end of diastole. For example, haemorrhage decreases the preload, leading to reduced myocardial stretch and reduced SV, while fluid administration increases the preload, leading to wide myocardial stretch and increased SV (as described by the Frank–Starling law of the heart, see below).

Contractility is the intrinsic contractile function of the ventricle and can be defined as the force generated by the myocardium independent of preload and afterload. Sympathetic nervous stimulation and inotropic drugs such as beta-adrenergic agonists increase the

contractility, while anaesthetic agents, severe acidosis, hypoxia or hypoxaemia decrease the contractility of the myocardium.

Finally, afterload is the pressure that the heart must work against to eject blood during systole. Clinically, the afterload is equivalent to the amount of vasoconstriction or vascular impedance against which the ventricle ejects. Vasoconstrictors such as alpha-adrenergic agonists increase the afterload, tending to reduce the SV, while vasodilators reduce the afterload, tending to increase the SV.

2.2 The Frank–Starling law of the heart

Under most conditions, CO is determined by the venous return, which is the rate of blood flow into the heart through the vena cavae. A century ago, Dr. Otto Frank and Ernest Starling demonstrated increased ventricular contraction when the ventricle was stretched prior to contraction. Thus, an increase in ventricular filling pressure, which can be achieved by increasing venous return, augmented SV and CO. This is now called the Frank–Starling law of the heart. The well-known Frank-Starling curve depicts changes in SV or CO in response to changes in venous return, preload, or right atrial pressure (Figure 1.2-a). An increase in venous return increases the right ventricular filling pressure. Increased right ventricular filling pressure stretches sarcomere length in myocytes and increasing the sarcomere length increases troponin C calcium sensitivity, which increases the rate of cross-bridge attachment and detachment, and the amount of tension developed by the muscle fibre, and then augments contraction because the actin and myosin filaments are brought to a more nearly optimal degree of overlap for force generation, and this subsequently increases SV and thus CO (Guyton & Hall, 2015). Moreover, recent muscle physiology research identified a third filament system composed of the giant

elastic protein titin, which is responsible for most passive stiffness in the physiological sarcomere length range. A significant coupling of active and passive forces in cardiac muscle, where titin-based passive force promotes cross-bridge recruitment, resulted in greater active force production in response to stretch. This focus has been placed on the troponin-based "on-off" switching of the thin filament state in the regulation of length-dependent activation (Fukuda, Terui, Ohtsuki, Ishiwata, & Kurihara, 2009). The Frank-Starling curve is also influenced by contractility and afterload, and therefore reflects the ventricular function. Figure 1.2-b depicts the curve when the contractility is increased or the afterload is reduced, while the curve of Figure 1.2-c depicts the curve when the contractility is decreased, or the afterload is increased. If contractility and afterload remain constant, the preload determines SV and CO. Thus, when fluid loading increases venous return and preload, it will result in an increased SV and CO. Indeed, SV and CO cannot increase without an increase in venous return; clarified by the Frank- Starling principle that 'the heart pumps what it receives'. However, excessive preload overstretches the myocyte, resulting in unchanged or decreased SV. Although increasing preload by fluid loading will increase SV and CO when the patient is on the ascending portion of the curve, fluid loading will have little effect when the patient is near the flat portion (Guyton, 1955). Thus, if preload is increased when the heart is unable to pump the excess volume, it will lead to increased venous pressure and oedema.

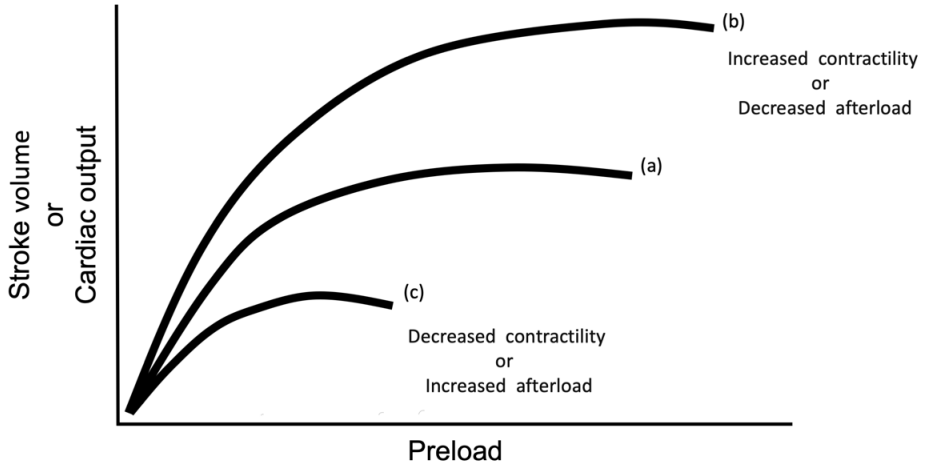

Figure 1.2 Frank-Starling curve. An increase in preload increase in stroke volume and cardiac output. Figure b showed the curve when the contractility is increased or the afterload is reduced, while the curve of Figure c is depicted when the contractility is decreased, or the afterload is increased. However, the excessive preload overstretches the myocyte, resulting in decreased or unchanged stroke volume.

2.3 Fluid responsiveness

The main purpose of fluid administration is to prevent or treat organ hypoperfusion, particularly brain, myocardium, and kidneys, while simultaneously avoiding excessive fluid administration. If fluid administration increases SV or CO, then tissue perfusion improves, but if the heart is unable to increase SV or CO in response, then fluid loading will cause congestion and oedema in the lungs and other tissues. Of critical importance, however, is that it is clinically challenging to identify whether tissue hypoperfusion exists, or predict if fluid administration will be effective, as subclinical signs of hypoperfusion can easily be missed. Thus, it is typical in clinical practice to administer intravenous fluids in ignorance of where the haemodynamic status of a given patient is on the Frank-Starling curve. Perhaps unsurprisingly then, only roughly half of haemodynamically unstable patients respond to a fluid administration, defined as an increase in SV or CO

upon fluid loading (Marik, Cavallazzi, Vasu, & Hirani, 2009; Michard & Teboul, 2002).

When tissue hypoperfusion is deemed likely, it is essential to find out a patient's position on the Frank-Starling curve to predict whether an increase in SV and CO is to be expected from fluid administration. When a patient is located on the steep portion of the Frank-Starling curve, it is called a fluid "Responder" because increased SV and thus CO is expected after the fluid loading, while a fluid "Non-responder" is on the flat portion of the curve because SV and CO will remain unchanged following fluid administration (Figure 1.3). Fluid administration in a non-responder will cause a rise in cardiac filling pressures and thus hydrostatic pressures in pulmonary and venous circulation causing pulmonary and systemic oedema. Thus, a perioperative positive fluid balance was associated with increased postoperative mortality or acute kidney injury in intensive care unit patients (Oh, Song, Do, Jheon, & Lim, 2019). Conversely, if fluid administration is restricted in a patient that would be responsive, it is more likely that the unresolved hypotension will be treated with vasopressor therapy. In those under-treated potential responders, the vasopressor therapy may impair critical organ perfusion, threaten local tissues oxygenation, and cause cardiac arrhythmias which will further reduce CO (Fischer et al., 2013; Plurad et al., 2011). This is especially important since supranormal CO and oxygen delivery achieved using only an inotropic drugs failed to improve postoperative outcomes in critically ill patients (Hayes et al., 1994). Therefore, methods for predicting fluid responsiveness are required in order to identify those patients who will benefit from fluid loading and avoid ineffective and potentially detrimental fluid administration to non-responders where positive inotropic drugs may more effective and safer.

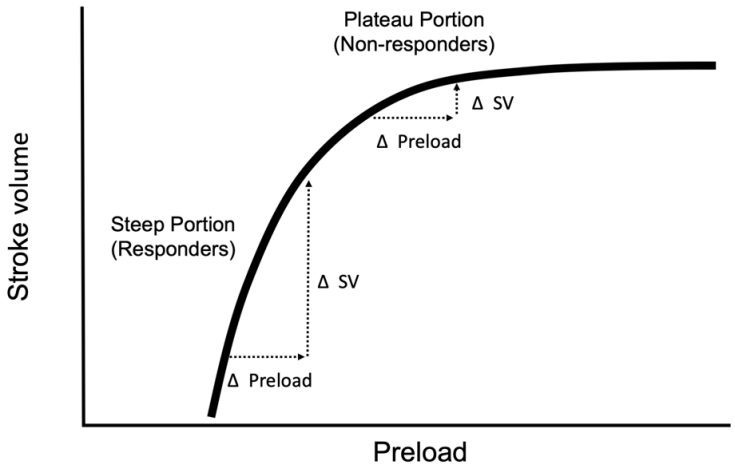

Figure 1.3 Fluid responder and non-responder. On the steep portion of the curve, changes in preload will result in large changes in stroke volume. A patient on the steep portion of the curve would have an increased stroke volume in response to fluid loading. Thus, this patient is a fluid responder. On the plateau portion of the curve, identical changes in preload will not alter stroke volume as much as they would for a patient on the steep portion of the curve. Hence, this patient is a fluid non-responder.

3. ASSESSMENT OF FLUID RESPONSIVENESS

3.1 Static predictors of fluid responsiveness

As mentioned, recent evidence has revealed that outcomes such as pneumonia, acute kidney injury, wound infection and hospital length of stay, are improved if fluid therapy is individualised based on the patient's fluid responsiveness (Chong et al., 2018). Medical history, clinical examination findings such as skin turgor, blood pressure, pulse rate, and urine output, and routine laboratory tests, are important but of limited sensitivity and specificity to predict fluid responsiveness (Michard & Teboul, 2002; Vincent & Weil, 2006). Central venous pressure (CVP) has traditionally been used as an indirect measure of venous return or preload in critically ill patients (De Backer & Vincent, 2018). It was believed that CVP is always proportional to preload, and thus a patient with low CVP will be a fluid responder, and a patient with high CVP will be a fluid non-responder. However, many studies have shown that neither static filling pressures nor the rate of change of static pressures following fluid administration was accurate in predicting fluid responsiveness (Magder, 2010; Marik, Baram, & Vahid, 2008). Filling pressures, such as CVP, are intramural pressures, and are dependent on atrial and ventricular compliance, which varies widely between patients, especially when critically ill (Magder, 2015), and are influenced by venous flow. Therefore, CVP is unable to function as a reliable indicator, neither of preload nor of fluid responsiveness (Marik et al., 2008). Thus, there is interest in developing more reliable predictors of fluid responsiveness.

3.2 Dynamic predictors of fluid responsiveness

3.2.1 Stroke volume variation

Over the last decade, dynamic parameters of fluid responsiveness have been described in anaesthetised patients that use stroke volume variations (SVV) induced by mechanical

ventilation. Positive pressure mechanical ventilation transiently compresses the vena cavae in the chest, which decreases the venous flow and preload, which in turn reduces SV (Mutz et al., 1984; Robotham et al., 1983). The degree to which SV decreases during positive pressure ventilation is proportional to the slope of the Frank-Starling curve, in the same way that fluid administration may or may not increase SV (Figure 1.4). On the steep portion of the curve, a given reduction in preload during insufflation will result in a larger SVV than in a patient on the shallower portion of the curve. Thus, the SVV can be used to predict if the patient is a fluid responder.

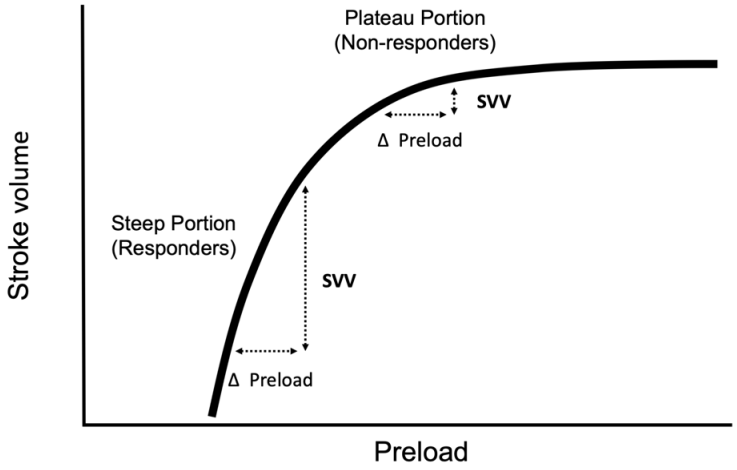

Figure 1.4 Stroke volume variation (SVV) A patient on the steep portion of the curve would have an increased SV in response to fluid loading. Thus, the patient with large SVV is a fluid responder. On the plateau portion of the curve, same changes in preload will not alter SV as much as they would for a patient on the steep portion of the curve. Hence, the patient with small SVV is a fluid non-responder.

In figure 1.5, patients A and B have different curves because of different contractility and

afterload. Given the same change in absolute preload, patient A has a greater change in SV (and greater SVV) than does patient B. So, patient A is a responder even though absolute changes in preload are identical for these two patients.

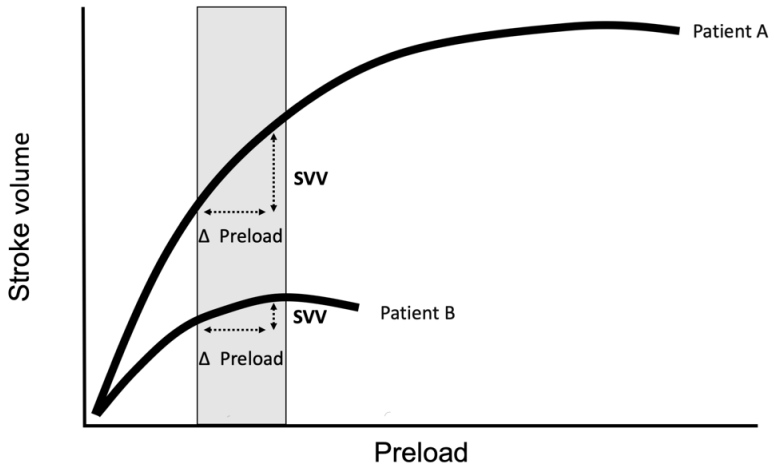

Figure 1.5 Patients A and B have different curves because of different contractility and afterload. Given a same change in absolute preload, patient A has a greater change in SV than does patient B. Patient A is a responder and therefore has a greater variation in SV even though absolute changes in preload are identical for these two patients.

To define the SVV of a patient, it is necessary to continuously measure SV or CO by echocardiography, or pulse contour analysis (Feissel et al., 2001; Yi, Liu, Qiao, Wan, & Mu, 2017) (see Section 4. MEASUREMENT OF CARDIA OUTPUT IN DOGS in Chapter 1). However, in veterinary clinical practice, it is challenging to accurately measure SV. Therefore, an alternative dynamic parameter reflecting SVV has been

introduced, called pulse pressure variation (PPV). PPV is obtained directly from the peripheral arterial pressure waveform (Yang & Du, 2014). Unlike continuous echocardiography, it is feasible to place an arterial line in dogs to measure direct arterial pressure during anaesthesia.

3.2.2 Pulse pressure variation (PPV)

Pulse pressure (PP: difference between systolic and diastolic arterial pressure) is almost proportional to SV, and inversely proportional to arterial compliance (Bighamian & Hahn, 2014). During positive pressure ventilation, arterial compliance remain relatively constant over the course of a single breath, thus PP can be used as surrogate of SV, and thus PPV is almost identical to SVV. In other words, SVV or PPV induced by positive pressure mechanical ventilation will be greater when the patient sits on the steep portion of the curve, predicting an increase in SV following fluid loading. PPV is calculated as the maximal pulse pressure less the minimum pulse pressure divided by the average of these two pressures (Michard et al., 1999)(Figure 5).

$$PPV = 100 \text{ X } \frac{(\text{Maximum PP} - \text{Minimum PP})}{\text{Average PP}}$$

Where Average PP = (Maximum PP + Minimum PP)/ 2

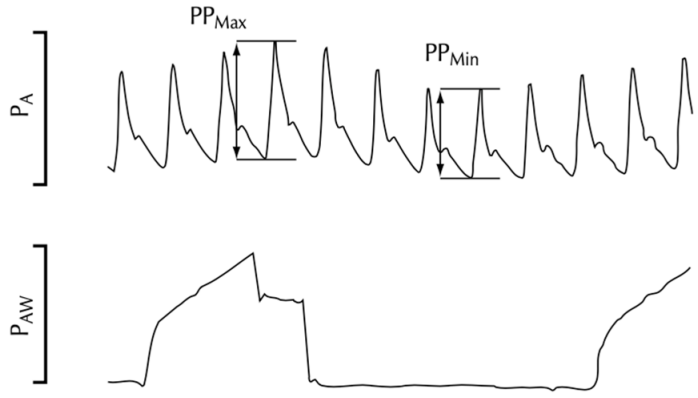

Figure 1.6 Pulse pressure variation (PPV) PPV, 100 × (PP_{Max} - PP_{Min})/Average PP; Average PP, (PP_{Max} + PP_{Min})/2; P_A, arterial pressure; P_{AW}, airway pressure; PP_{Max}, maximum pulse pressure after a positive pressure breath; PP_{Min}, minimum pulse pressure after a positive pressure breath (from Scott et al. 2001).

PPV has been shown to be highly predictive of fluid responsiveness in a systematic review of its use in people (Marik et al., 2009). The major finding of this systematic review and meta-analysis was that PPV during positive pressure mechanical ventilation with a tidal volume > 8 mL/kg was an accurate predictor of fluid responsiveness in critically ill people, with a sensitivity of 0.88 [95% confidence interval (95%CI): 0.81 to 0.92)], a specificity of 0.89 (95%CI: 0.84 to 0.92), and summary receiver operating characteristic (ROC) area under the curve (AUC) of 0.94 (95%CI: 0.91 to 0.95) (Yang & Du, 2014). Another meta-analysis evaluated 22 studies that included a total of 807 mechanically ventilated patients, to evaluate the value of PPV in predicting fluid responsiveness (Hong et al., 2014). This reported a summary ROC AUC for PPV of 0.94 (95%CI: 0.91 to 0.95), with a pooled sensitivity of 0.88 (95%CI: 0.81 to 0.92) and a pooled specificity of 0.89 (95%CI: 0.84 to 0.92) (Yang & Du, 2014). Overall, the median

cutoff value for PPV (expressed as a percentage) to predict fluid responsiveness in this meta-analysis was 12% (interquartile range 10 to 13%) with a sensitivity of 0.88 and a specificity of 0.89. The grey zone (below which patients are unlikely to respond, and above which they are expected to respond) was identified as being between 10 and 13%.

3.2.3 Pulse pressure variation (PPV) for dogs

In mechanically ventilated anaesthetised experimental dogs, I found that the PPV predicted fluid responsiveness with ROC AUC of 0.85 (95%CI: 0.70 to 1.00, $p = 0.038$) and cutoff values of 11% (sensitivity 79%; specificity 80%) (**Chapter 2**)(Sano, Seo, et al., 2018). At the time of submission of that study for publication, there were no previously published studies in dogs. Subsequently, Fantoni et al (2017) reported the predictive value of PPV in 33 and 39 mechanically ventilated anaesthetised dogs that underwent orthopaedic surgery and ovariohysterectomy respectively. Those authors found the PPV predicted fluid responsiveness with ROC AUC of 0.89 and 0.98, and the best cutoff values were deemed to be 15% and 16% (Fantoni et al., 2017). Both the results of Fantoni et al and those presented in Chapter 2 are similar to the data in people. A further study in 63 client-owned dogs found that a PPV ≥ 13% reliably predicted fluid responsiveness in 82.9% of cases (Drozdzynska, Chang, Stanzani, & Pelligand, 2018). Therefore, PPV is recommended as a useful clinical tool to detect occult hypovolaemia or to prevent hypervolaemia and predict the cardiovascular response to fluid challenge in anaesthetised dogs being mechanically ventilated. However, PPV requires invasive arterial catheter placement, which may be challenging in small and/or hypotensive dogs. Therefore, a non-invasive technique would be desirable in veterinary practices.

3.2.4 Pleth Variability Index (PVI)

PVI is a dynamic parameter that is measured non-invasively, that allows for continuous and automated calculation of mechanical ventilation-induced variations in the pulse oximetry waveform amplitude (Desebbe et al., 2010). In contrast to measurement of PPV, PVI can be calculated from the non-invasive placement of a pulse oximeter probe, which provides sufficient information to predict fluid responsiveness.

The perfusion index (PI) is an assessment of the pulsatile strength at the monitoring site. The PI is an indirect and non-invasive measure of peripheral perfusion. It is calculated by means of pulse oximetry by expressing the pulsatile signal (during arterial inflow) as a percentage of the non-pulsatile signal, both of which are derived from the amount of infrared light absorbed (Goldman, Petterson, Kopotic, & Barker, 2000; Sun & Huang, 2014). It is comparable to pulse pressure.

$$PI = 100\ \mathrm{X}\ \frac{\text{direct current}}{\text{alternate current}}$$

PVI can be automatically calculated by anaesthetic monitoring equipment using maximum and minimum values of PI during mechanically ventilatory cycles (Sun & Huang, 2014) (Figure 6).

$$PPV = 100\ \mathrm{X}\ \frac{(\text{Maximum PI} - \text{Minimum PI})}{\text{Maximum PI}}$$

PVI can predict fluid responsiveness in mechanically ventilated people (Cannesson, Delannoy, et al., 2008; Cannesson, Desebbe, et al., 2008). A systematic review and meta-analysis of 18 studies involving 665 people showed that the PVI has a reasonable ability to predict fluid responsiveness with the pooled ROC AUC of 0.88 (95%CI: 0.84 to 0.91), the pooled sensitivity of 0.73 (95%CI: 0.68 to 0.78) and specificity of 0.82 (95%CI: 0.77

to 0.86) (Chu, Wang, Sun, & Wang, 2016). Furthermore, PVI measured in sedated patients prior to induction, has been reported as a good predictor of hypotension following general anaesthesia induction (Tsuchiya, Yamada, & Asada, 2010). Therefore, these results could be extrapolated to dogs.

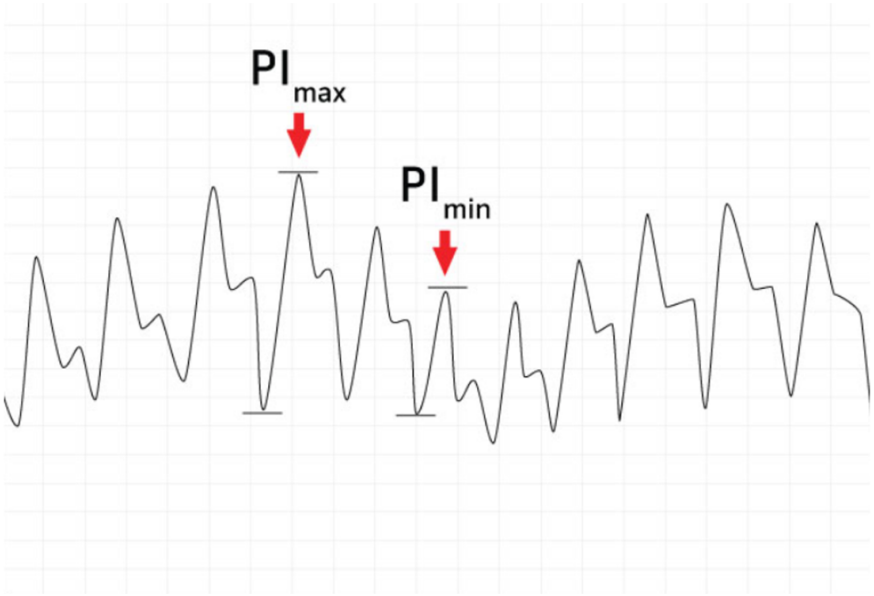

Figure 1.7 Pleth Variability Index (PVI) PVI, 100 × (PI_{max} - PI_{min})/PI_{max}; PI_{max}, Maximum perfusion index; PI_{min}, Minimum PI (from Masimo HP; https://www.masimo.com/pvi/)

3.2.5 Pleth Variability Index (PVI) for dogs

In mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine, PVI predicted fluid responsiveness with ROC AUC of 0.84 (95%CI: 0.68 to 1.00, $p = 0.043$) and cutoff values of 9.3% (sensitivity 0. 86; specificity 0.70) (**Chapter 2**) (Sano, Seo, et al., 2018). At the time of publication, there were no previous studies in dogs, however a subsequent clinical study of 39 mechanically ventilated anaesthetised dogs has been published (Celeita-Rodríguez et al., 2019). In that subsequent study, the PVI could reliably predict fluid responsiveness with ROC AUC of 0.91 using a cut-off

value of 11% (Celeita-Rodríguez et al., 2019). PVI can be more easily used in routine veterinary practice than PPV, since PVI can be measured noninvasively and does not require an arterial catheter, however mechanical ventilation is still needed. Unfortunately, availability of mechanical ventilation in veterinary practice is not common (Sano, Barker, et al., 2018) and therefore, alternative methods need to be investigated.

3.3 Limitation of dynamic predictors of fluid responsiveness

The dynamic predictors described above are less reliable to predict fluid responsiveness in spontaneously breathing patients than in mechanically ventilated patients (Soubrier et al., 2007). However, as stated, mechanical ventilation is not widely available in veterinary practice, and also those techniques cannot be used in unanaesthetised animals. Moreover, dynamic indices are affected by not only the preload but also other factors such as heart rate (Morgan, Abel, Mullins, & Guntheroth, 1966), respiratory rate (De Backer, Taccone, Holsten, Ibrahimi, & Vincent, 2009), pleural pressure (Liu et al., 2016), and tidal volume (Díaz, Erranz, Donoso, Salomon, & Cruces, 2015; Kim & Pinsky, 2008). These confounding factors may decrease clinical application (Marik & Lemson, 2014). Therefore, further investigations to determine their accuracy for the assessment of fluid responsiveness are essential.

3.4 Mini-fluid challenge

The mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in SV after a small loading dose of fluid. Theoretically, the change in SV at the steep portion of the Frank-Starling curve will be greater than at the plateau portion after the administration of a small fluid load, or a larger fluid volume (Figure 1.8). Muller et al (2011) showed that there was a good correlation between the increase in SV after 100 mL

of the mini-fluid challenge, and the increase in SV after 500 mL of the fluid challenge (r = 0.81, 95%CI: 0.66 – 0.90 (Muller et al., 2011). Therefore, the magnitude of the change in SV after a small fluid loading could predict responsiveness to a larger fluid bolus. Studies in people have shown that a mini-fluid challenge could predict fluid responsiveness in both mechanically ventilated and spontaneously breathing people (Guinot et al., 2015; Muller et al., 2011). Messina et al (2019) published a systematic review and a metanalysis of 21 studies of the reliability of the mini-fluid challenge in predicting fluid responsiveness in a total of 805 human patients in the intensive care unit and operating room. The pooled ROC AUC for the mini-fluid challenge was 0.91 (95%CI: 0.85 to 0.97) and the pooled sensitivity and specificity were 0.82 (95%CI: 0.76 to 0.88) and 0.83 (95%CI: 0.77 to 0.89) respectively, with a best threshold of 5% (grey zone: 3.0 to 7.0%) (Messina et al., 2019). To my knowledge, the study described in Chapter 4 of this thesis is the only one published that has evaluated the mini-fluid challenge in dogs (Sano, Fujiyama, et al., 2019), (Sano, Fujiyama, et al., 2019). In that study, I found that the mini-fluid challenge of 3mL/kg was a reliable predictor of fluid responsiveness in mechanically ventilated dogs, with a ROC AUC of 0.93 (95%CI: 0.79 to 1.00), sensitivity of 1.00 and specificity of 0.90. However, that study was in clinically healthy dogs, and an assessment of its accuracy in clinical patients will be required before widespread use in practice can be recommended.

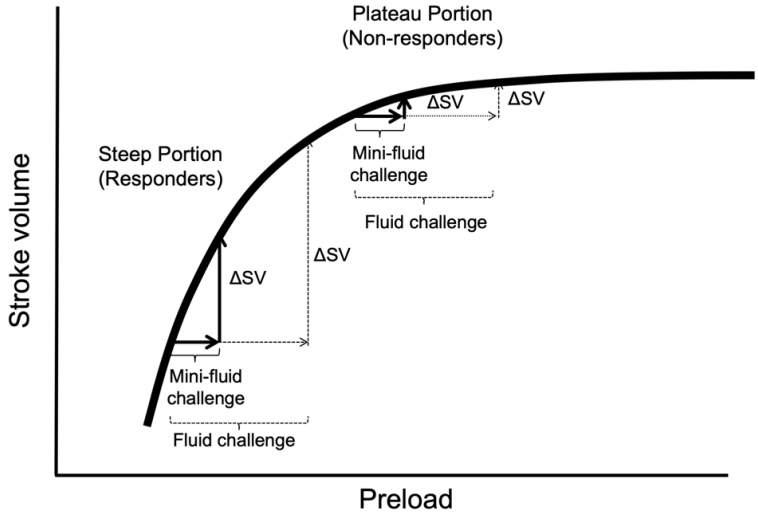

Figure 1.8 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

4. MEASUREMENT OF CARDIA OUTPUT IN DOGS

4.1 Clinical gold standard technique

Measurement of SV and CO can facilitate cardiovascular management during anaesthesia. Assessment of trends in SV and CO are also important when treating with inotropic, vasoactive drugs and intravenous fluids (Hasanin, 2015). The thermodilution technique with a pulmonary artery catheter is considered the clinical standard method to measure CO in dogs based on the Stewart–Hamilton equation. It was described (Fegler, 1954) and tested in dogs using regression analysis, resulting in a good agreement with the Fick method (Hendriks, Schipperheyn, & Quanjer, 1978). However, the placement of a pulmonary artery catheter is clinically invasive and challenging in veterinary practice and associated with increased postoperative complications in human medicine (Harvey et al., 2005; Sakka, Reinhart, Wegscheider, & Meier-Hellmann, 2000). Therefore, there is a need for more clinically feasible yet accurate means of measuring SV and CO in veterinary practice.

4.2 Minimally invasive technique

Several minimally invasive methods for CO measurement have been tested in dogs. Arterial pressure contour analysis is a minimally invasive technique for continuous CO determination. Systems for the arterial pulse contour analysis require calibration by an invasive technique such as a thermodilution technique, which has been performed for the PiCCO (Pulsion Medical Systems, Germany) (Garofalo et al., 2016; Hofer, Cecconi, Marx, & della Rocca, 2009) and the LiDCO (LiDCO Ltd., Cambridge, UK)(Mathews & Singh, 2008) in people. However, comparisons between those systems and with the gold standard thermodilution technique in dogs have produced conflicting results (Garofalo et al., 2016; Morgaz et al., 2014). The FloTrac/Vigileo system (Edwards Lifesciences AG,

Switzerland), without prior calibration, uses the arterial pressure contour to continuously monitor and/or calculate SV, CO, SVV and systemic vascular resistance through a standard peripheral arterial line based on algorithm using the relationship between aortic pulse pressure, SV and aortic compliance (Bektas et al., 2012). Although this system is less invasive, it requires an arterial line and did not have a good agreement with the thermodilution technique in dogs (Bektas et al., 2012).

Ultrasonography is able to estimate blood flow through a valve orifice using colour-flow Doppler. The area under the velocity-time curve can be calculated automatically in an ultrasound machine. This area under the curve is called the velocity time integral, and it is proportional to how far blood moves during the time period. If the valve orifice is assumed to be circular, the orifice area can be calculated using the diameter (area = $\pi \times \text{radius}^2$), which can be measured using ultrasonography. The formula for SV is area multiplied by VTI (SV = area × VTI). CO can be calculated by heart rate multiplied by SV (CO = HR × VTI). Importantly, ultrasonography is non-invasive, and transthoracic echocardiography at the main pulmonary artery provides an accurate measurement of CO with an excellent correlation with thermodilution techniques in dogs (Lopes et al., 2010). Similarly, transoesophageal echocardiography provides good agreement with the thermodilution technique in dogs (Mantovani et al., 2017; Yamashita et al., 2007) and transoesophageal Doppler devices accurately reflected the direction and magnitude of the changes of CO over time during abrupt hemodynamic changes in dogs (de Figueiredo, Cruz, Silva, & Rocha, 2004). However, all of these techniques require an expensive machine and an expert with a high level of skill and are thus not easily applicable to general veterinary practice.

4.3 Pulse wave transit time

Pulse wave transit time (PWTT) is the time from the electrocardiogram (ECG) R-wave peak to the rise point of the pulse oximeter wave (Sugo et al., 2010). The rise point of the pulse wave is defined as the point at which the differentiated pulse wave reached 30% of its peak amplitude (Figure 1.9). PWTT, a measure of velocity, has proven to be inversely proportional to SV and has a strong correlation with SV in dogs (Sugo et al., 2010) and people (Ishihara et al., 2004). Based on this relationship, Sugo et al. developed a system to estimate SV and CO using PWTT (Sugo, Sakai, Terao, Ukawa, & Ochiai, 2012). This requires an initial 3-minute period of stable haemodynamics for calibration against another CO measurement system, or an automatic patient information calibration based on the patient's information, and their arterial pulse pressure (Ishihara et al., 2004), which is similar to arterial pressure contour analysis. It is easy to use, inexpensive, minimally invasive and requires only routine anaesthetic monitoring available in most veterinary clinics (pulse oximetry, ECG, non-invasive or invasive arterial blood pressure monitoring). In a canine experiment, the correlation of estimated CO with CO obtained using electromagnetic flow meters on the aorta, was high ($r = 0.825$), however the agreement between the two methods was not clearly reported (Sugo et al., 2010). Because a high correlation does not always indicate good agreement between the two methods, precision of agreement with percentage error using Bland–Altman analysis should be used to assess interchangeability of two methods (Critchley, Lee, & Ho, 2010).

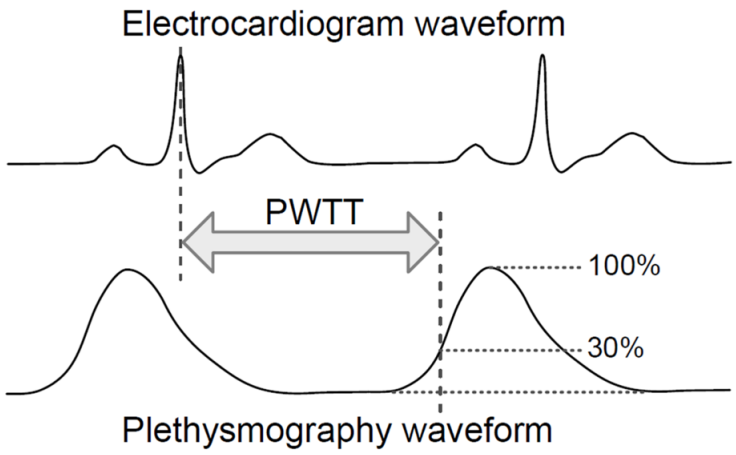

Figure 1.9 Pulse Wave Transit Time (PWTT). PWTT was calculated as the time from the ECG R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

5. PERIOPERATIVE HAEMODYNAMIC MANAGEMENT IN DOGS

5.1 Purpose of perioperative haemodynamic management

Anaesthesia is essential when animals need major or minor surgeries, endoscopy, diagnostic imaging, or other invasive diagnostic techniques. Anaesthesia provides unconsciousness, amnesia, muscle relaxation, and analgesia to animals, allowing these procedures to be performed humanely without pain or excessive physiological responses. However, haemodynamic depression can be caused concurrently, as anaesthetic agents are usually cardiovascular depressants (D. Brodbelt, 2009; Gaynor et al., 1999). Haemodynamic depression may preclude sufficient oxygen delivery (DO_2) to tissues, causing tissue hypoxia. Inappropriate perioperative haemodynamic management has been shown to cause severe tissue hypoxia during anaesthesia, resulting in many clinical complications such as surgical infection, delayed wound healing, prolonged hospitalization, and multiple organ dysfunction syndrome in dogs (Snowdon, Smeak, & Chiang, 2016; Turk, Singh, & Weese, 2015) and people (Monk, Saini, Weldon, & Sigl, 2005; Tassoudis et al., 2011).

The main goal of perioperative haemodynamic management is to provide adequate DO_2 to major organs such as the brain, heart, and kidneys, and to peripheral tissues. DO_2 depends on haemoglobin (Hb) concentration, saturation of arterial Hb with oxygen (SaO_2), and CO. CO is directly reduced by anaesthetic agents, but Hb and SaO_2 are not. Assuming normal Hb and SaO_2 (although both can be reduced under anaesthesia under some circumstances), CO determines DO_2. Therefore, optimisation of CO will lead to successful perioperative haemodynamic management. Consistent with that, a systematic review and meta-analysis showed that optimisation of CO has been shown to decrease

perioperative morbidity and hospital length of stay in cardiac patientss (Aya, Cecconi, Hamilton, & Rhodes, 2013).

5.2 Perioperative blood pressure management

As described above, it is challenging to measure CO in veterinary practices. Thus, instead of monitoring CO, blood pressure, especially mean arterial pressure (MAP), is used as a surrogate measure of tissue perfusion in dogs and people (Monk et al., 2005). Cerebral blood flow autoregulation is supposed to maintain normal perfusion between a MAP of 60 and 150 mm Hg (Figure 1.10) (Dagal & Lam, 2009; Paulson, Strandgaard, & Edvinsson, 1990) whilst autoregulation of renal perfusion flow drops off steeply at a MAP of 70 mmHg (Shipley & Study, 1951) (Figure 1.11) . Thus, MAP needs to be maintained at least 60 mmHg to prevent brain ischaemia, or more than 70 mmHg to provide enough blood perfusion to maintain normal kidney function during anaesthesia (Iizuka, Kamata, Yanagawa, & Nishimura, 2013; Redondo et al., 2007). However, most anaesthetic agents have vasodilatory and negative inotropic effects that can cause hypotension (D. Brodbelt, 2009; Gaynor et al., 1999). In fact, severe hypotension (MAP < 60 mmHg or systolic arterial pressure < 80 mmHg) occurs in up to 65% of anaesthetised in dogs and cats (Iizuka et al., 2013; Redondo et al., 2007). Therefore, monitoring of MAP and the prompt treatment of low MAP is necessary.

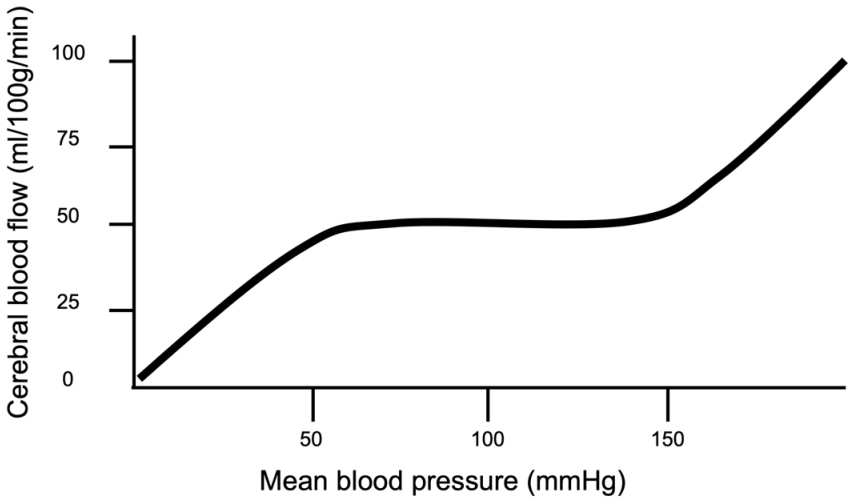

Figure 1.10 Cerebral blood flow autoregulation (Paulson et al., 1990). It typically operates between MAP of the order of 60 and 150 mmHg (normal MAP).

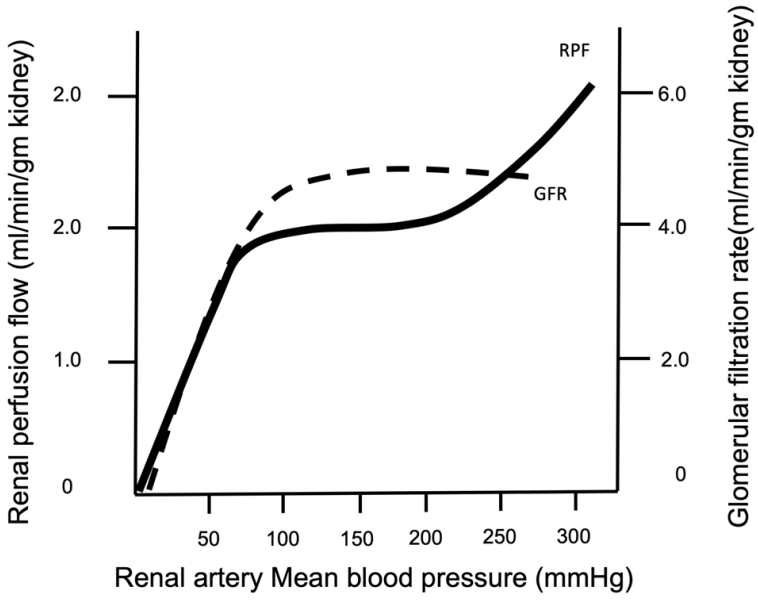

Figure 1.11 Renal perfusion flow autoregulation (Shipley & Study, 1951). Renal perfusion flow (RPF) drops off at 70 mmHg but the plateau really starts at about 110 mmHg. Glomerular filtration (GFR) plateaus at 120 mmHg and by the time renal artery mean blood pressure is 70 mmHg the GFR is at about 70%.

5.3 Treatment for hypotension in small animals

Hypotension under anaesthesia is a frequent occurrence, even in healthy patients. When observed, assessment of anaesthetic depth should be the first action because deep anaesthesia is a common cause (Monk et al., 2005). Probably the most common treatment of hypotension if anaesthetic depth is deemed appropriate is the administration of a fluid bolus. However, caution should be taken when using fluid therapy as the sole method to correct anaesthesia-related hypotension as high rates of fluids can exacerbate complications rather than prevent them (Muir et al., 2011; Voldby & Brandstrup, 2016). Based on 2013 AAHA/ AAFP fluid therapy guidelines in dogs and cats (Davis et al., 2013), the following process is the recommended treatment of hypotension in the anaesthetised dogs:

1. Decrease anaesthetic depth and/or inhalant concentration.
2. Provide an intravenous bolus of an isotonic crystalloid such as lactated Ringer's solution (3–10 mL/kg). Repeat once if needed.
3. If response is inadequate, consider intravenous administration of a colloid such as hetastarch. Slowly administer 5–10 mL/kg for dogs and 1–5 mL/kg for cats, titrating to effect to minimize the risk of vascular overload (measure blood pressure every 3–5 minutes) (Chappell et al., 2008). Colloids are more likely to increase blood pressure than crystalloids (Aarnes, Bednarski, Lerche, Hubbell, & Muir, 2009).
4. If response to crystalloid and/or colloid boluses is inadequate and patient is not hypovolemic, techniques other than fluid therapy may be needed (e.g., inopressors or, balanced anaesthetic techniques) (Chappell et al., 2008).

As described above, excessive fluid administration can result from the treatment of hypotension in the anaesthetised dogs. Therefore, prevention of hypotension using inopressors such as noradrenaline could reduce the incidence of fluid overload in clinical

veterinary practices (Sano, Chambers, & Bridges, 2019). However, peripheral perfusion may be impeded due to excessive vasoconstriction, which can be severe enough to cause peripheral gangrene (Kwon, Hong, & Park, 2018).

6 AIM AND OBJECTIVES OF THESIS

This literature review established that although fluid therapy is important in order to improve haemodynamics in dogs and people, excessive fluid administration is detrimental. The clinical challenge is to determine which patient will respond to fluid administration, and to determine the correct amount of fluid for each patient. Currently, there are several methods to determine fluid responsiveness in people but studies in dogs are scarce. Dynamic parameters to predict fluid responsiveness are often used in people but have many limitations. Application of dynamic parameters to dogs is possible but clinically limited because of their invasiveness and the requirement for a positive pressure mechanical ventilator. The mini-fluid challenge is an alternative method to identify fluid responsiveness and may be used in spontaneously breathing dogs. However, reliable measurement of the change in stroke volume is necessary. Pulse wave transit time, which can be clinically obtained non-invasively in dogs, may be used as a surrogate parameter for SV. The mini-fluid challenge using pulse wave transit time has the potential to detect fluid responsiveness in dogs non-invasively. However, those methods are still cumbersome in clinical veterinary practice because there is currently a lack of suitable equipment. Simple prophylactic noradrenaline infusion may be able to prevent hypotension in anaesthetised dogs and reduce the incidence of fluid overload caused by the typical treatment of hypotension in veterinary practice: a bolus of fluid.

6.1 Thesis aim

The aim of the research presented in this thesis was to develop clinically feasible methods to determine fluid responsiveness and prevent fluid overload in anaesthetised dogs.

6.2 Thesis Objectives

The investigation of clinical methods to prevent excessive fluid administration was described by the following objectives:

Chapter 2 To evaluate whether PPV and PVI are more accurate than CVP for predicting fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine.

Chapter 3 To evaluate the ability of PWTT to detect changes in SV and to estimate CO compared with the thermodilution technique in isoflurane-anaesthetised dogs.

Chapter 4 To investigate whether percentage changes in PWTT induced by mini-fluid challenges predict fluid responsiveness in mechanically ventilated anaesthetised dogs.

Chapter 5 To investigate whether percentage changes in PWTT following mini-fluid challenge could predict fluid responsiveness in spontaneously breathing anaesthetised dogs.

Chapter 6 To investigate whether noradrenaline infusion prior to hypotension improves anaesthetic management in dogs undergoing ovariohysterectomy.

Three experimental studies in healthy dogs, and two clinical trials in client owned healthy dogs were conducted to achieve these objectives.

Abbreviations

AUC	area under a curve
CO	cardiac output (mL)
CVP	central venous pressure (mmHg)
DO_2	oxygen delivery (mL/minute)
Hb	haemoglobin (g/dL)
HR	heart rate (beat/minute)
MAP	mean arterial pressure (mmHg)
PI	perfusion index
PPV	pulse pressure variation (%)
PVI	pleth Variability Index (%)
PWTT	pulse wave transit time (msecond)
ROC	receiver operating characteristic
SV	stroke volume (mL)
SVV	stroke volume variation (%)
VTI	velocity time integral (cm)
95%CI	95% confidence interval

7 REFERENCES

Aarnes, T. K., Bednarski, R. M., Lerche, P., Hubbell, J. A., & Muir, W. W., 3rd. (2009). Effect of intravenous administration of lactated Ringer's solution or hetastarch for the treatment of isoflurane-induced hypotension in dogs. *Am J Vet Res, 70*(11), 1345-1353. doi:10.2460/ajvr.70.11.1345

Arieff, A. I. (1999). Fatal postoperative pulmonary edema: pathogenesis and literature review. *Chest, 115*(5), 1371-1377. doi:10.1378/chest.115.5.1371

Aya, H. D., Cecconi, M., Hamilton, M., & Rhodes, A. (2013). Goal-directed therapy in cardiac surgery: a systematic review and meta-analysis. *Br J Anaesth, 110*(4), 510-517. doi:10.1093/bja/aet020

Bednarski, R., Grimm, K., Harvey, R., Lukasik, V. M., Penn, W. S., Sargent, B., & Spelts, K. (2011). AAHA anesthesia guidelines for dogs and cats. *J Am Anim Hosp Assoc, 47*(6), 377-385. doi:10.5326/jaaha-ms-5846

Bektas, R. N., Kutter, A. P., Hartnack, S., Jud, R. S., Schnyder, M., Matos, J. M., & Bettschart-Wolfensberger, R. (2012). Evaluation of a minimally invasive non-calibrated pulse contour cardiac output monitor (FloTrac/Vigileo) in anaesthetized dogs. *Vet Anaesth Analg, 39*(5), 464-471. doi:10.1111/j.1467-2995.2012.00741.x

Bighamian, R., & Hahn, J. O. (2014). Relationship between stroke volume and pulse pressure during blood volume perturbation: a mathematical analysis. *Biomed Res Int, 2014*, 459269. doi:10.1155/2014/459269

Bishop, M. H., Shoemaker, W. C., Appel, P. L., Meade, P., Ordog, G. J., Wasserberger, J., . . . et al. (1995). Prospective, randomized trial of survivor values of cardiac index, oxygen delivery, and oxygen consumption as resuscitation endpoints in severe trauma. *J Trauma, 38*(5), 780-787. doi:10.1097/00005373-199505000-00018

Bishop, M. H., Shoemaker, W. C., Appel, P. L., Wo, C. J., Zwick, C., Kram, H. B., . . . Fleming, A. W. (1993). Relationship between supranormal circulatory values, time delays, and outcome in severely traumatized patients. *Crit Care Med, 21*(1), 56-63. doi:10.1097/00003246-199301000-00013

Boyd, O., Grounds, R. M., & Bennett, E. D. (1993). A randomized clinical trial of the effect of deliberate perioperative increase of oxygen delivery on mortality in high-risk surgical patients. *Jama, 270*(22), 2699-2707.

Brandstrup, B., Svensen, C., & Engquist, A. (2006). Hemorrhage and operation cause a contraction of the extracellular space needing replacement--evidence and implications? A systematic review. *Surgery, 139*(3), 419-432. doi:10.1016/j.surg.2005.07.035

Brandstrup, B., Tonnesen, H., Beier-Holgersen, R., Hjortso, E., Ording, H., Lindorff-Larsen, K., . . . Pott, F. (2003). Effects of intravenous fluid restriction on postoperative complications: comparison of two perioperative fluid regimens: a randomized assessor-blinded multicenter trial. *Ann Surg, 238*(5), 641-648. doi:10.1097/01.sla.0000094387.50865.23

Breznock, E. M., & Strack, D. (1982). Blood volume of nonsplenectomized and splenectomized cats before and after acute hemorrhage. *Am J Vet Res, 43*(10), 1811-1814.

Brodbelt, D. (2009). Perioperative mortality in small animal anaesthesia. *Vet J, 182*(2), 152-161. doi:10.1016/j.tvjl.2008.06.011

Brodbelt, D. C., Pfeiffer, D. U., Young, L. E., & Wood, J. L. (2007). Risk factors for anaesthetic-related death in cats: results from the confidential enquiry into

perioperative small animal fatalities (CEPSAF). *Br J Anaesth, 99*(5), 617-623. doi:10.1093/bja/aem229

Bundgaard-Nielsen, M., Secher, N. H., & Kehlet, H. (2009). 'Liberal' vs. 'restrictive' perioperative fluid therapy--a critical assessment of the evidence. *Acta Anaesthesiol Scand, 53*(7), 843-851. doi:10.1111/j.1399-6576.2009.02029.x

Cannesson, M., Delannoy, B., Morand, A., Rosamel, P., Attof, Y., Bastien, O., & Lehot, J. J. (2008). Does the Pleth variability index indicate the respiratory-induced variation in the plethysmogram and arterial pressure waveforms? *Anesth Analg, 106*(4), 1189-1194, table of contents. doi:10.1213/ane.0b013e318167ab1f

Cannesson, M., Desebbe, O., Rosamel, P., Delannoy, B., Robin, J., Bastien, O., & Lehot, J. J. (2008). Pleth variability index to monitor the respiratory variations in the pulse oximeter plethysmographic waveform amplitude and predict fluid responsiveness in the operating theatre. *Br J Anaesth, 101*(2), 200-206. doi:10.1093/bja/aen133

Celeita-Rodríguez, N., Teixeira-Neto, F. J., Garofalo, N. A., Dalmagro, T. L., Girotto, C. H., Oliveira, G. C. V., & Santos, I. F. (2019). Comparison of the diagnostic accuracy of dynamic and static preload indexes to predict fluid responsiveness in mechanically ventilated, isoflurane anesthetized dogs. *Vet Anaesth Analg, 46*(3), 276-288. doi:10.1016/j.vaa.2018.12.004

Chappell, D., Jacob, M., Hofmann-Kiefer, K., Conzen, P., & Rehm, M. (2008). A rational approach to perioperative fluid management. *Anesthesiology, 109*(4), 723-740. doi:10.1097/ALN.0b013e3181863117

Chong, M. A., Wang, Y., Berbenetz, N. M., & McConachie, I. (2018). Does goal-directed haemodynamic and fluid therapy improve peri-operative outcomes?: A systematic review and meta-analysis. *Eur J Anaesthesiol, 35*(7), 469-483. doi:10.1097/eja.0000000000000778

Chu, H., Wang, Y., Sun, Y., & Wang, G. (2016). Accuracy of pleth variability index to predict fluid responsiveness in mechanically ventilated patients: a systematic review and meta-analysis. *J Clin Monit Comput, 30*(3), 265-274. doi:10.1007/s10877-015-9742-3

Coller, F. A., Dick, V. S., & Maddock, W. G. (1936). Maintenance of normal water exchange with intravenous fluids. *Journal of the American Medical Association, 107*, 1522–1527.

Corcoran, T., Rhodes, J. E., Clarke, S., Myles, P. S., & Ho, K. M. (2012). Perioperative fluid management strategies in major surgery: a stratified meta-analysis. *Anesth Analg, 114*(3), 640-651. doi:10.1213/ANE.0b013e318240d6eb

Cotton, B. A., Guy, J. S., Morris, J. A., Jr., & Abumrad, N. N. (2006). The cellular, metabolic, and systemic consequences of aggressive fluid resuscitation strategies. *Shock, 26*(2), 115-121. doi:10.1097/01.shk.0000209564.84822.f2

Critchley, L. A., Lee, A., & Ho, A. M. (2010). A critical review of the ability of continuous cardiac output monitors to measure trends in cardiac output. *Anesth Analg, 111*(5), 1180-1192. doi:10.1213/ANE.0b013e3181f08a5b

Dagal, A., & Lam, A. M. (2009). Cerebral autoregulation and anesthesia. *Curr Opin Anaesthesiol, 22*(5), 547-552. doi:10.1097/ACO.0b013e32833020be

Davis, H., Jensen, T., Johnson, A., Knowles, P., Meyer, R., Rucinsky, R., & Shafford, H. (2013). 2013 AAHA/AAFP fluid therapy guidelines for dogs and cats. *J Am Anim Hosp Assoc, 49*(3), 149-159. doi:10.5326/jaaha-ms-5868

De Backer, D., Taccone, F. S., Holsten, R., Ibrahimi, F., & Vincent, J. L. (2009). Influence of respiratory rate on stroke volume variation in mechanically ventilated

patients. *Anesthesiology, 110*(5), 1092-1097. doi:10.1097/ALN.0b013e31819db2a1

De Backer, D., & Vincent, J. L. (2018). Should we measure the central venous pressure to guide fluid management? Ten answers to 10 questions. *Crit Care, 22*(1), 43. doi:10.1186/s13054-018-1959-3

de Figueiredo, L. F., Cruz, R. J., Jr., Silva, E., & Rocha, E. S. M. (2004). Cardiac output determination during experimental hemorrhage and resuscitation using a transesophageal Doppler monitor. *Artif Organs, 28*(4), 338-342. doi:10.1111/j.1525-1594.2004.47351.x

Desebbe, O., Boucau, C., Farhat, F., Bastien, O., Lehot, J. J., & Cannesson, M. (2010). The ability of pleth variability index to predict the hemodynamic effects of positive end-expiratory pressure in mechanically ventilated patients under general anesthesia. *Anesth Analg, 110*(3), 792-798. doi:10.1213/ANE.0b013e3181cd6d06

Díaz, F., Erranz, B., Donoso, A., Salomon, T., & Cruces, P. (2015). Influence of tidal volume on pulse pressure variation and stroke volume variation during experimental intra-abdominal hypertension. *BMC Anesthesiol, 15*, 127. doi:10.1186/s12871-015-0105-x

Doherty, M., & Buggy, D. J. (2012). Intraoperative fluids: how much is too much? *Br J Anaesth, 109*(1), 69-79. doi:10.1093/bja/aes171

Drozdzynska, M. J., Chang, Y. M., Stanzani, G., & Pelligand, L. (2018). Evaluation of the dynamic predictors of fluid responsiveness in dogs receiving goal-directed fluid therapy. *Vet Anaesth Analg, 45*(1), 22-30. doi:10.1016/j.vaa.2017.06.001

Durham, R. M., Neunaber, K., Mazuski, J. E., Shapiro, M. J., & Baue, A. E. (1996). The use of oxygen consumption and delivery as endpoints for resuscitation in critically ill patients. *J Trauma, 41*(1), 32-39; discussion 39-40. doi:10.1097/00005373-199607000-00007

Egerton Jennings, C. (1882). The intra-venuos injection of fluid for severe haemorrhage. *Lancet, 120*(3082), 436–437.

Fantoni, D. T., Ida, K. K., Gimenes, A. M., Mantovani, M. M., Castro, J. R., Patrício, G. C. F., . . . Otsuki, D. A. (2017). Pulse pressure variation as a guide for volume expansion in dogs undergoing orthopedic surgery. *Vet Anaesth Analg, 44*(4), 710-718. doi:10.1016/j.vaa.2016.11.011

Fegler, G. (1954). Measurement of cardiac output in anaesthetized animals by a thermodilution method. *Q J Exp Physiol Cogn Med Sci, 39*(3), 153-164. doi:10.1113/expphysiol.1954.sp001067

Feissel, M., Michard, F., Mangin, I., Ruyer, O., Faller, J. P., & Teboul, J. L. (2001). Respiratory changes in aortic blood velocity as an indicator of fluid responsiveness in ventilated patients with septic shock. *Chest, 119*(3), 867-873. doi:10.1378/chest.119.3.867

Fischer, P. E., Nunn, A. M., Wormer, B. A., Christmas, A. B., Gibeault, L. A., Green, J. M., & Sing, R. F. (2013). Vasopressor use after initial damage control laparotomy increases risk for anastomotic disruption in the management of destructive colon injuries. *Am J Surg, 206*(6), 900-903. doi:10.1016/j.amjsurg.2013.07.034

Fukuda, N., Terui, T., Ohtsuki, I., Ishiwata, S., & Kurihara, S. (2009). Titin and troponin: central players in the frank-starling mechanism of the heart. *Curr Cardiol Rev, 5*(2), 119-124. doi:10.2174/157340309788166714

Garofalo, N. A., Teixeira-Neto, F. J., Rodrigues, J. C., Cerejo, S. A., Aguiar, A. J., & Becerra-Velásquez, D. R. (2016). Comparison of Transpulmonary Thermodilution and Calibrated Pulse Contour Analysis with Pulmonary Artery

Thermodilution Cardiac Output Measurements in Anesthetized Dogs. *J Vet Intern Med, 30*(4), 941-950. doi:10.1111/jvim.13984

Gattinoni, L., Brazzi, L., Pelosi, P., Latini, R., Tognoni, G., Pesenti, A., & Fumagalli, R. (1995). A trial of goal-oriented hemodynamic therapy in critically ill patients. SvO2 Collaborative Group. *N Engl J Med, 333*(16), 1025-1032. doi:10.1056/nejm199510193331601

Gaynor, J. S., Dunlop, C. I., Wagner, A. E., Wertz, E. M., Golden, A. E., & Demme, W. C. (1999). Complications and mortality associated with anesthesia in dogs and cats. *J Am Anim Hosp Assoc, 35*(1), 13-17. doi:10.5326/15473317-35-1-13

Goldman, J. M., Petterson, M. T., Kopotic, R. J., & Barker, S. J. (2000). Masimo signal extraction pulse oximetry. *J Clin Monit Comput, 16*(7), 475-483. doi:10.1023/a:1011493521730

Grubb, T., Sager, J., Gaynor, J. S., Montgomery, E., Parker, J. A., Shafford, H., & Tearney, C. (2020). 2020 AAHA Anesthesia and Monitoring Guidelines for Dogs and Cats. *J Am Anim Hosp Assoc, 56*(2), 59-82. doi:10.5326/jaaha-ms-7055

Guinot, P. G., Bernard, E., Defrancq, F., Petiot, S., Majoub, Y., Dupont, H., & Lorne, E. (2015). Mini-fluid challenge predicts fluid responsiveness during spontaneous breathing under spinal anaesthesia: An observational study. *Eur J Anaesthesiol, 32*(9), 645-649. doi:10.1097/eja.0000000000000175

Guyton, A. C. (1955). Determination of cardiac output by equating venous return curves with cardiac response curves. *Physiol Rev, 35*(1), 123-129. doi:10.1152/physrev.1955.35.1.123

Guyton, A. C., & Hall, J. E. (2015). *Textbook of Medical Physiolog* (13th ed.).

Harvey, S., Harrison, D. A., Singer, M., Ashcroft, J., Jones, C. M., Elbourne, D., . . . Rowan, K. (2005). Assessment of the clinical effectiveness of pulmonary artery catheters in management of patients in intensive care (PAC-Man): a randomised controlled trial. *Lancet, 366*(9484), 472-477. doi:10.1016/s0140-6736(05)67061-4

Hasanin, A. (2015). Fluid responsiveness in acute circulatory failure. *J Intensive Care, 3*, 50. doi:10.1186/s40560-015-0117-0

Hayes, M. A., Timmins, A. C., Yau, E. H., Palazzo, M., Hinds, C. J., & Watson, D. (1994). Elevation of systemic oxygen delivery in the treatment of critically ill patients. *N Engl J Med, 330*(24), 1717-1722. doi:10.1056/nejm199406163302404

Hendriks, F. F., Schipperheyn, J. J., & Quanjer, P. H. (1978). Thermal dilution measurement of cardiac output in dogs using an analog computer. *Basic Res Cardiol, 73*(5), 459-468. doi:10.1007/bf01906526

Hofer, C. K., Cecconi, M., Marx, G., & della Rocca, G. (2009). Minimally invasive haemodynamic monitoring. *Eur J Anaesthesiol, 26*(12), 996-1002. doi:10.1097/eja.0b013e3283300d55

Holte, K., Jensen, P., & Kehlet, H. (2003). Physiologic effects of intravenous fluid administration in healthy volunteers. *Anesth Analg, 96*(5), 1504-1509, table of contents. doi:10.1213/01.ane.0000055820.56129.ee

Hong, J. Q., He, H. F., Chen, Z. Y., Du, Z. S., Liu, W. F., Weng, P. Q., & Huang, H. B. (2014). Comparison of stroke volume variation with pulse pressure variation as a diagnostic indicator of fluid responsiveness in mechanically ventilated critically ill patients. *Saudi Med J, 35*(3), 261-268.

Iizuka, T., Kamata, M., Yanagawa, M., & Nishimura, R. (2013). Incidence of intraoperative hypotension during isoflurane-fentanyl and propofol-fentanyl anaesthesia in dogs. *Vet J, 198*(1), 289-291. doi:10.1016/j.tvjl.2013.06.021

Ishihara, H., Okawa, H., Tanabe, K., Tsubo, T., Sugo, Y., Akiyama, T., & Takeda, S. (2004). A new non-invasive continuous cardiac output trend solely utilizing routine cardiovascular monitors. *J Clin Monit Comput, 18*(5-6), 313-320. doi:10.1007/s10877-005-2452-5

Jacob, M., Chappell, D., & Rehm, M. (2009). The 'third space'--fact or fiction? *Best Pract Res Clin Anaesthesiol, 23*(2), 145-157. doi:10.1016/j.bpa.2009.05.001

Jia, F. J., Yan, Q. Y., Sun, Q., Tuxun, T., Liu, H., & Shao, L. (2017). Liberal versus restrictive fluid management in abdominal surgery: a meta-analysis. *Surg Today, 47*(3), 344-356. doi:10.1007/s00595-016-1393-6

Kim, H. K., & Pinsky, M. R. (2008). Effect of tidal volume, sampling duration, and cardiac contractility on pulse pressure and stroke volume variation during positive-pressure ventilation. *Crit Care Med, 36*(10), 2858-2862. doi:10.1097/CCM.0b013e3181865aea

Kwon, J. W., Hong, M. K., & Park, B. Y. (2018). Risk Factors of Vasopressor-Induced Symmetrical Peripheral Gangrene. *Ann Plast Surg, 80*(6), 622-627. doi:10.1097/sap.0000000000001314

Lang, K., Boldt, J., Suttner, S., & Haisch, G. (2001). Colloids versus crystalloids and tissue oxygen tension in patients undergoing major abdominal surgery. *Anesth Analg, 93*(2), 405-409 , 403rd contents page. doi:10.1097/00000539-200108000-00034

Lassen, K., Soop, M., Nygren, J., Cox, P. B., Hendry, P. O., Spies, C., . . . Dejong, C. H. (2009). Consensus review of optimal perioperative care in colorectal surgery: Enhanced Recovery After Surgery (ERAS) Group recommendations. *Arch Surg, 144*(10), 961-969. doi:10.1001/archsurg.2009.170

Latta, T. (1832). Malignant cholera: Documents communicated by the Central Board of Health, London, relative to the treatment of cholera by the copious injection of aqueous and saline fluids into the vein. *Lancet, 18*(457), 274–280.

Liu, Y., Wei, L. Q., Li, G. Q., Yu, X., Li, G. F., & Li, Y. M. (2016). Pulse Pressure Variation Adjusted by Respiratory Changes in Pleural Pressure, Rather Than by Tidal Volume, Reliably Predicts Fluid Responsiveness in Patients With Acute Respiratory Distress Syndrome. *Crit Care Med, 44*(2), 342-351. doi:10.1097/ccm.0000000000001371

Lobo, D. N., Bostock, K. A., Neal, K. R., Perkins, A. C., Rowlands, B. J., & Allison, S. P. (2002). Effect of salt and water balance on recovery of gastrointestinal function after elective colonic resection: a randomised controlled trial. *Lancet, 359*(9320), 1812-1818. doi:10.1016/s0140-6736(02)08711-1

Lopes, P. C., Sousa, M. G., Camacho, A. A., Carareto, R., Nishimori, C. T., Santos, P. S., & Nunes, N. (2010). Comparison between two methods for cardiac output measurement in propofol-anesthetized dogs: thermodilution and Doppler. *Vet Anaesth Analg, 37*(5), 401-408. doi:10.1111/j.1467-2995.2010.00552.x

Lowell, J. A., Schifferdecker, C., Driscoll, D. F., Benotti, P. N., & Bistrian, B. R. (1990). Postoperative fluid overload: not a benign problem. *Crit Care Med, 18*(7), 728-733. doi:10.1097/00003246-199007000-00010

MacKay, G., Fearon, K., McConnachie, A., Serpell, M. G., Molloy, R. G., & O'Dwyer, P. J. (2006). Randomized clinical trial of the effect of postoperative intravenous fluid restriction on recovery after elective colorectal surgery. *Br J Surg, 93*(12), 1469-1474. doi:10.1002/bjs.5593

Magder, S. (2010). Fluid status and fluid responsiveness. *Curr Opin Crit Care, 16*(4), 289-296. doi:10.1097/MCC.0b013e32833b6bab

Magder, S. (2015). Understanding central venous pressure: not a preload index? *Curr Opin Crit Care, 21*(5), 369-375. doi:10.1097/mcc.0000000000000238

Mantovani, M. M., Fantoni, D. T., Gimenes, A. M., de Castro, J. R., Flor, P. B., Ida, K. K., & Schwartz, D. S. (2017). Clinical monitoring of cardiac output assessed by transoesophageal echocardiography in anaesthetised dogs: a comparison with the thermodilution technique. *BMC Vet Res, 13*(1), 325. doi:10.1186/s12917-017-1227-9

Marik, P. E., Baram, M., & Vahid, B. (2008). Does central venous pressure predict fluid responsiveness? A systematic review of the literature and the tale of seven mares. *Chest, 134*(1), 172-178. doi:10.1378/chest.07-2331

Marik, P. E., Cavallazzi, R., Vasu, T., & Hirani, A. (2009). Dynamic changes in arterial waveform derived variables and fluid responsiveness in mechanically ventilated patients: a systematic review of the literature. *Crit Care Med, 37*(9), 2642-2647. doi:10.1097/CCM.0b013e3181a590da

Marik, P. E., & Lemson, J. (2014). Fluid responsiveness: an evolution of our understanding. *Br J Anaesth, 112*(4), 617-620. doi:10.1093/bja/aet590

Matas, R. (1924). THE CONTINUED INTRAVENOUS "DRIP": WITH REMARKS ON THE VALUE OF CONTINUED GASTRIC DRAINAGE AND IRRIGATION BY NASAL INTUBATION WITH A GASTRODUODENAL TUBE (JUTTE) IN SURGICAL PRACTICE. *Ann Surg, 79*(5), 643-661. doi:10.1097/00000658-192405000-00001

Mathews, L., & Singh, R. K. (2008). Cardiac output monitoring. *Ann Card Anaesth, 11*(1), 56-68. doi:10.4103/0971-9784.38455

McArdle, G. T., McAuley, D. F., McKinley, A., Blair, P., Hoper, M., & Harkin, D. W. (2009). Preliminary results of a prospective randomized trial of restrictive versus standard fluid regime in elective open abdominal aortic aneurysm repair. *Ann Surg, 250*(1), 28-34. doi:10.1097/SLA.0b013e3181ad61c8

Messina, A., Dell'Anna, A., Baggiani, M., Torrini, F., Maresca, G. M., Bennett, V., . . . Cecconi, M. (2019). Functional hemodynamic tests: a systematic review and a metanalysis on the reliability of the end-expiratory occlusion test and of the mini-fluid challenge in predicting fluid responsiveness. *Crit Care, 23*(1), 264. doi:10.1186/s13054-019-2545-z

Michard, F., Chemla, D., Richard, C., Wysocki, M., Pinsky, M. R., Lecarpentier, Y., & Teboul, J. L. (1999). Clinical use of respiratory changes in arterial pulse pressure to monitor the hemodynamic effects of PEEP. *Am J Respir Crit Care Med, 159*(3), 935-939. doi:10.1164/ajrccm.159.3.9805077

Michard, F., & Teboul, J. L. (2002). Predicting fluid responsiveness in ICU patients: a critical analysis of the evidence. *Chest, 121*(6), 2000-2008. doi:10.1378/chest.121.6.2000

Monk, T. G., Saini, V., Weldon, B. C., & Sigl, J. C. (2005). Anesthetic management and one-year mortality after noncardiac surgery. *Anesth Analg, 100*(1), 4-10. doi:10.1213/01.Ane.0000147519.82841.5e

Morgan, B. C., Abel, F. L., Mullins, G. L., & Guntheroth, W. G. (1966). Flow patterns in cavae, pulmonary artery, pulmonary vein, and aorta in intact dogs. *Am J Physiol, 210*(4), 903-909. doi:10.1152/ajplegacy.1966.210.4.903

Morgaz, J., Granados Mdel, M., Muñoz-Rascón, P., Dominguez, J. M., Fernández-Sarmiento, J. A., Gómez-Villamandos, R. J., & Navarrete, R. (2014). Comparison of thermodilution, lithium dilution, and pulse contour analysis for the measurement of cardiac output in 3 different hemodynamic states in dogs. *J Vet Emerg Crit Care (San Antonio), 24*(5), 562-570. doi:10.1111/vec.12219

Muir, W. W., 3rd, Kijtawornrat, A., Ueyama, Y., Radecki, S. V., & Hamlin, R. L. (2011). Effects of intravenous administration of lactated Ringer's solution on hematologic, serum biochemical, rheological, hemodynamic, and renal measurements in healthy isoflurane-anesthetized dogs. *J Am Vet Med Assoc, 239*(5), 630-637. doi:10.2460/javma.239.5.630

Muller, L., Toumi, M., Bousquet, P. J., Riu-Poulenc, B., Louart, G., Candela, D., . . . Lefrant, J. Y. (2011). An increase in aortic blood flow after an infusion of 100 ml colloid over 1 minute can predict fluid responsiveness: the mini-fluid challenge study. *Anesthesiology, 115*(3), 541-547. doi:10.1097/ALN.0b013e318229a500

Mutz, N., Baum, M., Benzer, H., Koller, W., Moritz, E., & Pauser, G. (1984). Intraoperative application of high-frequency ventilation. *Crit Care Med, 12*(9), 800-802. doi:10.1097/00003246-198409000-00027

Myles, P., Bellomo, R., Corcoran, T., Forbes, A., Wallace, S., Peyton, P., . . . Parke, R. (2017). Restrictive versus liberal fluid therapy in major abdominal surgery (RELIEF): rationale and design for a multicentre randomised trial. *BMJ Open, 7*(3), e015358. doi:10.1136/bmjopen-2016-015358

Myles, P. S., Bellomo, R., Corcoran, T., Forbes, A., Peyton, P., Story, D., . . . Wallace, S. (2018). Restrictive versus Liberal Fluid Therapy for Major Abdominal Surgery. *N Engl J Med, 378*(24), 2263-2274. doi:10.1056/NEJMoa1801601

Navarro, L. H., Bloomstone, J. A., Auler, J. O., Jr., Cannesson, M., Rocca, G. D., Gan, T. J., . . . Kramer, G. C. (2015). Perioperative fluid therapy: a statement from the international Fluid Optimization Group. *Perioper Med (Lond), 4*, 3. doi:10.1186/s13741-015-0014-z

Nisanevich, V., Felsenstein, I., Almogy, G., Weissman, C., Einav, S., & Matot, I. (2005). Effect of intraoperative fluid management on outcome after intraabdominal surgery. *Anesthesiology, 103*(1), 25-32. doi:10.1097/00000542-200507000-00008

Noel-Morgan, J., & Muir, W. W. (2018). Anesthesia-Associated Relative Hypovolemia: Mechanisms, Monitoring, and Treatment Considerations. *Front Vet Sci, 5*, 53. doi:10.3389/fvets.2018.00053

Norberg, A., Hahn, R. G., Li, H., Olsson, J., Prough, D. S., Borsheim, E., . . . Svensen, C. H. (2007). Population volume kinetics predicts retention of 0.9% saline infused in awake and isoflurane-anesthetized volunteers. *Anesthesiology, 107*(1), 24-32. doi:10.1097/01.anes.0000268387.34758.6d

O'Shaughnessy, W. B. (1831). Proposal of a new method of treating the blue epidemic cholera by the injection of highly oxygenised salts into the venous system. *Lancet, 17*(432), 366–371.

Oh, T. K., Song, I. A., Do, S. H., Jheon, S., & Lim, C. (2019). Association of perioperative weight-based fluid balance with 30-day mortality and acute kidney injury among patients in the surgical intensive care unit. *J Anesth, 33*(3), 354-363. doi:10.1007/s00540-019-02630-8

Paulson, O. B., Strandgaard, S., & Edvinsson, L. (1990). Cerebral autoregulation. *Cerebrovasc Brain Metab Rev, 2*(2), 161-192.

Plurad, D. S., Talving, P., Lam, L., Inaba, K., Green, D., & Demetriades, D. (2011). Early vasopressor use in critical injury is associated with mortality independent from volume status. *J Trauma, 71*(3), 565-570; discussion 570-562. doi:10.1097/TA.0b013e3182213d52

Rahbari, N. N., Zimmermann, J. B., Schmidt, T., Koch, M., Weigand, M. A., & Weitz, J. (2009). Meta-analysis of standard, restrictive and supplemental fluid

administration in colorectal surgery. *Br J Surg, 96*(4), 331-341. doi:10.1002/bjs.6552

Ratner, A. J., Lysenko, E. S., Paul, M. N., & Weiser, J. N. (2005). Synergistic proinflammatory responses induced by polymicrobial colonization of epithelial surfaces. *Proc Natl Acad Sci U S A, 102*(9), 3429-3434. doi:10.1073/pnas.0500599102

Redondo, J. I., Rubio, M., Soler, G., Serra, I., Soler, C., & Gómez-Villamandos, R. J. (2007). Normal values and incidence of cardiorespiratory complications in dogs during general anaesthesia. A review of 1281 cases. *J Vet Med A Physiol Pathol Clin Med, 54*(9), 470-477. doi:10.1111/j.1439-0442.2007.00987.x

Robotham, J. L., Cherry, D., Mitzner, W., Rabson, J. L., Lixfeld, W., & Bromberger-Barnea, B. (1983). A re-evaluation of the hemodynamic consequences of intermittent positive pressure ventilation. *Crit Care Med, 11*(10), 783-793. doi:10.1097/00003246-198310000-00005

Ruttmann, T. G., James, M. F., & Aronson, I. (1998). In vivo investigation into the effects of haemodilution with hydroxyethyl starch (200/0.5) and normal saline on coagulation. *Br J Anaesth, 80*(5), 612-616. doi:10.1093/bja/80.5.612

Sakka, S. G., Reinhart, K., Wegscheider, K., & Meier-Hellmann, A. (2000). Is the placement of a pulmonary artery catheter still justified solely for the measurement of cardiac output? *J Cardiothorac Vasc Anesth, 14*(2), 119-124. doi:10.1016/s1053-0770(00)90002-8

Sano, H., Barker, K., Odom, T., Lewis, K., Giordano, P., Walsh, V., & Chambers, J. P. (2018). A survey of dog and cat anaesthesia in a sample of veterinary practices in New Zealand. *N Z Vet J, 66*(2), 85-92. doi:10.1080/00480169.2017.1413959

Sano, H., Chambers, P., & Bridges, J. (2019). Effects of preemptive norepinephrine infusion prior to hypotension on anesthetic management in dogs undergoing ovariohysterectomy. *Veterinary Anaesthesia and Analgesia, 46*(6), 831.e814-831.e815. doi:10.1016/j.vaa.2019.08.039

Sano, H., Fujiyama, M., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2019). Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. *J Vet Emerg Crit Care (San Antonio), 29*(4), 391-398. doi:10.1111/vec.12860

Sano, H., Seo, J., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2018). Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anesthetized dogs. *J Vet Emerg Crit Care (San Antonio), 28*(4), 301-309. doi:10.1111/vec.12728

Schol, P. B., Terink, I. M., Lance, M. D., & Scheepers, H. C. (2016). Liberal or restrictive fluid management during elective surgery: a systematic review and meta-analysis. *J Clin Anesth, 35*, 26-39. doi:10.1016/j.jclinane.2016.07.010

Self, W. H., Semler, M. W., Bellomo, R., Brown, S. M., deBoisblanc, B. P., Exline, M. C., . . . Shapiro, N. I. (2018). Liberal Versus Restrictive Intravenous Fluid Therapy for Early Septic Shock: Rationale for a Randomized Trial. *Ann Emerg Med, 72*(4), 457-466. doi:10.1016/j.annemergmed.2018.03.039

Shipley, R. E., & Study, R. S. (1951). Changes in renal blood flow, extraction of inulin, glomerular filtration rate, tissue pressure and urine flow with acute alterations of renal artery blood pressure. *Am J Physiol, 167*(3), 676-688. doi:10.1152/ajplegacy.1951.167.3.676

Shires, T., Williams, J., & Brown, F. (1961). Acute change in extracellular fluids associated with major surgical procedures. *Ann Surg, 154*, 803-810. doi:10.1097/00000658-196111000-00005

Shoemaker, W. C., Appel, P. L., Kram, H. B., Waxman, K., & Lee, T. S. (1988). Prospective trial of supranormal values of survivors as therapeutic goals in high-risk surgical patients. *Chest, 94*(6), 1176-1186. doi:10.1378/chest.94.6.1176

Snowdon, K. A., Smeak, D. D., & Chiang, S. (2016). Risk Factors for Dehiscence of Stapled Functional End-to-End Intestinal Anastomoses in Dogs: 53 Cases (2001-2012). *Vet Surg, 45*(1), 91-99. doi:10.1111/vsu.12413

Soubrier, S., Saulnier, F., Hubert, H., Delour, P., Lenci, H., Onimus, T., . . . Durocher, A. (2007). Can dynamic indicators help the prediction of fluid responsiveness in spontaneously breathing critically ill patients? *Intensive Care Med, 33*(7), 1117-1124. doi:10.1007/s00134-007-0644-9

Sugo, Y., Sakai, T., Terao, M., Ukawa, T., & Ochiai, R. (2012). The comparison of a novel continuous cardiac output monitor based on pulse wave transit time and echo Doppler during exercise. *Conf Proc IEEE Eng Med Biol Soc, 2012*, 236-239. doi:10.1109/embc.2012.6345913

Sugo, Y., Ukawa, T., Takeda, S., Ishihara, H., Kazama, T., & Takeda, J. (2010). A novel continuous cardiac output monitor based on pulse wave transit time. *Conf Proc IEEE Eng Med Biol Soc, 2010*, 2853-2856. doi:10.1109/iembs.2010.5626343

Sun, S., & Huang, S. Q. (2014). Role of pleth variability index for predicting hypotension after spinal anesthesia for cesarean section. *Int J Obstet Anesth, 23*(4), 324-329. doi:10.1016/j.ijoa.2014.05.011

Tang, J., Wu, G., & Peng, L. (2011). Pharmacokinetics of propofol in patients undergoing total hip replacement : effect of acute hypervolemic hemodilution. *Anaesthesist, 60*(9), 835-840. doi:10.1007/s00101-011-1907-y

Tassoudis, V., Vretzakis, G., Petsiti, A., Stamatiou, G., Bouzia, K., Melekos, M., & Tzovaras, G. (2011). Impact of intraoperative hypotension on hospital stay in major abdominal surgery. *J Anesth, 25*(4), 492-499. doi:10.1007/s00540-011-1152-1

Thomas, W. T. (1898). Injection of saline solution in shock. *Lancet, 152*, 1390– 1391.

Tsuchiya, M., Yamada, T., & Asada, A. (2010). Pleth variability index predicts hypotension during anesthesia induction. *Acta Anaesthesiol Scand, 54*(5), 596-602. doi:10.1111/j.1399-6576.2010.02225.x

Turk, R., Singh, A., & Weese, J. S. (2015). Prospective surgical site infection surveillance in dogs. *Vet Surg, 44*(1), 2-8. doi:10.1111/j.1532-950X.2014.12267.x

Valverde, A., Gianotti, G., Rioja-Garcia, E., & Hathway, A. (2012). Effects of high-volume, rapid-fluid therapy on cardiovascular function and hematological values during isoflurane-induced hypotension in healthy dogs. *Can J Vet Res, 76*(2), 99-108.

Velmahos, G. C., Demetriades, D., Shoemaker, W. C., Chan, L. S., Tatevossian, R., Wo, C. C., . . . Berne, T. V. (2000). Endpoints of resuscitation of critically injured patients: normal or supranormal? A prospective randomized trial. *Ann Surg, 232*(3), 409-418. doi:10.1097/00000658-200009000-00013

Vermeulen, H., Hofland, J., Legemate, D. A., & Ubbink, D. T. (2009). Intravenous fluid restriction after major abdominal surgery: a randomized blinded clinical trial. *Trials, 10*, 50. doi:10.1186/1745-6215-10-50

Vincent, J. L., & Weil, M. H. (2006). Fluid challenge revisited. *Crit Care Med, 34*(5), 1333-1337. doi:10.1097/01.Ccm.0000214677.76535.A5

Voldby, A. W., & Brandstrup, B. (2016). Fluid therapy in the perioperative setting-a clinical review. *J Intensive Care, 4*, 27. doi:10.1186/s40560-016-0154-3

Xu, C., Peng, J., Liu, S., Huang, Y., Guo, X., Xiao, H., & Qi, D. (2018). Goal-directed fluid therapy versus conventional fluid therapy in colorectal surgery: A meta analysis of randomized controlled trials. *Int J Surg, 56*, 264-273. doi:10.1016/j.ijsu.2018.06.034

Yamashita, K., Ueyama, Y., Miyoshi, K., Igarashi, R., Kushiro, T., Umar, M. A., & Muir, W. W. (2007). Minimally invasive determination of cardiac output by transthoracic bioimpedance, partial carbon dioxide rebreathing, and transesophageal Doppler echocardiography in beagle dogs. *J Vet Med Sci, 69*(1), 43-47. doi:10.1292/jvms.69.43

Yang, X., & Du, B. (2014). Does pulse pressure variation predict fluid responsiveness in critically ill patients? A systematic review and meta-analysis. *Crit Care, 18*(6), 650. doi:10.1186/s13054-014-0650-6

Yi, L., Liu, Z., Qiao, L., Wan, C., & Mu, D. (2017). Does stroke volume variation predict fluid responsiveness in children: A systematic review and meta-analysis. *PLoS One, 12*(5), e0177590. doi:10.1371/journal.pone.0177590

Chapter 2

Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anaesthetized dogs

Sano H, Seo J, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P
School of Veterinary Science, Massey University, Palmerston North, New Zealand

This chapter was published in *Journal of Veterinary Emergency and Critical Care:*

Sano H, Seo J, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P: Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anesthetized dogs. *Journal of Veterinary Emergency and Critical Care.* 28(4):301-309, 2018. doi: 10.1111/vec.12728.

This study was supported by Massey University Research Funds.

PREFACE

As we have seen from Chapter 1, both hypovolaemia and hypervolaemia are known to increase perioperative morbidity and mortality in people. In veterinary medicine, objective assessment of hydration status is not established, and many veterinarians have a tendency to provide more fluid to animals "just in case", which is likely to cause hypervolaemia. Therefore, fluid responsiveness should be closely monitored in order to avoid hypervolaemia. Static parameters such as central venous pressure (CVP) are still believed to be a clinical gold standard to estimate preload and fluid responsiveness in veterinary medicine even though studies in people have shown that CVP is not useful to predict fluid responsiveness. In the last decade, dynamic parameters such as pulse pressure variation (PPV) and pleth variability index (PVI) have been proved to be reliable predictors for fluid responsiveness in people. However, evaluation of dynamic parameters to predict fluid responsiveness had not been investigated in dogs at the time of the study. PPV and PVI are clinically measurable in veterinary practice as PPV can be calculated form invasive arterial blood pressure wave forms using an electronic pressure transducer and PVI can be obtained from non-invasive plethysmographic waveforms using a pulse oximeter. Therefore, in Chapter 2, we investigated whether PPV and PVI were more accurate than CVP for predicting fluid responsiveness in dogs.

This chapter was published in *Journal of Veterinary Emergency and Critical Care* and is formatted for the style of the journal.

ROLES OF EACH OF THE AUTHORS

Sano H: Primarily contributed to the study conception and design, collected and interpreted data with statistical analysis, prepared the manuscript and approved the final version to be published.

Seo J: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Wightman P: Contributed to ultrasound performance and prepared the manuscript.

Cave NJ: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Gieseg MA: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Johnson C: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Chambers P: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

ABSTRACT

Objective: To evaluate whether PPV and PVI are more accurate than CVP for predicting fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine.

Design: Prospective experimental trial.

Setting: University teaching hospital.

Animals: Twelve Harrier hound dogs.

Interventions: Each dog was anaesthetised and had a fluid challenge performed. This was repeated 4 weeks later for a total of 24 fluid challenges. After premedication with intramuscular acepromazine, anaesthesia was induced with propofol and maintained with isoflurane. The dogs were mechanically ventilated with constant settings. The fluid challenge consisted of 10 mL/kg of 6% hydroxyethyl starch intravenously over 13 minutes.

Measurements and Main Results: Before and after the fluid challenge, PPV, PVI, CVP, and other haemodynamics were recorded. Change in velocity time integral of pulmonary arterial blood flow by echocardiography was calculated as an indication of change in stroke volume. A fluid responder was defined as an increase in velocity time integral ≥ 15%. Receiver operator characteristic (ROC) curves were used to determine cutoff values. Areas under ROC curve were calculated and compared. Dogs responded on 14 fluid challenges and did not on 10. Cutoff values for PPV and PVI were 11% (sensitivity 79%; specificity 80%) and 9.3% (sensitivity 86%; specificity 70%) respectively. The areas under the ROC curve of PPV [0.85, 95% confidence interval (95%CI): 0.70-1.00, $p = 0.038$] and PVI (0.84, 95%CI: 0.68-1.00, $p = 0.043$) were significantly higher than CVP (0.56, 95%CI: 0.32-0.81). Grey zone values were identified ranges of 9-12% [7 out of 24 fluid challenges (29%)] for PPV and 8 - 12% [8 out of 24 fluid challenges (33%)]

for PVI.

Conclusions: PPV and PVI predicted fluid responsiveness more accurately than CVP and may be useful to guide fluid administration in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine.

Keywords: Frank-Starling, pulse oximetry, pulse pressure, stroke volume

Abbreviations

CO	cardiac output (mL)
CVP	central venous pressure (mmHg)
$F_E'Iso$	end-tidal concentration of isoflurane (%)
$P_E'CO_2$	end-tidal partial pressure of carbon dioxide (mmHg)
PIP	peak inspiratory pressure (cmH_2O)
PPV	pulse pressure variation (%)
PVI	pleth variability index (%)
ROC	receiver operator characteristic
SV	stroke volume (mL)
VT	tidal volume (mL/kg)
VTI	velocity time integral (cm)
95%CI	95% confidence interval

INTRODUCTION

Volume expansion with fluid administration is one of the standard treatments to optimise the haemodynamics of anaesthetised and critical patients. The goal of fluid administration is to increase preload and improve stroke volume (SV), cardiac output (CO), and thus tissue perfusion. However, fluid administration may not improve haemodynamics in a significant percentage of patients (Pinsky & Teboul, 2005). In an ICU, almost 50% of septic patients did not respond to fluid administration (Kelm et al., 2015) and it could have caused adverse effects including haemodilution (Aarnes, Bednarski, Lerche, Hubbell, & Muir, 2009; Valverde, Gianotti, Rioja-Garcia, & Hathway, 2012) and tissue oedema (Holte, Jensen, & Kehlet, 2003), both of which could have impaired oxygen and nutrient delivery to tissues (Cotton, Guy, Morris, & Abumrad, 2006). In addition, excessive perioperative fluid administration can contribute to postoperative complications, including prolonged length of hospital stay, organ failure, and mortality in people (Boyd, Forbes, Nakada, Walley, & Russell, 2011; Kelm et al., 2015; Lobo, Macafee, & Allison, 2006; Rosenberg, Dechert, Park, & Bartlett, 2009). Therefore, volume expansion should be considered only for those patients that are likely to respond.

Assessment of fluid responsiveness has been challenging in clinical practice because SV is dependent on integration of preload, afterload, and cardiac contractility, all of which are clinically difficult to evaluate. Traditionally static parameters such as CVP were believed to be good predictors for fluid responsiveness. The CVP, the pressure measured in the vena cava close to the right atrium, indicates right ventricular filling pressure, which approximates to preload and should be proportional to SV. However, the CVP has been shown to be unreliable for predicting fluid responsiveness in people (Kumar et al., 2004; Marik & Cavallazzi, 2013) and dogs (Bandt C, 2012; Berkenstadt et al., 2005; Sasaki,

Mutoh, Mutoh, Kawashima, & Tsubone, 2016; Taguchi et al., 2011) because the preload is determined not only by cardiac filling pressures, but also on the compliance of the heart and venous tone. Dynamic parameters for predicting fluid responsiveness have been developed and introduced into clinical use in human anaesthesia and critical care medicine (Cavallaro, Sandroni, & Antonelli, 2008). In healthy normal animals, SV is proportional to preload (Frank-Starling curve), but this relationship reaches a plateau and then becomes inversely proportional. In mechanically ventilated patients who are in the steep portion of the Frank-Starling curve, the SV will change more in response to the variation of preload caused by positive-pressure ventilation (Figure 2.1: Responders)(Michard, Lopes, & Auler, 2007; Vieillard-Baron et al., 2004). In contrast, patients who are on the plateau portion of the curve will experience only a small change in SV in response to preload changes induced by the same positive-pressure ventilation (Figure 2.1: Non-responders)(Michard et al., 2007; Vieillard-Baron et al., 2004). Thus, fluid responsiveness, (i.e. the ability of the heart to increase SV in response to fluid administration), is related to the magnitude of the ventilation induced changes in SV and hence arterial blood pressure (Cavallaro et al., 2008; Michard, 2005; Michard et al., 2007). Therefore, measurement of dynamic parameters has been shown to more accurately predict fluid responsiveness than traditional static parameters in mechanically ventilated people (Cannesson et al., 2008; Michard et al., 2000; Natalini et al., 2006; Preisman, Kogan, Berkenstadt, & Perel, 2005).

PPV (Marik, Cavallazzi, Vasu, & Hirani, 2009; Michard et al., 2000; Michard et al., 2007; Natalini et al., 2006) and PVI (Cannesson et al., 2007; Cannesson et al., 2008; Natalini et al., 2006) are dynamic parameters that are measurable in veterinary practice (Diniz et al., 2014; Klein et al., 2016). Both PPV and PVI are derived from respiratory changes in

arterial blood pressure waveform and the plethysmography waveform of a non-invasive pulse oximeter caused by positive pressure ventilation (Figure 2.1). In mechanically ventilated anaesthetised dogs, PPV and PVI increased consistently with decreased blood volume and decreased with volume expansion (Diniz et al., 2014; Klein et al., 2016; H.-G. N. Ricco C, Shih A, Bandt C, Pavlisko N, Killos M & Queiroz P, 2012; H.-G. N. Ricco C, Shih A, Pavlisko N, Killos M & Queiroz P, 2012). In hypovolaemic or haemorrhagic patients, the intrathoracic veins are compressed by the raised intrathoracic pressure, causing large oscillations in SV and arterial pressure, thus increase in PPV and PVI. However, the assessment of PPV and PVI to predict fluid responsiveness had not been reported in dogs with non-haemorrhagic conditions at the time of this study.

Therefore, the aims of this study were to: (1) obtain the cutoff values of PPV and PVI to discriminate between responders and non-responders, and (2) compare the accuracy of PPV and PVI with CVP for predicting fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine, a commonly used premedicant in clinical practices. The hypothesis was that PPV and PVI could predict fluid responsiveness more accurately than CVP in dogs in a simulated clinical setting.

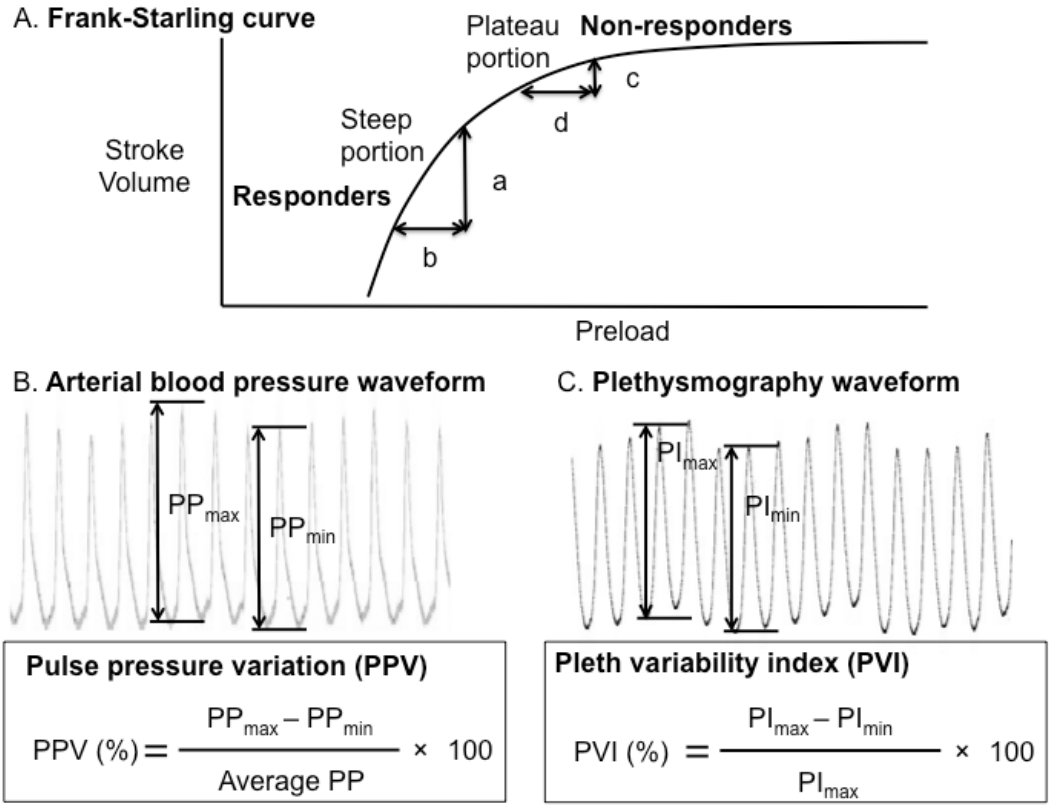

Figure 2.1 The Frank–Starling curve of the heart (A), definition of Pulse pressure variation (PPV) (B) and Pleth variability index (PVI) (C). In mechanically ventilated patients, stroke volume (SV) can change greater (a) in response to the variation of preload (b) caused by positive-pressure ventilation (Responders). In contrast, an increase in SV is relatively insensitive (c) to preload changes (d) on the plateau portion of the curve (Non-responders). PP, pulse pressure (systolic arterial pressure - diastolic arterial pressure); Average PP, (PPmax + PPmin)/2; PI, perfusion index; max, maximum; min, minimum.

MATERIALS AND METHODS

The study was approved by the animal ethics committee of Massey University (Protocol ID: 14/113). Twelve Harrier hound dogs (7 female, 5 male) were used. The dogs' median (interquartile range) age and mean (standard deviation) weight were 6 (3) years old and 25.4 (2.6) kg. The dogs were determined to be healthy on the basis of history, physical examination, stable bodyweight and blood work analysis. Each dog was anaesthetised twice with a 4-week interval between the 2 anaesthetics. As a part of a different study, dogs were randomised to either an overnight preanaesthetic fasting (12-16 hours), or a preanaesthetic feeding of 25% of calculated daily intake, using a moist diet (approximate 400 mL of water), given 3 hours prior to premedication. On the second anaesthesia, the dogs were crossed over to either be fed, or fasted. Water was provided ad libitum 3 hours prior to the administration of premedication. Therefore, a total of 24 anaesthetic occasions were included in this study.

Thirty minutes prior to the induction of anaesthesia, 0.05 mg/kg of acepromazine (Acezine 2, Ethical Agents Ltd, Manukau, NZ) was injected into the lumbar epaxial muscle. After placement of an 18- or 20-gauge catheter (Optiva I.V. catheter Radiopaque, Smiths Medical International Ltd, London, UK) into a cephalic vein, propofol (Repose, Norbrook NZ Ltd, Auckland, NZ) was administered intravenously to effect for the induction of anaesthesia (up to 4 mg/kg). Dogs were positioned in right lateral recumbency and orotracheally intubated for administration of isoflurane (Attane, Bayer Animal health New Zealand, Auckland, NZ) in 100% oxygen (2 L/minute), delivered via a rebreathing circuit (Excel 80 Anesthesia Machine, Datex-Ohmeda NZ Pty, Auckland, NZ). They were mechanically ventilated with volume-controlled mode (Excel 80 system with a 7800 Ventilator, Datex-Ohmeda NZ Pty, Auckland, NZ) with a tidal volume (VT)

of 15 mL/kg and inspiration-to-expiration ratio of 1:2 at a respiratory rate of 15 breath/minute and these respiratory settings were kept unchanged until the end of the study. An absence of spontaneous breathing was confirmed clinically by observing the capnograph waveform, variations in peak inspiratory pressure (PIP), and the dog's chest movements. Neuromuscular blocking agents, however, were not used in this study. The depth of anaesthesia was maintained with an end-tidal concentration of isoflurane ($F_E'Iso$) of 1.5 ± 0.05% throughout the study. A multipurpose bedside monitor (Life Scope BSM-3763, Nihon Kohden, Tokyo, Japan) was used to record heart rate, oxygen saturation of arterial haemoglobin, temperature, and blood pressure. End-tidal partial pressure of carbon dioxide ($P_E'CO_2$) and $F_E'Iso$ were measured by a second monitor (Anesthesia Gas monitor POET IQ, Criticare systems Inc, Waukesha, WI, USA). The dogs were maintained at normal rectal temperature by a warm air blanket (Mistral-air, Sound Veterinary Equipment, Ferntree Gully, VIC, AU).

Measurements

Arterial blood pressure and CVP

A 20- or 22-gauge catheter (Optiva I.V. catheter Radiopaque, Smiths Medical International Ltd, London, UK) was inserted into the dorsal pedal artery to measure arterial blood pressure and a 12-gauge long catheter (13 cm MILACATH, MILA International Inc, Erlanger, KY, USA) was placed into the left jugular vein to measure CVP (confirmed by pressure wave visualization). Both catheters were connected through a low compliance pressure line to an electronic pressure transducer (BD DTXPlus: DTX Plus TNF-R, Becton Dickinson Critical Care Systems PTE LTD, Singapore) that was calibrated to atmospheric pressure and positioned at the level of the manubrium. The system was pressurised to 300 mmHg and flushed with heparinised saline manually

before all measurements. A 2-point calibration (0-180 mmHg) was performed using a mercury manometer (FC-110DELUXE Mercurial Sphygmomanometer, Forcal Corporation, Chiba, Japan) and the damping coefficient was determined by a high-pressure flush test to ensure that it was around 0.7 before the study.

PPV and PVI

PPV was automatically calculated by the multipurpose bedside monitor (Life Scope BSM-3763, Nihon Kohden, Tokyo, Japan) attached to the arterial line. PVI was automatically calculated using a pulse oximeter (Radical 7, Masimo Corporation, Irvine, CA, USA) probe clipped on the tongue based on perfusion index, which is a numerical value that indicates the strength of the infrared signal returning from the monitoring site. Both equations were shown in Figure 2.1. After confirmation of cardiovascular stability, no further adjustment was performed through the study.

Velocity time integral (VTI) of main pulmonary artery, SV, and CO

Transthoracic echocardiography (Toshiba Xario 200 ultrasound unite with a 2–5 MHz phased array transducer, Toshiba, Tokyo, Japan) was used to determine VTI of main pulmonary arterial blood flow, SV, and CO. The transducer was positioned and fixed using a flexible holder in the 3rd intercostal space of the right hemithorax to obtain a longitudinal section of the right ventricular outlet. The VTI of the pulmonary artery was measured just distal to the valve cusps by pulsed wave Doppler at the end of expiration. The angle between the ultrasound beam and the blood flow was kept less than 20° and fixed throughout the study. The cross sectional area of the pulmonary artery was also estimated based on the measured diameter of the pulmonary artery. SV and CO were calculated as the following: SV = Cross sectional area × VTI, CO = SV × Heart rate.

Stroke volume index and cardiac index were also calculated by dividing SV and CO by each body weight. Percentage change in VTI was determined based on change in VTI before and after the fluid challenge. For each step of the study, VTI was measured in triplicate and averaged for the determination of the VTI value.

Experimental protocol

Forty to 45 minutes after the induction of anaesthesia, 15 minutes was allocated without any stimulation in order to confirm cardiopulmonary stability. Immediately before and a minute after the fluid challenge, measurements were recorded every minute for 5 minutes. The highest and lowest values were excluded, and the other 3 values were averaged to give a representative value. The fluid challenge consisted of administration of 10 mL/kg of 6% hydroxyethyl starch (Voluven 6%, Fresenius Kabi Australia Pty Limited, Pymble NSW, AU) via the CVP line over 13 minutes. On recovery, the dogs received 0.2 mg/kg of meloxicam (Metacam, Boehringer Ingelheim Vetmedica Inc, St Joseph, MO, USA) intravenously.

Statistical Methods

All data are presented as the mean (standard deviation) unless otherwise stated. Normality was assessed with the Shapiro-Wilk test. Comparisons of cardiorespiratory variables before and after the fluid challenge between responders and non-responders, and percentage changes in cardiorespiratory variables after the fluid challenge were assessed using an unpaired Student's t-test or a Mann-Whitney U test when appropriate. In order to confirm that dogs in the fasting group had a different fluid responsiveness from those in the feeding group, their changes in VTI after fluid challenge were compared using a paired Student's t-test. A responder was defined as having more than a 15% increase in

VTI, and a non-responder as less than a 15% increase in VTI based on human literature (Michard & Teboul, 2002). ROC curves were generated for PPV, PVI and CVP and the areas under the ROC curves were calculated and compared as previously described (Hanley & McNeil, 1983). The best cutoff values to discriminate responders and non-responders were defined by the point on the ROC curve determined by the maximum value of the Youden index (sensitivity + specificity - 1)(Ray, Le Manach, Riou, & Houle, 2010; Youden, 1950). The maximum index is represented graphically as the height above the diagonal line to minimize misclassification rates. To determine a clinically useful range (below which patients are unlikely to respond, and above which they are expected to respond), the grey zone was calculated (Cannesson et al., 2011). Youden index determination was then conducted for each bootstrapped population, resulting in a set of 1,000 "optimal" values. The mean value of these optimal values and its 95%CI were then estimated. Thus, the grey zone was defined along with the 95%CI (Cannesson et al., 2011). Values within the grey zone would correspond to a prediction not precise enough for fluid responsiveness. A sample size calculation showed that 22 fluid challenges were required to detect a difference of 0.15 in areas under the ROC curves between PPV and CVP based on a previous study (Natalini et al., 2006) and 24 fluid challenges were obtained from 12 anaesthetised dogs in this study. Data were analysed by statistical software (SPSS statistics 22, IBM, San Jose, CA, USA and MedCalc for Windows version 12.5, MedCalc Software, Oostende, Belgium).

RESULTS

Of the 24 fluid challenges, 14 fluid challenges increased VTI by more than 15 % (responders) and 10 did not (non-responders). Moreover, the VTI change after the fluid challenge in the over-night preanaesthetic fasting group (8 responders and 4 non-responders) was significantly higher than that in the preanaesthetic feeding group (4 responders and 8 non-responders) [21.5 (12.2) % vs 15.6 (10.1) %, $p = 0.012$]. The mean (standard deviation) dose of propofol administered for induction was 3.1 (0.2) mg/kg in responders and 3.2 (0.3) mg/kg in non-responders with no statistical difference ($p = 0.503$).

Cardiorespiratory variables compared between responders and non-responders

Before the fluid challenge, PPV and PVI were significantly higher in responders than in non-responders, and stroke volume index and cardiac index were significantly lower in responders than in non-responders (Table 2.1). There was no significant difference in arterial blood pressure, CVP, PIP, $P_E'CO_2$, $F_E'Iso$, and temperature between responders and non-responders before the fluid challenge (Table 2.1). After the fluid challenge, percentage changes in heart rate, systolic arterial pressure, diastolic arterial pressure, mean arterial pressure, stroke volume index, cardiac index, PPV, PVI, and VTI in responders were significantly greater than in non-responders, but change in CVP in non-responders was larger than in responders (Table 2.2).

Table 2.1 Comparisons of cardiorespiratory variables before and after the fluid challenge between responders and non-responders. Data are mean (standard deviation). *Significant difference between responders and non-responders ($p < 0.05$). CVP, central venous pressure; VTI, velocity time integral; PPV, pulse pressure variation; PVI, pleth variability index; PIP, peak inspiratory pressure; $P_E'CO_2$, End-tidal partial pressure of carbon dioxide; $F_E'Iso$, end-tidal concentration of isoflurane.

		Responders	Non-responders	*p*-Value
Heart rate (beat/minute)	Pre	91 (11)	95 (12)	0.428
	Post	107 (19)	99 (12)	0.209
Systolic arterial pressure (mmHg)	Pre	87 (8)	86 (8)	0.657
	Post	101 (13)	90 (9)	0.033*
Diastolic arterial pressure (mmHg)	Pre	48 (6)	46 (3)	0.156
	Post	54 (10)	46 (4)	0.014*
Mean arterial pressure (mmHg)	Pre	57 (6)	55 (4)	0.326
	Post	66 (11)	57 (4)	0.014*
Stroke volume index (mL/kg)	Pre	0.99 (0.13)	1.13 (0.13)	0.023*
	Post	1.26 (0.16)	1.21 (0.15)	0.484
Cardiac index (mL/minute/kg)	Pre	91 (19)	107 (17)	0.042*
	Post	136 (36)	120 (18)	0.145
CVP (mmHg)	Pre	4 (2)	3 (1)	0.625
	Post	6 (2)	6 (2)	0.991
VTI (cm)	Pre	12.0 (1.6)	13.1 (1.5)	0.126
	Post	15.2 (2)	14.0 (1.5)	0.139
PPV (%)	Pre	14 (4)	9 (2)	0.002*
	Post	8 (2)	8 (2)	0.712
PVI (%)	Pre	12 (4)	9 (3)	0.004*
	Post	8 (2)	7 (3)	0.644
PIP (cmH_2O)	Pre	11.4 (1.1)	11.0 (1.2)	0.339
	Post	11.4 (1.0)	11.1 (1.1)	0.478
$P_E'CO_2$ (mmHg)	Pre	35 (2)	34 (2)	0.64
	Post	37 (2)	36 (2)	0.706
$F_E'Iso$ (%)	Pre	1.51 (0.03)	1.5 (0.01)	0.437
	Post	1.50 (0.29)	1.5 (0.01)	0.609
Temperature (℃)	Pre	36.7 (0.9)	36.3 (1.1)	0.333
	Post	36.6 (0.9)	36.1 (1.3)	0.277

Table 2.2 Comparisons of percentage changes in cardiorespiratory variables after the fluid challenge between responders and non-responders. Data are mean (standard deviation). *Significant difference between responders and non-responders ($p < 0.05$). CVP, central venous pressure; VTI, velocity time integral; PPV, pulse pressure variation; PVI, pleth variability index; PIP, peak inspiratory pressure; $P_E'CO_2$, end-tidal partial pressure of carbon dioxide; $F_E'Iso$, end-tidal concentration of isoflurane.

	Responders	**Non-responders**	***p*-Value**
Heart rate (%)	18 (11)	4 (8)	0.003*
Systolic arterial pressure (%)	16 (12)	5 (6)	0.009*
Diastolic arterial pressure (%)	11 (15)	0 (7)	0.048*
Mean arterial pressure (%)	15 (15)	3 (7)	0.016*
Stroke volume index (%)	27 (7)	7 (4)	< 0.001*
Cardiac index (%)	50 (19)	12 (5)	< 0.001*
Central venous pressure (%)	46 (26)	72 (28)	0.028*
VTI (%)	27 (7)	7 (4)	< 0.001*
PPV (%)	-43 (14)	-18 (12)	< 0.001*
PVI (%)	-35 (14)	-12 (13)	0.001*
PIP (%)	0 (2)	1 (2)	0.285
$P_E'CO_2$ (%)	7 (2)	7 (5)	0.993
$F_E'Iso$ (%)	-1 (3)	0 (1)	0.403
Temperature (%)	0 (1)	0 (2)	0.709

ROC curve analysis for CVP, PPV and PVI

The areas under the ROC curve for CVP, PPV, and PVI were 0.56, 0.85, and 0.84 respectively and cutoff values to discriminate between responders and non-responders for PPV and PVI were 11% (sensitivity 79%; specificity 80%) and 9.3% (sensitivity 86%; specificity 70%), respectively (Table 2.3). There were significant differences in the areas under the ROC curve between PPV and CVP (p = 0.038), and between PVI and CVP (p = 0.043), however there was no significant difference in the areas under the ROC curve between PPV and PVI (p = 0.896) (Figure 2.2). Grey zone values were identified ranges of 9-12% [7 out of 24 fluid challenges (29%)] for PPV and 8-12% [8 out of 24 fluid challenges (33%)] for PVI.

Table 2.3 Areas under the receiver operator characteristic (ROC) curves and cutoff values for the prediction of fluid responsiveness. AUC: Area under the curve, 95%CI: 95% confidence interval, CVP: Central venous pressure, PPV: Pulse pressure variation, PVI: Pleth variability index, Sen: Sensitivity, Spe: Specificity.

	AUC	95%CI	*p*-value	Cutoff	Sen (%)	Spe (%)	Grey zone
CVP	0.56	0.32 - 0.81	0.605	2 mmHg	100	20	2 - 4 mmHg
PPV	0.85	0.70 - 1.00	0.004	11 %	78.57	80	9 - 12%
PVI	0.84	0.68 - 1.00	0.005	9.3 %	85.71	70	8 - 12%

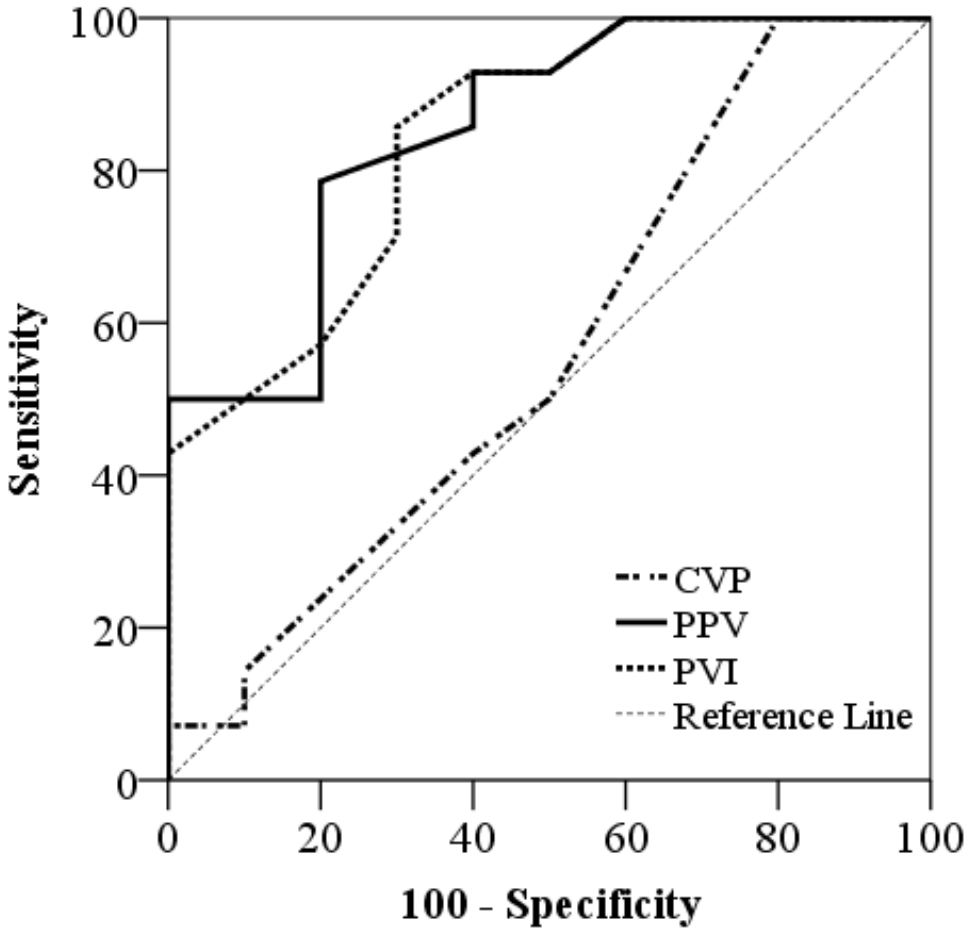

Figure 2.2 Comparison of areas under the receiver operator characteristic (ROC) curve for CVP, PPV and PVI. CVP, central venous pressure; PPV, pulse pressure variation; PVI, pleth variability index.

DISCUSSION

The study showed that PPV and PVI could predict fluid responsiveness more accurately than CVP in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine. This result is consistent with studies in ventilated human patients (Cannesson et al., 2008; Michard et al., 2000; Natalini et al., 2006; Preisman et al., 2005). In people, the cutoff values for PPV and PVI above which patients are considered to respond to the fluid administration are 9.4-17% (Cavallaro et al., 2008) and 9.5-17% (Cannesson et al., 2008; Loupec et al., 2011; Zimmermann et al., 2010) respectively, which is similar to the values in the dogs in this study. The grey zones for PPV and PVI in this study are also similar to those defined in people, where approximately 25% of patients lie within the grey zone of PPV that lies between 9% and 13% (Cannesson et al., 2011). In addition, the areas under the ROC curve for CVP (0.56) showed that it had no predictive value and should probably not be used clinically (Marik & Cavallazzi, 2013). The values in this study are comparable to these values in human studies (Cannesson et al., 2008; Cannesson et al., 2011; Loupec et al., 2011; Marik & Cavallazzi, 2013; Michard et al., 2000; Natalini et al., 2006; Preisman et al., 2005; Zimmermann et al., 2010).

Volume expansion has been the standard treatment for perioperative hypotension in veterinary practice. However, a fluid challenge improves haemodynamics in only 50% of human patients (Kelm et al., 2015; Pinsky & Teboul, 2005). In studies of deeply anaesthetised euvolaemic hypotensive dogs, high-volume rapid administration of an isotonic crystalloid or a colloid did not improve arterial blood pressure (Aarnes et al., 2009; Valverde et al., 2012), indicating that blood pressure was a poor predictor of the haemodynamic response to fluid administration (Muir et al., 2014). The present study also showed that high-volume colloid administration improved haemodynamics in only

14 out of 24 (58%) occasions in healthy anaesthetised dogs and there was no significant difference in blood pressure before the fluid challenge between responders and non-responders. Thus, dynamic parameters for predicting fluid responsiveness should be utilised when volume expansion is considered.

PPV is a percentage change in arterial pulse pressure over a mechanical ventilatory cycle. PPV has been widely accepted as a measure for predicting the fluid responsiveness in patients who are hypotensive during general anaesthesia (Cannesson et al., 2008; Lu, Dong, Xu, Shen, & Zheng, 2014; Natalini et al., 2006), when critically ill (Drvar et al., 2013; Loupec et al., 2011; Michard et al., 2000), and in those undergoing cardiac surgery (Bendjelid, Suter, & Romand, 2004; Preisman et al., 2005), elective coronary artery surgery (Broch et al., 2011), or colorectal surgery (Hood & Wilson, 2011). In fact, peri-anaesthetic goal-directed haemodynamic therapy based on PPV reduced the total amount of fluid administered and the length of stay in hospital, and improved postoperative outcome in human medicine (Forget, Lois, & de Kock, 2010; M. R. Lopes et al., 2007). The present study suggests that PPV can also predict fluid responsiveness in mechanically ventilated dogs. Therefore, the use of PPV may improve postoperative outcomes in dogs, although further studies to test that are required.

The pulse oximeter waveform is generated by blood volume changes in both arterial and venous vessels (J. M. Kim, Arakawa, Benson, & Fox, 1986) using the absorption of light at two different wavelengths. The perfusion index is defined as the ratio between constant absorption and pulsatile absorption, and is equivalent to the amplitude of the plethysmographic waveform. PVI reflects the respiratory variation from the pulse oximeter waveform in mechanically ventilated patients (Cannesson et al., 2008) and has

a good relationship to PPV (Cannesson et al., 2007), although there are differences between the plethysmography and arterial blood pressure waveform. In people, PVI-based goal-directed fluid management reduced the volume of intraoperative fluid infused and reduced intraoperative and postoperative lactate concentrations (Forget et al., 2010). In this study, PVI could predict fluid responsiveness as well as PPV in dogs. Furthermore, PVI can be more easily used in routine veterinary practice than PPV, since PVI can be measured noninvasively and does not require an arterial catheter, although mechanical ventilation is still needed.

Previous human studies of dynamic variables for fluid responsiveness used more than one assessment in the same patient (Natalini et al., 2006; Tavernier, Makhotine, Lebuffe, Dupont, & Scherpereel, 1998). The response to fluid administration can vary in the same patient because of variations in their hydration status, preload, afterload, cardiac contractility and underlying diseases. In this study, 24 fluid challenges were performed in 12 dogs that were anaesthetised twice, 4 weeks apart. The different fluid responsiveness may have been caused by 2 different feeding conditions based on perioperative oral rehydration therapy studies in people (Taniguchi et al., 2014; Taniguchi et al., 2009). Human studies indicated that both body weight and body fluid decreased more in the conventional fasting group than in the group who had perioperative oral solution 3 hours prior to surgery and urine volume significantly increased in the oral solution group compared to the fasting group, even though there were no differences in the volume of bleeding, dose of intraoperative infusion, and use of vasopressors (Taniguchi et al., 2014; Taniguchi et al., 2009). In fact, the VTI changes after fluid challenge in different feeding situations were significantly different in this study. This result showed that dogs were not inherently responders or non-responders, but changed

their responsiveness depending on their haemodynamic condition at the time of anaesthesia. Therefore, the 24 fluid challenges were considered independent in terms of fluid responsiveness in this study.

The appropriate volume, type of fluid and definition of responsiveness differ between studies in people. In adults, 500 mL of colloid has been arbitrarily chosen (Michard & Teboul, 2002). Fluid responsiveness in people has been defined as an increase in VTI or SV of up to 15% or an increase in CO of 10-20% (Michard & Teboul, 2002). Although the appropriate volume and the definition of fluid responsiveness for dogs are unknown, 10 mL/kg of colloid and a 15% increase in VTI were chosen based on extrapolation from human studies (Michard & Teboul, 2002).

Acepromazine was used as a premedication in this study for several reasons. First, it is a common premedication in veterinary practice and its use simulates a general clinical setting. Second, fluid administration during anaesthesia is often considered in the situation of hypotension where acepromazine could be involved. Third, other predications such as dexmedetomidine have been shown to alter PPV independent of volume status (Diniz et al., 2014). Thus, although avoiding premedication may reduce potential confounding factors, it makes extrapolation to the clinical situation more difficult. Acepromazine was considered unlikely to change haemodynamics abruptly during the fluid challenge since the elimination of acepromazine is slow (elimination half-life: 7.1 hours in dogs)(Hashem, Kietzmann, & Scherkl, 1992). However, potential effect of acepromazine on PPV and PVI in dogs have not been investigated. Therefore, further study is warranted.

Propofol is a short acting induction agent and clinical duration of action ranged from 23 to 40 minutes (Morgan & Legge, 1989) after a single injection of propofol in dogs (elimination half-life: 70 minute in dogs)(Reid & Nolan, 1993). As all measurements were initiated at least 1 hour after the induction and the same does of profopol was given into each group, propofol is unlikely to affect and bias the results of this study.

Arrhythmias could alter the values of dynamic parameters (Teboul & Vieillard-Baron, 2005). In people, it is reported that a heart rate/respiratory rate ratio needs to be more than 3.6 in order to measure accurate dynamic parameters (De Backer, Taccone, Holsten, Ibrahimi, & Vincent, 2009). In the current study, the absence of arrhythmias was confirmed by the visualization of ECG. The heart rate/respiratory rate ratio, depending on the heart rate ranging from 72 to 117 beat/minute with the constant respiratory rate of 15 breath/minute, varied between 4.8 and 7.8. However, care is required for clinical extrapolation of this result as opioids can cause bradycardia.

This study has several limitations. First, spontaneous respiratory effort during mechanical ventilation impacts on the measurements of dynamic parameters because of irregular pleural pressure (Teboul & Vieillard-Baron, 2005). Although an absence of spontaneous breathing was confirmed clinically by observing the capnograph waveform, variations in PIP, and the animal's chest movements, it is possible that small respiratory efforts affected values of PPV and PVI because a neuromuscular block agent was not used. Second, the measurement of PPV and PVI can be directly affected by a change in VT (Diaz, Erranz, Donoso, Salomon, & Cruces, 2015; H. K. Kim & Pinsky, 2008), PIP (Kang, Kim, Woo, & Yoon, 2014; Kawazoe et al., 2015), and a pleural pressure (Liu et al., 2016). In this study, the relatively high VT of 15 mL/kg was set in order to avoid a

low VT that may produce unreliable PPV (Oliveira-Costa, Friedman, Vieira, & Fialkow, 2012), but the current study's setting resulted in clinically acceptable PIP and $P_E'CO_2$ (Table 2.1). Although the actual VT was not measured, PIP, oxygen flow (2 L/minute), and $P_E'CO_2$ were almost equal between groups (Table 2.1). Although the pleural pressure could affect values of PPV and PVI directly, it was not measured in this study because the experimental dogs used were clinically healthy without any respiratory problems. Third, it has been shown that PPV does not predict fluid responsiveness in people with increased pulmonary artery pressure (Wyler von Ballmoos et al., 2010) and right ventricular dysfunction (Reuse, Vincent, & Pinsky, 1990). In this study, the cardiac function was evaluated using echocardiography prior to the fluid challenge and no abnormalities were found. However, since some dogs were relatively old, undetected occult cardiac dieses may have influenced our results. Fourth, the change in VTI using echorcardiography was used to differentiate fluid responsiveness in this study. Transthoracic echocardiography provides an accurate measurement of CO with an excellent correlation with thermodilution techniques in dogs (P. C. Lopes et al., 2010). SV is calculated from VTI at the pulmonary artery via pulse Doppler and the cross sectional area of the pulmonic artery (P. C. Lopes et al., 2010). The changes in SV are correlated to VTI variations based on the assumption that the diameter of the pulmonary artery was constant over a 13-minute fluid challenge. Thus, the changes in VTI are directly related to changes in SV and this approach has been used in several human studies (Maizel et al., 2007; Muller et al., 2011). In this study, we used a 15% increase in VTI to differentiate responders from non-responders. Since the variability for the measurement of VTI is reported at approximately 3–8% in people (Lewis, Kuo, Nelson, Limacher, & Quinones, 1984; Moulinier et al., 1991; Oren-Grinberg & Park, 2008), and minor changes in transducer position did not influence Doppler frequency shift integral substantially in

dogs (Steingart et al., 1980), an increase in VTI of 15% or more represents a real effect. However, the normal VTI variation is unknown in dogs.

In conclusion, PPV and PVI predicted fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine, and may be useful to guide fluid administration. When PPV and PVI fall into the grey zone, uncertainty exists and further information should be sought. However, the clinical use of PPV and PVI require tightly controlled conditions such as mechanical ventilation. Further investigation in clinical settings will be required to confirm the clinical utility of these parameters.

ACKNOWLEDGEMENTS

This work was performed at School of Veterinary Science, Massey University, Palmerston North, 4442, New Zealand. This study was supported by a grant from Massey University Research Funds. The authors thank Ms. Karin Weidgraaf and the staff of Massey University nutrition colony research facility for their invaluable assistance in this work. We are also indebted to Dr. Masako Fujiyama and Dr. Tjarda Reints Bok of Companion Animal Hospital, Massey University.

REFERENCES

Aarnes, T. K., Bednarski, R. M., Lerche, P., Hubbell, J. A., & Muir, W. W., 3rd. (2009). Effect of intravenous administration of lactated Ringer's solution or hetastarch for the treatment of isoflurane-induced hypotension in dogs. *Am J Vet Res, 70*(11), 1345-1353. doi:10.2460/ajvr.70.11.1345

Bandt C, B. L., Buckley G, Shih A, Guereiro H, Ricco C, Delaforcade A, Meola D, Thuramalla N. (2012). STATIC VOLUMETRIC MONITORING OF BLOOD VOLUMES IN CANINE HEMORRHAGIC MODEL. *Abstracts from the International Veterinary Emergency and Critical Care Symposium and the European Veterinary Emergency and Critical Care Society Annual Congress, 2012*.

Bendjelid, K., Suter, P. M., & Romand, J. A. (2004). The respiratory change in preejection period: a new method to predict fluid responsiveness. *J Appl Physiol (1985), 96*(1), 337-342. doi:10.1152/japplphysiol.00435.2003

Berkenstadt, H., Friedman, Z., Preisman, S., Keidan, I., Livingstone, D., & Perel, A. (2005). Pulse pressure and stroke volume variations during severe haemorrhage in ventilated dogs. *Br J Anaesth, 94*(6), 721-726. doi:10.1093/bja/aei116

Boyd, J. H., Forbes, J., Nakada, T. A., Walley, K. R., & Russell, J. A. (2011). Fluid resuscitation in septic shock: a positive fluid balance and elevated central venous pressure are associated with increased mortality. *Crit Care Med, 39*(2), 259-265. doi:10.1097/CCM.0b013e3181feeb15

Broch, O., Bein, B., Gruenewald, M., Hocker, J., Schottler, J., Meybohm, P., . . . Renner, J. (2011). Accuracy of the pleth variability index to predict fluid responsiveness depends on the perfusion index. *Acta Anaesthesiol Scand, 55*(6), 686-693. doi:10.1111/j.1399-6576.2011.02435.x

Cannesson, M., Attof, Y., Rosamel, P., Desebbe, O., Joseph, P., Metton, O., . . . Lehot, J. J. (2007). Respiratory variations in pulse oximetry plethysmographic waveform amplitude to predict fluid responsiveness in the operating room. *Anesthesiology, 106*(6), 1105-1111. doi:10.1097/01.anes.0000267593.72744.20

Cannesson, M., Desebbe, O., Rosamel, P., Delannoy, B., Robin, J., Bastien, O., & Lehot, J. J. (2008). Pleth variability index to monitor the respiratory variations in the pulse oximeter plethysmographic waveform amplitude and predict fluid responsiveness in the operating theatre. *Br J Anaesth, 101*(2), 200-206. doi:10.1093/bja/aen133

Cannesson, M., Le Manach, Y., Hofer, C. K., Goarin, J. P., Lehot, J. J., Vallet, B., & Tavernier, B. (2011). Assessing the diagnostic accuracy of pulse pressure variations for the prediction of fluid responsiveness: a "gray zone" approach. *Anesthesiology, 115*(2), 231-241. doi:10.1097/ALN.0b013e318225b80a

Cavallaro, F., Sandroni, C., & Antonelli, M. (2008). Functional hemodynamic monitoring and dynamic indices of fluid responsiveness. *Minerva Anestesiol, 74*(4), 123-135.

Cotton, B. A., Guy, J. S., Morris, J. A., Jr., & Abumrad, N. N. (2006). The cellular, metabolic, and systemic consequences of aggressive fluid resuscitation strategies. *Shock, 26*(2), 115-121. doi:10.1097/01.shk.0000209564.84822.f2

De Backer, D., Taccone, F. S., Holsten, R., Ibrahimi, F., & Vincent, J. L. (2009). Influence of respiratory rate on stroke volume variation in mechanically ventilated patients. *Anesthesiology, 110*(5), 1092-1097. doi:10.1097/ALN.0b013e31819db2a1

Diaz, F., Erranz, B., Donoso, A., Salomon, T., & Cruces, P. (2015). Influence of tidal volume on pulse pressure variation and stroke volume variation during experimental intra-abdominal hypertension. *BMC Anesthesiol, 15*(1), 127.

doi:10.1186/s12871-015-0105-x

Diniz, M. S., Teixeira-Neto, F. J., Candido, T. D., Zanuzzo, F. S., Teixeira, L. R., Klein, A. V., & do Nascimento, P., Jr. (2014). Effects of dexmedetomidine on pulse pressure variation changes induced by hemorrhage followed by volume replacement in isoflurane-anesthetized dogs. *J Vet Emerg Crit Care (San Antonio), 24*(6), 681-692. doi:10.1111/vec.12246

Drvar, Z., Pavlek, M., Drvar, V., Tomasevic, B., Baronica, R., & Peric, M. (2013). [Stroke volume and pulse pressure variation are good predictors of fluid responsiveness in sepsis patients]. *Acta Med Croatica, 67*(5), 407-414.

Forget, P., Lois, F., & de Kock, M. (2010). Goal-directed fluid management based on the pulse oximeter-derived pleth variability index reduces lactate levels and improves fluid management. *Anesth Analg, 111*(4), 910-914. doi:10.1213/ANE.0b013e3181eb624f

Hanley, J. A., & McNeil, B. J. (1983). A method of comparing the areas under receiver operating characteristic curves derived from the same cases. *Radiology, 148*(3), 839-843. doi:10.1148/radiology.148.3.6878708

Hashem, A., Kietzmann, M., & Scherkl, R. (1992). [The pharmacokinetics and bioavailability of acepromazine in the plasma of dogs]. *Dtsch Tierarztl Wochenschr, 99*(10), 396-398.

Holte, K., Jensen, P., & Kehlet, H. (2003). Physiologic effects of intravenous fluid administration in healthy volunteers. *Anesth Analg, 96*(5), 1504-1509, table of contents.

Hood, J. A., & Wilson, R. J. (2011). Pleth variability index to predict fluid responsiveness in colorectal surgery. *Anesth Analg, 113*(5), 1058-1063. doi:10.1213/ANE.0b013e31822c10cd

Kang, W. S., Kim, J. Y., Woo, N. S., & Yoon, T. G. (2014). The influence of different mechanical ventilator settings of peak inspiratory pressure on stroke volume variation in pediatric cardiac surgery patients. *Korean J Anesthesiol, 66*(5), 358-363. doi:10.4097/kjae.2014.66.5.358

Kawazoe, Y., Nakashima, T., Iseri, T., Yonetani, C., Ueda, K., Fujimoto, Y., & Kato, S. (2015). The impact of inspiratory pressure on stroke volume variation and the evaluation of indexing stroke volume variation to inspiratory pressure under various preload conditions in experimental animals. *J Anesth, 29*(4), 515-521. doi:10.1007/s00540-015-1995-y

Kelm, D. J., Perrin, J. T., Cartin-Ceba, R., Gajic, O., Schenck, L., & Kennedy, C. C. (2015). Fluid overload in patients with severe sepsis and septic shock treated with early goal-directed therapy is associated with increased acute need for fluid-related medical interventions and hospital death. *Shock, 43*(1), 68-73. doi:10.1097/shk.0000000000000268

Kim, H. K., & Pinsky, M. R. (2008). Effect of tidal volume, sampling duration, and cardiac contractility on pulse pressure and stroke volume variation during positive-pressure ventilation. *Crit Care Med, 36*(10), 2858-2862. doi:10.1097/CCM.0b013e3181865aea

Kim, J. M., Arakawa, K., Benson, K. T., & Fox, D. K. (1986). Pulse oximetry and circulatory kinetics associated with pulse volume amplitude measured by photoelectric plethysmography. *Anesth Analg, 65*(12), 1333-1339.

Klein, A. V., Teixeira-Neto, F. J., Garofalo, N. A., Lagos-Carvajal, A. P., Diniz, M. S., & Becerra-Velasquez, D. R. (2016). Changes in pulse pressure variation and plethysmographic variability index caused by hypotension-inducing hemorrhage followed by volume replacement in isoflurane-anesthetized dogs. *Am J Vet Res,*

77(3), 280-287. doi:10.2460/ajvr.77.3.280

Kumar, A., Anel, R., Bunnell, E., Habet, K., Zanotti, S., Marshall, S., . . . Parrillo, J. E. (2004). Pulmonary artery occlusion pressure and central venous pressure fail to predict ventricular filling volume, cardiac performance, or the response to volume infusion in normal subjects. *Crit Care Med, 32*(3), 691-699.

Lewis, J. F., Kuo, L. C., Nelson, J. G., Limacher, M. C., & Quinones, M. A. (1984). Pulsed Doppler echocardiographic determination of stroke volume and cardiac output: clinical validation of two new methods using the apical window. *Circulation, 70*(3), 425-431.

Liu, Y., Wei, L. Q., Li, G. Q., Yu, X., Li, G. F., & Li, Y. M. (2016). Pulse Pressure Variation Adjusted by Respiratory Changes in Pleural Pressure, Rather Than by Tidal Volume, Reliably Predicts Fluid Responsiveness in Patients With Acute Respiratory Distress Syndrome. *Crit Care Med, 44*(2), 342-351. doi:10.1097/ccm.0000000000001371

Lobo, D. N., Macafee, D. A., & Allison, S. P. (2006). How perioperative fluid balance influences postoperative outcomes. *Best Pract Res Clin Anaesthesiol, 20*(3), 439-455.

Lopes, M. R., Oliveira, M. A., Pereira, V. O., Lemos, I. P., Auler, J. O., Jr., & Michard, F. (2007). Goal-directed fluid management based on pulse pressure variation monitoring during high-risk surgery: a pilot randomized controlled trial. *Crit Care, 11*(5), R100. doi:10.1186/cc6117

Lopes, P. C., Sousa, M. G., Camacho, A. A., Carareto, R., Nishimori, C. T., Santos, P. S., & Nunes, N. (2010). Comparison between two methods for cardiac output measurement in propofol-anesthetized dogs: thermodilution and Doppler. *Vet Anaesth Analg, 37*(5), 401-408. doi:10.1111/j.1467-2995.2010.00552.x

Loupec, T., Nanadoumgar, H., Frasca, D., Petitpas, F., Laksiri, L., Baudouin, D., . . . Mimoz, O. (2011). Pleth variability index predicts fluid responsiveness in critically ill patients. *Crit Care Med, 39*(2), 294-299. doi:10.1097/CCM.0b013e3181ffde1c

Lu, W., Dong, J., Xu, Z., Shen, H., & Zheng, J. (2014). The pleth variability index as an indicator of the central extracellular fluid volume in mechanically ventilated patients after anesthesia induction: comparison with initial distribution volume of glucose. *Med Sci Monit, 20*, 386-392. doi:10.12659/msm.890073

Maizel, J., Airapetian, N., Lorne, E., Tribouilloy, C., Massy, Z., & Slama, M. (2007). Diagnosis of central hypovolemia by using passive leg raising. *Intensive Care Med, 33*(7), 1133-1138. doi:10.1007/s00134-007-0642-y

Marik, P. E., & Cavallazzi, R. (2013). Does the central venous pressure predict fluid responsiveness? An updated meta-analysis and a plea for some common sense. *Crit Care Med, 41*(7), 1774-1781. doi:10.1097/CCM.0b013e31828a25fd

Marik, P. E., Cavallazzi, R., Vasu, T., & Hirani, A. (2009). Dynamic changes in arterial waveform derived variables and fluid responsiveness in mechanically ventilated patients: a systematic review of the literature. *Crit Care Med, 37*(9), 2642-2647. doi:10.1097/CCM.0b013e3181a590da

Michard, F. (2005). Changes in arterial pressure during mechanical ventilation. *Anesthesiology, 103*(2), 419-428; quiz 449-415.

Michard, F., Boussat, S., Chemla, D., Anguel, N., Mercat, A., Lecarpentier, Y., . . . Teboul, J. L. (2000). Relation between respiratory changes in arterial pulse pressure and fluid responsiveness in septic patients with acute circulatory failure. *Am J Respir Crit Care Med, 162*(1), 134-138. doi:10.1164/ajrccm.162.1.9903035

Michard, F., Lopes, M. R., & Auler, J. O., Jr. (2007). Pulse pressure variation: beyond the

fluid management of patients with shock. *Crit Care, 11*(3), 131. doi:10.1186/cc5905

Michard, F., & Teboul, J. L. (2002). Predicting fluid responsiveness in ICU patients: a critical analysis of the evidence. *Chest, 121*(6), 2000-2008.

Morgan, D. W., & Legge, K. (1989). Clinical evaluation of propofol as an intravenous anaesthetic agent in cats and dogs. *Vet Rec, 124*(2), 31-33.

Moulinier, L., Venet, T., Schiller, N. B., Kurtz, T. W., Morris, R. C., Jr., & Sebastian, A. (1991). Measurement of aortic blood flow by Doppler echocardiography: day to day variability in normal subjects and applicability in clinical research. *J Am Coll Cardiol, 17*(6), 1326-1333.

Muir, W. W., Ueyama, Y., Pedraza-Toscano, A., Vargas-Pinto, P., Delrio, C. L., George, R. S., . . . Hamlin, R. L. (2014). Arterial blood pressure as a predictor of the response to fluid administration in euvolemic nonhypotensive or hypotensive isoflurane-anesthetized dogs. *J Am Vet Med Assoc, 245*(9), 1021-1027. doi:10.2460/javma.245.9.1021

Muller, L., Toumi, M., Bousquet, P. J., Riu-Poulenc, B., Louart, G., Candela, D., . . . Lefrant, J. Y. (2011). An increase in aortic blood flow after an infusion of 100 ml colloid over 1 minute can predict fluid responsiveness: the mini-fluid challenge study. *Anesthesiology, 115*(3), 541-547. doi:10.1097/ALN.0b013e318229a500

Natalini, G., Rosano, A., Taranto, M., Faggian, B., Vittorielli, E., & Bernardini, A. (2006). Arterial versus plethysmographic dynamic indices to test responsiveness for testing fluid administration in hypotensive patients: a clinical trial. *Anesth Analg, 103*(6), 1478-1484. doi:10.1213/01.ane.0000246811.88524.75

Oliveira-Costa, C. D., Friedman, G., Vieira, S. R., & Fialkow, L. (2012). Pulse pressure variation and prediction of fluid responsiveness in patients ventilated with low tidal volumes. *Clinics (Sao Paulo), 67*(7), 773-778.

Oren-Grinberg, A., & Park, K. W. (2008). Assessment of myocardial systolic function by TEE. *Int Anesthesiol Clin, 46*(2), 31-49. doi:10.1097/AIA.0b013e31811f45e7

Pinsky, M. R., & Teboul, J. L. (2005). Assessment of indices of preload and volume responsiveness. *Curr Opin Crit Care, 11*(3), 235-239.

Preisman, S., Kogan, S., Berkenstadt, H., & Perel, A. (2005). Predicting fluid responsiveness in patients undergoing cardiac surgery: functional haemodynamic parameters including the Respiratory Systolic Variation Test and static preload indicators. *Br J Anaesth, 95*(6), 746-755. doi:10.1093/bja/aei262

Ray, P., Le Manach, Y., Riou, B., & Houle, T. T. (2010). Statistical evaluation of a biomarker. *Anesthesiology, 112*(4), 1023-1040. doi:10.1097/ALN.0b013e3181d47604

Reid, J., & Nolan, A. M. (1993). Pharmacokinetics of propofol in dogs premedicated with acepromazine and maintained with halothane and nitrous oxide. *J Vet Pharmacol Ther, 16*(4), 501-505.

Reuse, C., Vincent, J. L., & Pinsky, M. R. (1990). Measurements of right ventricular volumes during fluid challenge. *Chest, 98*(6), 1450-1454.

Ricco C, H.-G. N., Shih A, Bandt C, Pavlisko N, Killos M & Queiroz P. (2012). Pulse pressure variation in a model of hemorrhagic shock in mechanically ventilated dogs. *Abstracts at the American College of Veterinary Anesthesia and Analgesia Annual Meeting, 9th September, 2012, San Antonio, TX, USA*.

Ricco C, H.-G. N., Shih A, Pavlisko N, Killos M & Queiroz P. (2012). Pleth variability index derived from the Radical-7 Masimo pulse oximeter as a non-invasive indicator of circulating volume changes in dogs. *Abstracts at the American College of Veterinary Anesthesia and Analgesia Annual Meeting, 9th September,*

2012, San Antonio, TX, USA.

Rosenberg, A. L., Dechert, R. E., Park, P. K., & Bartlett, R. H. (2009). Review of a large clinical series: association of cumulative fluid balance on outcome in acute lung injury: a retrospective review of the ARDSnet tidal volume study cohort. *J Intensive Care Med, 24*(1), 35-46. doi:10.1177/0885066608329850

Sasaki, K., Mutoh, T., Mutoh, T., Kawashima, R., & Tsubone, H. (2016). Electrical velocimetry for noninvasive cardiac output and stroke volume variation measurements in dogs undergoing cardiovascular surgery. *Vet Anaesth Analg*. doi:10.1111/vaa.12380

Steingart, R. M., Meller, J., Barovick, J., Patterson, R., Herman, M. V., & Teichholz, L. E. (1980). Pulsed doppler echocardiographic measurement of beat-to-beat changes in stroke volume in dogs. *Circulation, 62*(3), 542-548.

Taguchi, H., Ichinose, K., Tanimoto, H., Sugita, M., Tashiro, M., & Yamamoto, T. (2011). Stroke volume variation obtained with Vigileo/FloTrac system during bleeding and fluid overload in dogs. *J Anesth, 25*(4), 563-568. doi:10.1007/s00540-011-1175-7

Taniguchi, H., Sasaki, T., Fujita, H., Kobayashi, H., Kawasaki, R., Goloubev, M., . . . Yoshikawa, T. (2014). Modified ERAS protocol using preoperative oral rehydration therapy: outcomes and issues. *J Anesth, 28*(1), 143-147. doi:10.1007/s00540-013-1769-3

Taniguchi, H., Sasaki, T., Fujita, H., Takamori, M., Kawasaki, R., Momiyama, Y., . . . Goto, T. (2009). Preoperative fluid and electrolyte management with oral rehydration therapy. *J Anesth, 23*(2), 222-229. doi:10.1007/s00540-009-0743-6

Tavernier, B., Makhotine, O., Lebuffe, G., Dupont, J., & Scherpereel, P. (1998). Systolic pressure variation as a guide to fluid therapy in patients with sepsis-induced hypotension. *Anesthesiology, 89*(6), 1313-1321.

Teboul, J. L., & Vieillard-Baron, A. (2005). Clinical value of pulse pressure variations in ARDS. Still an unresolved issue? *Intensive Care Med, 31*(4), 499-500. doi:10.1007/s00134-005-2587-3

Valverde, A., Gianotti, G., Rioja-Garcia, E., & Hathway, A. (2012). Effects of high-volume, rapid-fluid therapy on cardiovascular function and hematological values during isoflurane-induced hypotension in healthy dogs. *Can J Vet Res, 76*(2), 99-108.

Vieillard-Baron, A., Chergui, K., Rabiller, A., Peyrouset, O., Page, B., Beauchet, A., & Jardin, F. (2004). Superior vena caval collapsibility as a gauge of volume status in ventilated septic patients. *Intensive Care Med, 30*(9), 1734-1739. doi:10.1007/s00134-004-2361-y

Wyler von Ballmoos, M., Takala, J., Roeck, M., Porta, F., Tueller, D., Ganter, C. C., . . . Jakob, S. M. (2010). Pulse-pressure variation and hemodynamic response in patients with elevated pulmonary artery pressure: a clinical study. *Crit Care, 14*(3), R111. doi:10.1186/cc9060

Youden, W. J. (1950). Index for rating diagnostic tests. *Cancer, 3*(1), 32-35.

Zimmermann, M., Feibicke, T., Keyl, C., Prasser, C., Moritz, S., Graf, B. M., & Wiesenack, C. (2010). Accuracy of stroke volume variation compared with pleth variability index to predict fluid responsiveness in mechanically ventilated patients undergoing major surgery. *Eur J Anaesthesiol, 27*(6), 555-561. doi:10.1097/EJA.0b013e328335fbd1

Chapter 3

Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs

Sano H & Chambers P
School of Veterinary Science, Massey University, Palmerston North, New Zealand

This chapter was published in *Veterinary Anaesthesia and Analgesia*:

Sano H & Chambers JP: Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Veterinary Anaesthesia and Analgesia.* 44(5):1057-1067, 2017. doi: 10.1016/j.vaa.2016.11.014.

PREFACE

As described in **Chapter 1**, the mini-fluid challenge requires the assessment of change in stroke volume (SV) or cardiac output (CO). A significant increase in SV following the mini-fluid challenge indicates that the animal is a fluid responder. The thermodilution technique with a pulmonary artery catheter is considered the clinical standard for the measurement of CO. However, the placement of a pulmonary artery catheter is clinically invasive and challenging in veterinary practice and unfortunately, it has been shown in people that the pulmonary artery catheter itself casued significant morbidity and mortality. Therefore, investigation of a non-invasive method to measure CO or evaluate changes in SV is necessary. The mini-fluid challenge does not require the measurement of absolute number of SV or CO but assessment of changes in SV or CO. The study in **Chapter 3** tested the ability of pulse wave transit time (PWTT) to detect changes in SV and to estimate CO compared to thermodilution technique in isoflurane-anaesthetised dogs.

This chapter was published in *Veterinary Anaesthesia and Analgesia* and is formatted for the style of the journal.

ROLES OF EACH OF THE AUTHORS

Sano H: Primarily contributed to the study conception and design, collected and interpreted data with statistical analysis, prepared the manuscript and approved the final version to be published.

Chambers P: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

ABSTRACT

Objectives: To evaluate the ability of PWTT to detect changes in SV and to estimate CO compared with the thermodilution technique in isoflurane-anaesthetised dogs.

Study design: Prospective, experimental study.

Animals: Eight adult laboratory dogs.

Methods: The dogs were anaesthetised with isoflurane and mechanically ventilated. Reference CO (TDCO) was measured via a pulmonary artery catheter using the thermodilution technique, and reference SV (TDSV) was calculated. PWTT was calculated as the time from the electrocardiogram (ECG) R-wave peak to the rise point of the pulse oximeter wave. Estimated CO (esCO) was derived from PWTT after calibration with arterial pulse pressure (both non-invasive and invasive methods) and TDCO at the baseline. Haemodynamic changes were induced by administration of phenylephrine (vasoconstriction), high isoflurane (vasodilatation and negative inotropy) and dobutamine (vasodilatation and positive inotropy). Trending between percentage change in PWTT and TDSV was assessed using concordance analysis and receiver operator characteristic (ROC) curve. The agreement between esCO and TDCO was evaluated using the Bland-Altman method.

Results: The direction of percentage change between consecutive PWTT and the corresponding TDSV showed a concordance rate of 95%, with correlation coefficients of -0.86 ($p < 0.001$). Area under the ROC curve for the change in PWTT to detect 15% change in TDSV was 0.91 ($p < 0.001$). TDCO compared to esCO calibrated with invasive and non-invasive blood pressure showed a bias (precision of agreement) of 0.58 (1.54) and 0.57 (1.59) L/minute with a percentage error of ±61% and ±63%, respectively.

Conclusion and clinical relevance: In isoflurane-anaesthetised dogs, PWTT showed a good trending ability to detect 15% changes in SV. This technique is easy to use,

inexpensive, non-invasive and could become routine anaesthetic monitoring. However, the agreement between absolute esCO and TDCO was unacceptable.

Keywords: Cardiac output, stroke volume, pulse oximeter, electrocardiography

Abbreviations

ANOVA	analysis of variance
CO	cardiac output (L/minute)
CVP	central venous pressure (mmHg)
ECG	electrocardiogram
esCO	CO estimated from PWTT (L/minute)
$esCO_{IBP}$	CO estimated from PWTT and calibrated with IBP (L/minute)
$esCO_{NIBP}$	CO estimated from PWTT and calibrated with NIBP (L/minute)
esSV	SV estimated from PWTT (mL)
HR	heart rate (beat/min)
IBP	invasive blood pressure (mmHg)
MAP	mean arterial pressure (mmHg)
NIBP	non-invasive blood pressure (mmHg)
PWTT	pulse wave transit time (msecond)
ROC	receiver operator characteristic
SV	stroke volume (mL)
SVR	systemic vascular resistance (dynes/second/cm^5)
TDCO	CO measured by thermodilution technique (L/minute)
TDSV	SV measured by thermodilution technique (mL)
95%CI	95% confidence interval

INTRODUCTION

Measurement of SV and CO can facilitate cardiovascular management during anaesthesia. Goal-directed therapy with monitoring of CO improved postoperative outcome in high-risk surgical patients (Aya, Cecconi, Hamilton, & Rhodes, 2013; Cecconi et al., 2013; Donati et al., 2007; Gan et al., 2002; Ripolles-Melchor et al., 2016). Assessment of trend in SV and CO is also important for cardiovascular responsiveness to treatments with inotropic, vasoactive and fluid therapy (Guinot et al., 2015; Hasanin, 2015). Therefore, clinically feasible accurate measurement of SV and CO in veterinary practices should be investigated.

The thermodilution technique with a pulmonary artery catheter is considered the clinical standard method to measure CO (TDCO) in dogs based on the Stewart–Hamilton equation since it was described (Fegler, 1954) and tested in dogs using regression analysis, resulting in a good agreement with the Fick method (Hendriks, Schipperheyn, & Quanjer, 1978). However, the placement of a pulmonary artery catheter is clinically invasive and challenging in veterinary practice and associated with increased postoperative complications in human medicine (Harvey et al., 2005; Sakka, Reinhart, Wegscheider, & Meier-Hellmann, 2000). Hence, several minimally invasive methods for CO measurement have been tested in dogs (Bektas et al., 2012; Canfran, Cediel, Sandez, Caro-Vadillo, & Gomez de Segura, 2015; Garofalo et al., 2016; Kutter, Bettschart-Wolfensberger, Romagnoli, & Bektas, 2016; Morgaz et al., 2014).

PWTT is the time from the ECG R-wave peak to the rise point of the pulse oximeter wave (Sugo et al., 2010). The rise point of the pulse wave is defined as the point at which the differentiated pulse wave reached 30% of its peak amplitude (Figure 3.1). PWTT has

proven to be inversely proportional to SV and has a strong correlation with SV in dogs (Sugo et al., 2010) and human volunteers (Ishihara et al., 2004). Based on this relationship, Sugo et al. developed a system to estimate SV (esSV) and CO (esCO) using PWTT (Sugo, Sakai, Terao, Ukawa, & Ochiai, 2012; Sugo et al., 2010). This requires an initial 3-minute period of stable haemodynamics for calibration against another CO measurement system or calibration based on a demographic patient information, and arterial pulse pressure (Ishihara et al., 2004). It is easy to use, inexpensive, minimally invasive and requires only routine anaesthetic monitoring (pulse oximetry, ECG, non-invasive or invasive arterial blood pressure). In a canine experiment, the correlation of esCO compared with CO obtained using electromagnetic flow meters on the aorta was high (r = 0.825); however, the agreement between two methods was not clearly reported (Sugo et al., 2010). Because a high correlation does not always indicate good agreement between the two methods, precision of agreement with percentage error using Bland–Altman analysis should be used to assess the interchangeability of two methods (Critchley, Lee, & Ho, 2010).

Therefore, the aims of this study were: 1) to evaluate the ability of percentage change in PWTT to detect a trend of thermodilution SV (TDSV) correctly, and 2) to evaluate the agreement between TDCO and esCO both over a wide range of CO in isoflurane-anaesthetised dogs.

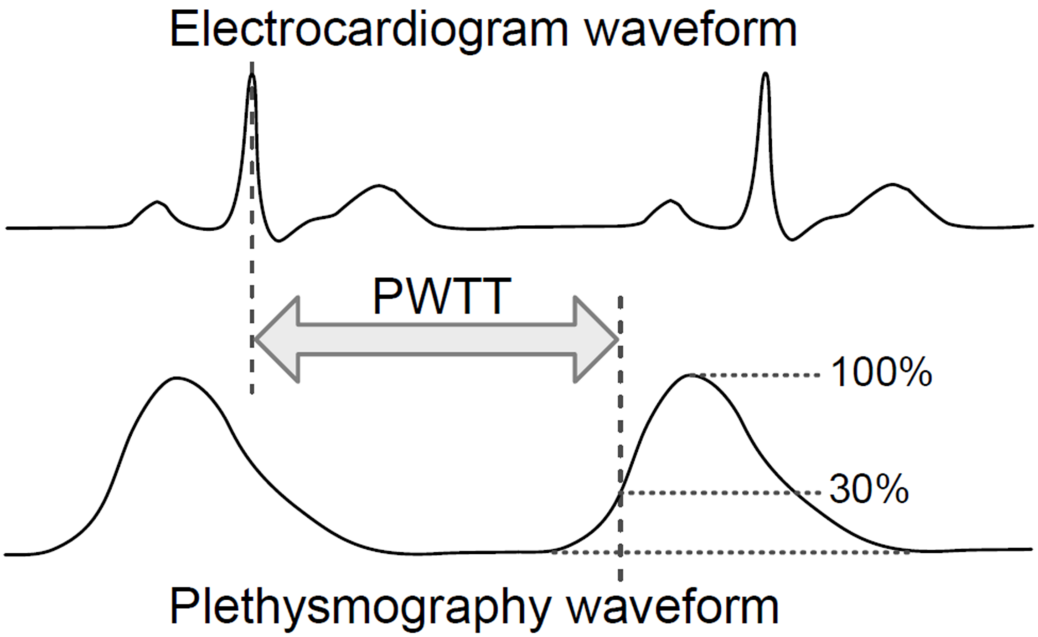

Figure 3.1 Pulse wave transit time (PWTT). PWTT was calculated as the time from the electrocardiogram R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

MATERIALS AND METHODS

The study was approved by the Animal Ethics Committee of Massey University (Protocol Number: 14/112). Eight adult laboratory Foxhounds were used. These eight dogs participated in a concurrent study to validate a non-invasive blood pressure (NIBP) monitor based on the American College of Veterinary Internal Medicine issued guidelines for validation of monitors for NIBP measurement in dogs and cats (Brown et al., 2007). The dogs were determined to be healthy on the basis of history, physical examination, stable bodyweight and blood work analysis.

Anaesthesia and instrumentation

Food was withheld overnight and water was provided *ad libitum*. An 18- or 20-gauge catheter (Optiva I.V. catheter Radiopaque, Smiths Medical International Ltd, London, UK) was placed into a cephalic vein for anaesthetic induction. Anaesthesia was induced with propofol (Propofol, Norbrook NZ Ltd, Auckland, New Zealand) to effect and the trachea was intubated with a cuffed endotracheal tube connected to a circle system with an anaesthetic machine (Excel 80 Anesthesia Machine, Datex-Ohmeda NZ Pty, Auckland, NZ). Anaesthesia was maintained with isoflurane (Isoflurane, Bayer New Zealand Ltd, Auckland, New Zealand) in 2 L/minute of 100% oxygen to achieve 1.50±0.05% end-tidal isoflurane concentration. Mechanical ventilation (Excel 80 system with a 7800 Ventilator, Datex-Ohmeda NZ Pty, Auckland, NZ) was initiated with 12 breath/minute and a tidal volume of 10-15 mL/kg to maintain end-tidal partial pressure of carbon dioxide between 35 and 40 mmHg. A 20- or 22-gauge catheter (Optiva I.V. catheter Radiopaque, Smiths Medical International Ltd, London, UK) was inserted into a dorsal pedal artery to measure invasive blood pressure (IBP). The catheter was connected to an electronic pressure transducer (BD DTXPlus: DTX Plus TNF-R, Becton Dickinson

Critical Care Systems PTE LTD, Singapore) via a noncompliant tube filled with heparinised saline. The transducer was positioned at the level of the manubrium and zeroed to atmospheric pressure. Before the experiment, a 2-point standard calibration was performed with a mercury manometer, and the IBP waveform was checked for stability and consistency. NIBP was measured by oscillometry using a commercial device (Life Scope BSM-3763, Nihon Kohden, Tokyo, Japan). The cuff size was selected according to the manufacturer's guideline (approximately 40% of circumference of the cuff site). The occlusive cuff was placed above the tarsus and at the same height as the transducer. For every measurement, four values were taken with a 20-second interval. The first value was discarded, and the following three consecutive values were averaged to determine a representative value. An 8.5 Fr introducer (Intro-Flex, Edwards Lifesciences LLC, Irvine, CA, USA) was placed in the left jugular vein using a Seldinger technique. A 7.5 Fr × 110 cm Swan-Ganz catheter (Swan-Ganz TD Catheter 131HF7, Edwards Lifesciences LLC, Irvine, CA, USA) was advanced via the introducer into the pulmonary artery, and the correct placement of the catheter was confirmed by characteristic pressure waveforms. ECG, oxygen saturation of arterial haemoglobin, IBP and NIBP were recorded by a multipurpose bedside monitor (Life Scope BSM-3763, Nihon Kohden, Japan), and end-tidal isoflurane concentration and end-tidal partial pressure of carbon dioxide were measured by another monitor (Anesthesia Gas monitor POET IQ, Criticare systems Inc, Waukesha, Wis, USA).

PWTT measurement

PWTT was calculated automatically by averaging 64 consecutive heartbeats (Life Scope BSM-3763, Nihon Kohden, Japan). Average PWTT and heart rate (HR) were based on data retrieved within every 1-second interval. Data with a large variability in PWTT (>20

milliseconds) or pulse amplitude deviating from median values (>30%) during calculation are excluded. In addition, the calculation of PWTT was automatically inhibited when >25% of the 64 beats were excluded in the following conditions: 1) either an ECG or pulse-oximetry pulse wave signal was not obtained and 2) either R wave or the start point of the ascending portion of pulse oximetry wave was not clearly identified (Sugo et al., 2012).

esCO and esSC calculation

Based on the Windkessel model and previous studies (Sugo et al., 2010), esCO and esSV were calculated using the following formula:

$$esCO = HR \times esSV$$

$$esSV = K \times (-0.25 \times PWTT + b)$$

The -0.25 was an individually nondependent constant, calculated as ΔPulse pressure/ΔPWTT obtained as a median from four experimental dogs from the Sugo et al.'s study. *K* and *b*, individual constants, were calculated according to the methods described by Sugo et al. (Sugo et al., 2010) by using the formulae below and were only determined for the first calibration with another CO measurement.

$$K = \frac{SV}{pulse\ pressure}$$

$$b = \frac{SV}{K} + 0.25 \times PWTT$$

Pulse pressure was measured by both IBP and NIBP measurements. The SV for these equations was obtained from TDCO. Therefore, after the determination of constants *K* and *b*, the continuous esCO measurement started using measurement of following PWTT (every 64 beats).

TDCO and TDSV measurement

The thermodilution technique with a Swan-Ganz catheter was used to measure TDCO. Consecutive boluses of 10 mL ice-cold saline were manually injected at the end of expiration by the same operator into the proximal port of the catheter located in the right atrium. The temperature change induced by the fluid bolus was detected by a thermistor-tipped catheter. The TDCO was calculated based on the Stewart-Hamilton equation by the same monitor (Life Scope BSM-3763, Nihon Kohden, Japan). An average of three thermodilution measurements with a variation of less than 10% were used to calculate TDCO. If more than three measurements with less than 10% variation were obtained, the closest values to the mean were used. The TDSV was also calculated by dividing TDCO by the HR.

Haemodynamic states

Three different haemodynamic states were induced by administration of phenylephrine (Phenylephrine, Hospira Inc., NC, USA), isoflurane and dobutamine (Dobutamine, Hameln Pharmaceuticals gmbh, Langes Feld, Germany) in this order (Figure 3.2). For every haemodynamic state, each drug' dosage was increased once after a 10-minute interval. There was also a 30-minute washout period before changing haemodynamic states. Vasoconstriction was induced by 0.5 μg/kg/minute (PH1) and 1 μg/kg/minute (PH2) of phenylephrine. End-tidal isoflurane concentrations of 2.0-2.5% (ISO1) and 2.5-3.0% (ISO2) were used to induce vasodilation and negative inotropy. Vasodilation and positive inotropy was produced by 1 μg/kg/minute (DO1) and 2 μg/kg/minute (DO2) of dobutamine. In each haemodynamic state, TDSV, PWTT, TDCO, esCO with IBP and NIBP calibration ($esCO_{IBP}$ and $esCO_{NIBP}$), HR, mean arterial pressure (MAP), central venous pressure (CVP) and systemic vascular resistance (SVR) were recorded or

calculated. After the calibration, 10 measurements were recorded on each dog including the last baseline after the cessation of the dobutamine infusion.

All dogs were administered 0.2 mg/kg meloxicam (Meloxicam, Boehringer Ingelheim Vetmedica, Inc, St Joseph, Mo, USA) intravenously during recovery.

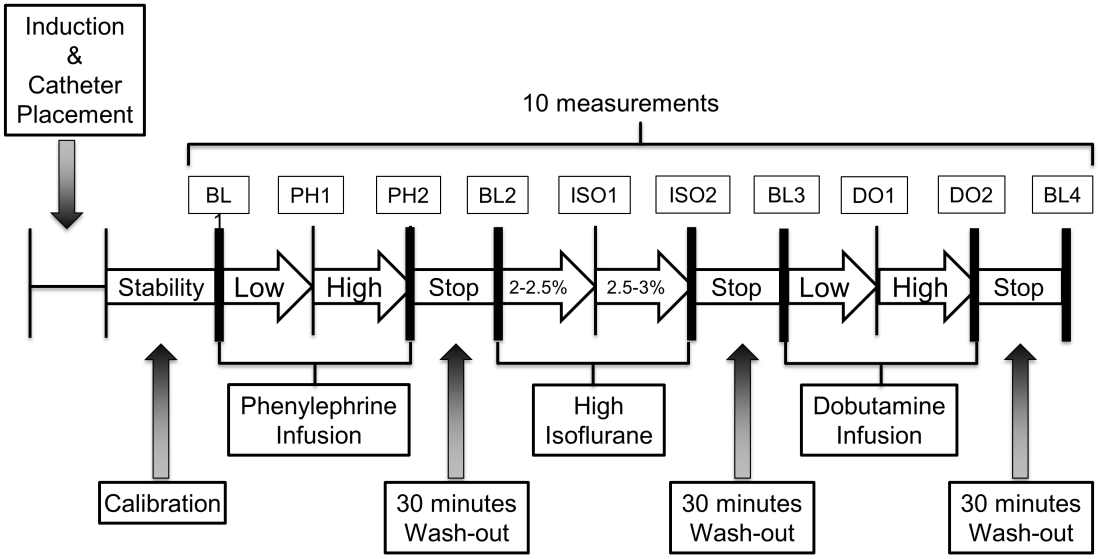

Figure 3.2 Timeline of the study. After the calibration, 10 measurements were taken; before drug administration (baseline1: BL1), during phenylephrine administration (0.5 µg/kg/minute: PH1; 1 µg/kg/minute: PH2), 30 minutes after cessation of PH1 (baseline2: BL2), during high isoflurane administration (2.0-2.5%: ISO1; 2.5-3.0%: ISO2), 30 minutes after cessation of ISO2 (baseline3: BL3), during dobutamine administration (1 µg/kg/minute: DO1; 2 µg/kg/minute: DO2) and 30 minutes after cessation of DO2 (baseline4: BL4).

Statistical analysis

Normality for all variables was assessed with the Shapiro-Wilk test. Haemodynamic variables at each state were compared by an analysis of variance (ANOVA) for repeated measurements followed by a Bonferroni correction.

The ability of PWTT to assess trend in TDSV was evaluated by plotting all consecutive sets of percentage change in PWTT and TDSV on a four-quadrant plot with linear regression analysis. Data points in the lower right and upper left quadrant were considered correct directions of SV between TDSV and PWTT. The concordance rate was calculated as the percentage of the total number of changes that were in the correct directions. Concordance rates of >95% were considered good, those between 90% and 95% marginal (acceptable), and <90% poor (Critchley et al., 2010; Montenij, Buhre, Jansen, Kruitwagen, & de Waal, 2016). In order for percentage change in PWTT to detect more than 15% of change in TDSV, a ROC curve was generated and the area under the ROC curve was calculated to determine a cutoff value with sensitivity and specificity, which was defined by the point on the ROC curve determined by the maximum value of the Youden index (sensitivity + specificity - 1) (Montenij et al., 2016; Youden, 1950). In consideration of random statistical effects, data sets < 15% percentage change in TDSV and lower than the cutoff value of PWTT determined using the ROC curve were excluded from concordance analysis.

Agreement of TDCO and esCO was assessed by Bland–Altman analysis for multiple data points per subject, calculating the mean (bias) and standard deviation (precision of agreement) of the differences (Bland & Altman, 2007). In addition, the bias between TDCO and esCO during baseline, phenylephrine, high isoflurane and dobutamine

administration were compared using one-way repeated measures ANOVA, followed by a Bonferroni correction. The percentage error was calculated as 1.96 times the standard deviation of the mean bias divided by the mean CO. The precision of method of triplicate TDCO measurements was calculated as 2 times the coefficient of error [precision of method = (2 × coefficient of variation)/√ number of replicates]. Data are presented as median (range) or mean ± standard deviation.

RESULTS

There were five intact males and three intact females with a body weight of 28.4 (23.5-36.5) kg and age of 3.2 (2.0-4.5) years included in the study. A total of 80 pairs of triplicate CO measurements were obtained for the TDCO and esCO comparisons and 72 pairs of consecutive percentage change in TDSV and PWTT were calculated from the measurements. Compared to each baseline before drug administration, phenylephrine (PH2) increased TDSV, MAP, CVP and SVR, and decreased PWTT and HR ($p < 0.05$; Table 3.1). High isoflurane (ISO2) reduced TDSV, TDCO, MAP, and SVR, and caused a significant increase in PWTT and HR. Dobutamine (DO2) increased TDSV, TDCO, $esCO_{IBP}$ and $esCO_{NIBP}$, and decreased PWTT, CVP and SVR significantly. The individual nondependent constant in the formula of esCO was -0.58 (-0.05 to -1.04) in this study.

Table 3.1 Cardiovascular parameters recorded in eight isoflurane-anaesthetised dogs before drug administration (baseline1: BL1), during phenylephrine administration (0.5 μg/kg/minute: PH1; 1 μg/kg/minute: PH2), 30 minutes after cessation of PH1 (baseline2: BL2), during high isoflurane administration (2.0-2.5%: ISO1; 2.5-3.0%: ISO2), 30 minutes after cessation of ISO2 (baseline3: BL3), during dobutamine administration (1 μg/kg/minute: DO1; 2 μg/kg/minute: DO2) and 30 minutes after cessation of DO2 (baseline4: BL4)

	BL1	PH1	PH2	BL2	ISO1	ISO2	BL3	DO1	DO2	BL4
TDSV (mL)	34 ± 10	41 ± 15	48 ± 19 [a]	51 ± 17	44 ± 15	34 ± 11 [b]	42 ± 13	64 ± 21 [c]	81 ± 27 [c]	47 ± 12
PWTT (msec)	254 ± 26	239 ± 21 [a]	232 ± 18 [a]	234 ± 19	240 ± 17	253 ± 17 [b]	239 ± 15	217 ± 22 [c]	203 ± 19 [c]	228 ± 16
TDCO (L/minute)	3.9 ± 1.3	3.6 ± 1.3	3.8 ± 1.3	5.0 ± 1.3	4.8 ± 1.5	3.9 ± 1.2 [b]	4.7 ± 1.6	7.3 ± 2.3 [c]	9.9 ± 3.0 [c]	5.6 ± 1.5
$esCO_{IBP}$ (L/minute)	4.4 ± 1.1	3.7 ± 1.1	3.5 ± 0.9	4.2 ± 0.9	4.5 ± 1.1	4.4 ± 1.2	4.6 ± 1.4	5.4 ± 1.6	6.3 ± 1.3 [c]	5.3 ± 1.5
$esCO_{NIBP}$ (L/minute)	4.3 ± 1.1	3.7 ± 1.1	3.4 ± 0.9	4.3 ± 1.0	4.6 ± 1.2	4.4 ± 1.3	4.6 ± 1.4	5.4 ± 1.6	6.3 ± 1.9 [c]	5.3 ± 1.6
HR (beat/minute)	117 ± 18	92 ± 27 [a]	84 ± 24 [a]	102 ± 12	112 ± 11	116 ± 7 [b]	113 ± 17	116 ± 17	126 ± 21	120 ± 16
MAP (mmHg)	78 ± 7	91 ± 13 [a]	120 ± 9 [a]	111 ± 9	82 ± 11 [b]	67 ± 11 [b]	99 ± 14	100 ± 8	94 ± 11	90 ± 12
CVP (mmHg)	5.9 ± 1.3	5.6 ± 0.7	9.2 ± 1.7 [a]	7.5 ± 1.2	7.0 ± 1.4	7.2 ± 1.5	7.0 ± 1.8	5.8 ± 1.2 [c]	5.5 ± 1.5 [c]	5.0 ± 1.2
SVR (dynes/sec/cm^5)	1622 ± 515	2032 ± 477 [a]	2527 ± 631 [a]	1712 ± 327	1284 ± 194 [b]	1286 ± 218 [b]	1686 ± 429	1104 ± 286 [c]	767 ± 219 [c]	1319 ± 430

[a, b, c] Means followed by different superscript letters are significantly different from each baseline ($p < 0.05$). TDSV, thermodilution stroke volume; PWTT, pulse wave transit time; TDCO, thermodilution cardiac output; $esCO_{IBP}$, estimated cardiac output using PWTT calibrated with invasive blood pressure; $esCO_{NIBP}$, estimated cardiac output using PWTT calibrated with non-invasive blood pressure; HR, heart rate; MAP, mean arterial pressure; CVP, central venous pressure; SVR, systemic vascular resistance.

Ability of PWTT to detect trending of TDSV

The area under the ROC curve for the change in PWTT to detect a 15% change in TDSV was 0.91 [95% confidence interval (95%CI), 0.85 - 0.98; $p < 0.001$; Figure 3.3). The cutoff value of the percentage change in PWTT was 2.7% (sensitivity: 86%, specificity: 81%).

Out of 72 sets on the four-quadrant plot, 16 points were excluded because they were located within less than 15% change in TDSV and less than 2.7% change in PWTT for the concordance analysis (Figure 3.4). On the four-quadrant plot, 53 points were detected in the correct directions and three were not. These 3 points were related to phenylephrine administration. Altogether, the concordance rate was 95% and the correlation coefficient was -0.86 (95%CI: -0.92 to -0.77; $p < 0.001$).

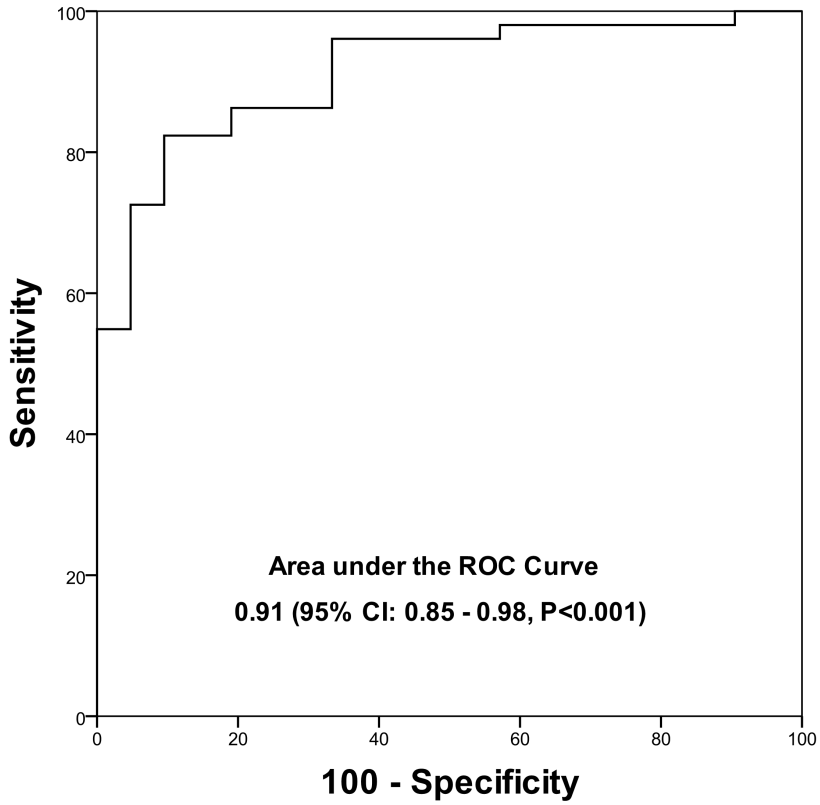

Figure 3.3 Area under the receiver operator characteristic (ROC) curve for the percentage change in pulse wave transit time (PWTT) to detect a 15% change in stroke volume derived from the thermodilution technique (TDSV). The closer to 1.0 of an area under the ROC curve indicates, the more reliable diagnostic method. 95%, 95% confidence interval. The area under the ROC curve and the cutoff value of the percentage change in PWTT to detect 15% change in stroke volume were 0.91 (95%CI, 0.85e0.98; $p < 0.001$) and 2.7% (sensitivity: 86%; specificity: 81%), respectively.

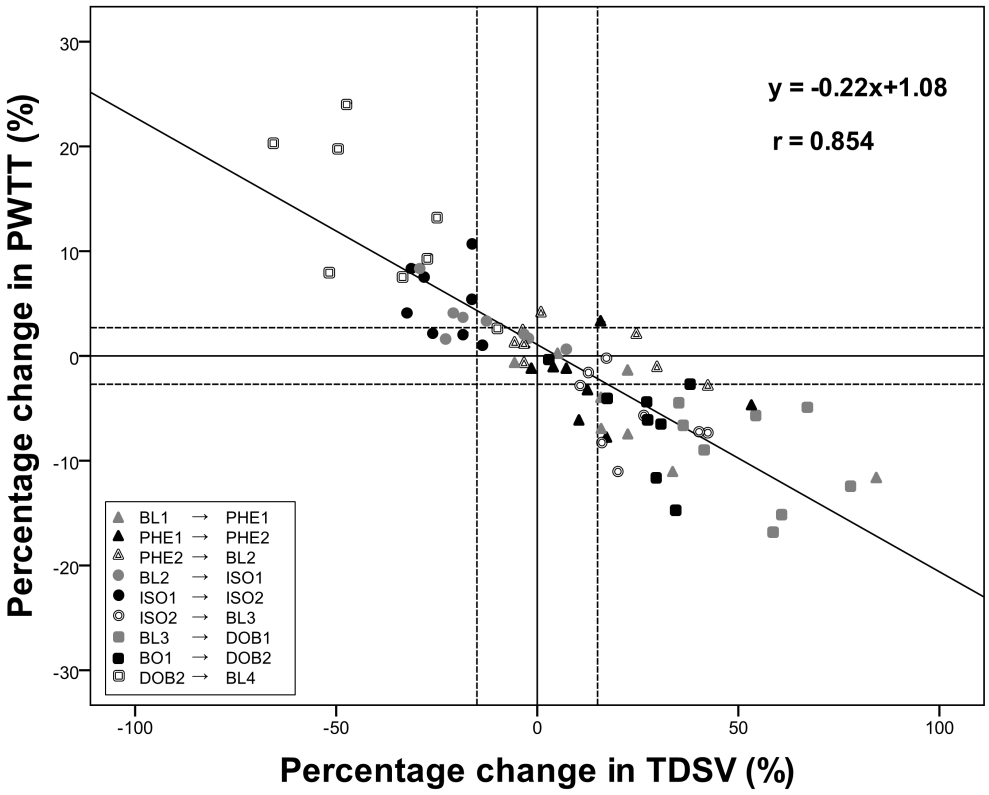

Figure 3.4 Four-quadrant plot with linear regression analysis between percentage changes in PWTT and TDSV. The dotted lines limit an exclusion zone of ±15% for TDSV and 2.7% for PWTT. PWTT, pulse wave transit time; TDSV, stroke volume measured by the thermodilution technique.

Agreement between TDCO and esCO

Bland–Altman plots for the difference between the methods were plotted (Figure 3.5). In 80 data sets, TDCO was 5.21 ± 2.46 L/minute, while $esCO_{IBP}$ and $esCO_{NIBP}$ were 4.63 ± 1.47 and 4.63 ± 1.51 L/minute, respectively. Overall, bias (precision of agreement) and percentage error between TDCO and $esCO_{IBP}$ and, $esCO_{NIBP}$ were 0.58 ± 1.54 L/minute and 61%, and 0.57 ± 1.59 L/minute and 63%, respectively. The bias at DO1 and DO2 were significantly higher than those before dobutamine infusion (p = 0.01 and 0.01 for $esCO_{IBP}$, p = 0.02 and 0.01 for $esCO_{NIBP}$; Table 3.2). After the phenylephrine administration was started, percentage error increased but did not return to baseline when administration was stopped. The precision of method of TDCO was 8.4%.

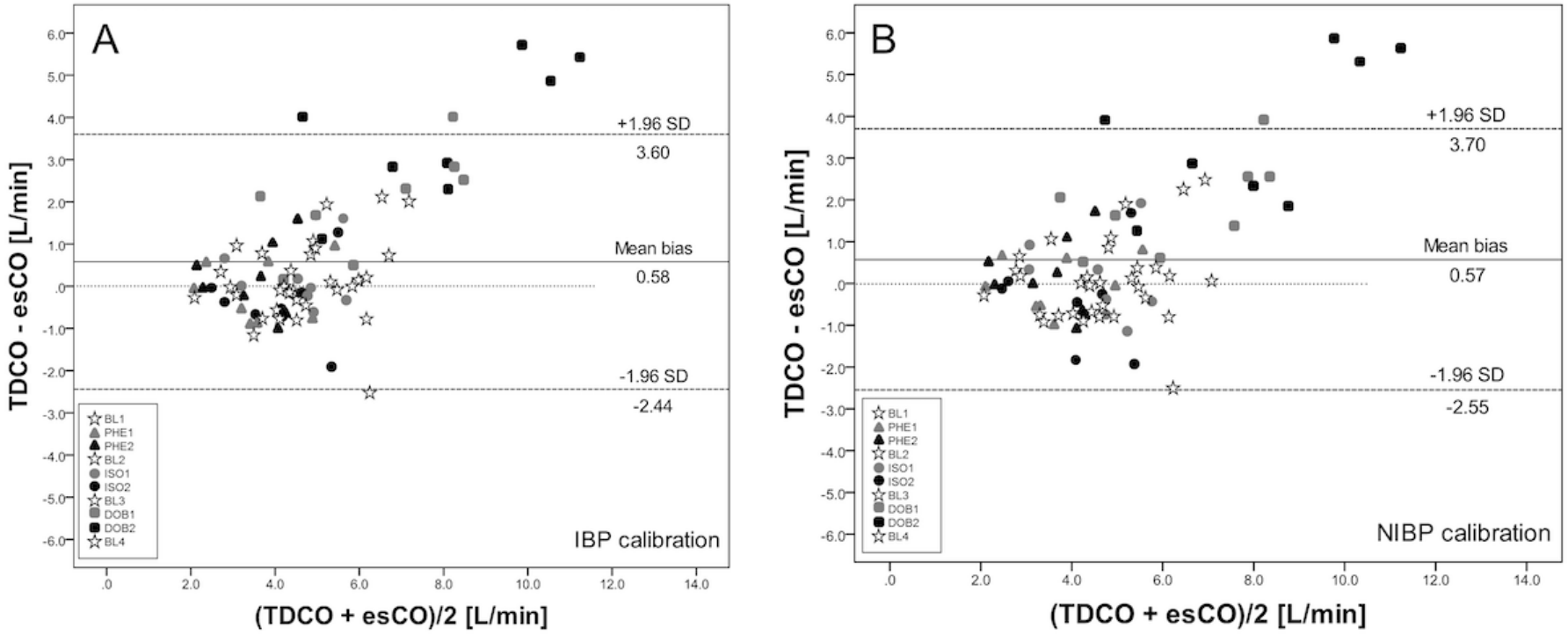

Figure 3.5 Bland-Altman plots for the difference between the methods plotted against their mean. (A) TDCO versus esCO with NIBP calibration, (B) TDCO versus esCO with IBP calibration. The solid line represents the mean bias, the two broken lines indicate the mean bias ± 1.96 standard deviation; the dotted line is the line of equality. esCO, estimated cardiac output based on pulse wave transit time; IBP, invasive blood pressure; NIBP, non-invasive blood pressure; SD, standard deviation; TDCO, cardiac output measured by the thermodilution technique.

DISCUSSION

The PWTT was able to show trends in CO. Changes in PWTT correlated with changes in TDSV with a concordance rate of 95%. The area under the ROC curve to detect 15% change in TDSV was 0.91 with a cutoff value of 2.7%. The precision of esCO compared to TDCO was clinically unacceptable with a percentage error of ±61% ($esCO_{IBP}$) and ±63% ($esCO_{NIBP}$) in isoflurane-anaesthetised dogs.

An original study of a total of 560 data sets in four anaesthetised dogs by Sugo et al. (Sugo et al., 2010) showed a correlation coefficient of -0.71 between ΔPWTT and ΔSV and -0.87 between PWTT and SV, but did not assess the trending ability to track change in the reference SV under different conditions. The current study revealed that change in PWTT has not only a strong negative correlation to change in SV with a good concordance rate, but also high sensitivity and specificity to detect a 15% change in TDSV. This result suggests change in PWTT may be clinically useful as a non-invasive method to track changes in SV in anaesthetised dogs using only routine cardiovascular monitoring.

In the present study, which had a small number of dogs in a broad range of haemodynamic states, PWTT was able to detect changes in TDSV. However, the percentage change in PWTT when SV was low was smaller than when SV was high even though the actual change in SV was the same. Therefore, the cutoff value of PWTT to detect more than 15% change in TDSV should perhaps be less than 2.7% in very low SV states and further studies are necessary. Moreover, the monitor used in this study uses algorithms to exclude artefacts or errors in recording in clinical settings such as a large variability in PWTT (>20 ms) or pulse amplitude deviating from median values (>30%). Thus, arrhythmias

that cause irregular PWTT variability and pulse amplitude are likely to be excluded and their impact on cardiovascular function not detected.

On the four-quadrant plot, three points were plotted in the opposite direction, and all were related to phenylephrine administration (Figure 3.4). Thus, vasoconstriction may affect PWTT. The PWTT is a time, not a volume such as SV. The PWTT is inversely proportional to blood velocity when the distance between the heart and the pulse oximetry probe remain unchanged. The blood velocity (distance per unit time) is proportional to SV (volume per unit time) if the total vessel cross-sectional area remains unchanged. Thus, a change in SV induced by vasoactive drugs or haemorrhage may affect the relationship between PWTT and SV because of changes to the diameter of the vessels. However, a study demonstrated that administration of phenylephrine, pentobarbital and blood removal did not affect the linear relationship between PWTT and SV in anaesthetised dogs (Sugo et al., 2010). The PWTT consists of a pre-ejection period, a time taken for pulse wave from the aorta to the muscular artery (central artery period), and a time from the muscular artery to the further peripheral arteries (peripheral artery period). Sugo et al. suggested that the administration of phenylephrine increases the pre-ejection period because of a rise in afterload, decreases the central artery period because of increased blood pressure caused by vasoconstriction and prolongs the peripheral artery period because of the small diameter of artery. These differential changes resulted in an unchanged linear relationship between PWTT and SV in both Sugo et al.'s study and the current study.

The percentage error of the limit of agreement has been proposed in order to evaluate agreement and interchangeability between two methods and compare studies with different CO ranges (Critchley et al., 2010). Generally, a percentage error of less than

30% is considered clinical acceptable when the reference method is TDCO. The present study showed percentage errors greater than 60%, and Sugo et al. revealed an estimated percentage error greater than 40% in smaller dogs (Sugo et al., 2010). In addition, studies that have evaluated esCO in human subjects at clinical settings, resulted in percentage error of 70% (Ishihara et al., 2004), 54% (Yamada et al., 2012), 38% (Tsutsui et al., 2013) and 44 -60% (Ball et al., 2013) compared with TDCO. However, using a percentage error of 30% has been debated (Cecconi, Rhodes, Poloniecki, Della Rocca, & Grounds, 2009; Hapfelmeier, Cecconi, & Saugel, 2016; Peyton & Chong, 2010). The value is based on the assumption that TDCO has a precision (POM) of ± 20% and is based on the formula (percentage error = $\sqrt{(POM_a)^2 + (POM_b)^2)}$(Cecconi et al., 2009; Hapfelmeier et al., 2016). Thus, if TDCO has a different precision, the acceptable percentage error is altered. If we consider that the precision of TDCO was 8.4% in this study, the estimate of acceptable percentage error becomes ± 12%. Moreover, this discrepancy could be explained not only by the precision of agreement from the Bland-Altman plots, but also by the general variability of CO measurements compared to the true CO values (defined as trueness) (Hapfelmeier et al., 2016). Thus, the precision of each method should be calculated from repeated measurements. Using an estimated percentage error may lead to false conclusions if it does not take into account the methods' variability around the trueness.

All percentage errors in this study were above the determined acceptable percentage errors (± 12%), indicating poor agreement between absolute esCO and TDCO. Therefore, the formula calculating esCO may need to be re-evaluated [esCO = K × (-0.25 × PWTT + b) × HR]. Sugo et al. considered that a constant of -0.25, calculated as ΔPulse pressure/ΔPWTT obtained as a median from four experimental dogs, was individually nondependent. However, actual ΔPulse pressure/ΔPWTT in our eight experimental dogs

were varied and not constant [-0.58 (-0.05 to -1.04)]. Therefore, this individually nondependent constant may need to be dependent.

As a possible limitation of the study, the order of drug administration was not randomised but fixed. Thus, a time effect, or a residual drug effect of prior drugs could affect the results. As shown in Table 3.1, each baseline (BL1, BL2, BL3 and BL4) was different although there was 30-minute washout period before changing haemodynamic states. Lastly, the Bland-Altman plot (Figure 3.5) showed a rise of bias with increasing CO. Thus, the calculation of agreement and percentage error is not ideal and the results may need to be presented as ratios, percentage bias or logarithmic transformation to remove this association.

In conclusion, PWTT showed a good trending ability to be able to detect 15% changes in SV in isoflurane-anaesthetised dogs. This technique is easy to use, inexpensive, non-invasive and could become routine anaesthetic monitoring. However, the agreement between absolute esCO and TDCO was unacceptable. Further development of this method will be required in dogs and other veterinary species.

ACKNOWLEDGEMENTS

The authors thank Dr. Yoshihiro Sugo for his invaluable assistant in this work. They are also indebted to Dr. Joonbum Seo and Dr. Masako Fujiyama of Companion Animal Hospital, Massey University.

REFERENCES

Aya, H. D., Cecconi, M., Hamilton, M., & Rhodes, A. (2013). Goal-directed therapy in cardiac surgery: a systematic review and meta-analysis. *Br J Anaesth, 110*(4), 510-517. doi:10.1093/bja/aet020

Ball, T. R., Tricinella, A. P., Kimbrough, B. A., Luna, S., Gloyna, D. F., Villamaria, F. J., & Culp, W. C., Jr. (2013). Accuracy of noninvasive estimated continuous cardiac output (esCCO) compared to thermodilution cardiac output: a pilot study in cardiac patients. *J Cardiothorac Vasc Anesth, 27*(6), 1128-1132. doi:10.1053/j.jvca.2013.02.019

Bektas, R. N., Kutter, A. P., Hartnack, S., Jud, R. S., Schnyder, M., Matos, J. M., & Bettschart-Wolfensberger, R. (2012). Evaluation of a minimally invasive non-calibrated pulse contour cardiac output monitor (FloTrac/Vigileo) in anaesthetized dogs. *Vet Anaesth Analg, 39*(5), 464-471. doi:10.1111/j.1467-2995.2012.00741.x

Bland, J. M., & Altman, D. G. (2007). Agreement between methods of measurement with multiple observations per individual. *J Biopharm Stat, 17*(4), 571-582. doi:10.1080/10543400701329422

Brown, S., Atkins, C., Bagley, R., Carr, A., Cowgill, L., Davidson, M., . . . Stepien, R. (2007). Guidelines for the identification, evaluation, and management of systemic hypertension in dogs and cats. *J Vet Intern Med, 21*(3), 542-558. doi:10.1892/0891-6640(2007)21[542:gftiea]2.0.co;2

Canfran, S., Cediel, R., Sandez, I., Caro-Vadillo, A., & Gomez de Segura, I. A. (2015). Evaluation of an oesophageal Doppler device for monitoring cardiac output in anaesthetised healthy normotensive dogs. *J Small Anim Pract, 56*(7), 450-455. doi:10.1111/jsap.12362

Cecconi, M., Corredor, C., Arulkumaran, N., Abuella, G., Ball, J., Grounds, R. M., . . . Rhodes, A. (2013). Clinical review: Goal-directed therapy-what is the evidence in surgical patients? The effect on different risk groups. *Crit Care, 17*(2), 209. doi:10.1186/cc11823

Cecconi, M., Rhodes, A., Poloniecki, J., Della Rocca, G., & Grounds, R. M. (2009). Bench-to-bedside review: the importance of the precision of the reference technique in method comparison studies--with specific reference to the measurement of cardiac output. *Crit Care, 13*(1), 201. doi:10.1186/cc7129

Critchley, L. A., Lee, A., & Ho, A. M. (2010). A critical review of the ability of continuous cardiac output monitors to measure trends in cardiac output. *Anesth Analg, 111*(5), 1180-1192. doi:10.1213/ANE.0b013e3181f08a5b

Donati, A., Loggi, S., Preiser, J. C., Orsetti, G., Munch, C., Gabbanelli, V., . . . Pietropaoli, P. (2007). Goal-directed intraoperative therapy reduces morbidity and length of hospital stay in high-risk surgical patients. *Chest, 132*(6), 1817-1824. doi:10.1378/chest.07-0621

Fegler, G. (1954). MEASUREMENT OF CARDIAC OUTPUT IN ANÆSTHETIZED ANIMALS BY A THERMO-DILUTION METHOD. *Quarterly Journal of Experimental Physiology and Cognate Medical Sciences, 39*(3), 153-164. doi:10.1113/expphysiol.1954.sp001067

Gan, T. J., Soppitt, A., Maroof, M., el-Moalem, H., Robertson, K. M., Moretti, E., . . . Glass, P. S. (2002). Goal-directed intraoperative fluid administration reduces length of hospital stay after major surgery. *Anesthesiology, 97*(4), 820-826. Retrieved from http://ovidsp.tx.ovid.com/ovftpdfs/FPDDNCGCMCCNIB00/fs046/ovft/live/gv023/00000542/00000542-200210000-00012.pdf

Garofalo, N. A., Teixeira-Neto, F. J., Rodrigues, J. C., Cerejo, S. A., Aguiar, A. J., & Becerra-Velasquez, D. R. (2016). Comparison of Transpulmonary Thermodilution and Calibrated Pulse Contour Analysis with Pulmonary Artery Thermodilution Cardiac Output Measurements in Anesthetized Dogs. *J Vet Intern Med*. doi:10.1111/jvim.13984

Guinot, P. G., Bernard, E., Deleporte, K., Petiot, S., Dupont, H., & Lorne, E. (2015). Mini-fluid challenge can predict arterial pressure response to volume expansion in spontaneously breathing patients under spinal anaesthesia. *Anaesth Crit Care Pain Med, 34*(6), 333-337. doi:10.1016/j.accpm.2015.06.007

Hapfelmeier, A., Cecconi, M., & Saugel, B. (2016). Cardiac output method comparison studies: the relation of the precision of agreement and the precision of method. *J Clin Monit Comput, 30*(2), 149-155. doi:10.1007/s10877-015-9711-x

Harvey, S., Harrison, D. A., Singer, M., Ashcroft, J., Jones, C. M., Elbourne, D., . . . Rowan, K. (2005). Assessment of the clinical effectiveness of pulmonary artery catheters in management of patients in intensive care (PAC-Man): a randomised controlled trial. *Lancet, 366*(9484), 472-477. doi:10.1016/s0140-6736(05)67061-4

Hasanin, A. (2015). Fluid responsiveness in acute circulatory failure. *J Intensive Care, 3*, 50. doi:10.1186/s40560-015-0117-0

Hendriks, F. F., Schipperheyn, J. J., & Quanjer, P. H. (1978). Thermal dilution measurement of cardiac output in dogs using an analog computer. *Basic Res Cardiol, 73*(5), 459-468.

Ishihara, H., Okawa, H., Tanabe, K., Tsubo, T., Sugo, Y., Akiyama, T., & Takeda, S. (2004). A new non-invasive continuous cardiac output trend solely utilizing routine cardiovascular monitors. *J Clin Monit Comput, 18*(5-6), 313-320.

Kutter, A. P., Bettschart-Wolfensberger, R., Romagnoli, N., & Bektas, R. N. (2016). Evaluation of agreement and trending ability between transpulmonary thermodilution and calibrated pulse contour and pulse power cardiac output monitoring methods against pulmonary artery thermodilution in anesthetized dogs. *J Vet Emerg Crit Care (San Antonio)*. doi:10.1111/vec.12439

Montenij, L. J., Buhre, W. F., Jansen, J. R., Kruitwagen, C. L., & de Waal, E. E. (2016). Methodology of method comparison studies evaluating the validity of cardiac output monitors: a stepwise approach and checklist. *Br J Anaesth, 116*(6), 750-758. doi:10.1093/bja/aew094

Morgaz, J., Granados Mdel, M., Munoz-Rascon, P., Dominguez, J. M., Fernandez-Sarmiento, J. A., Gomez-Villamandos, R. J., & Navarrete, R. (2014). Comparison of thermodilution, lithium dilution, and pulse contour analysis for the measurement of cardiac output in 3 different hemodynamic states in dogs. *J Vet Emerg Crit Care (San Antonio), 24*(5), 562-570. doi:10.1111/vec.12219

Peyton, P. J., & Chong, S. W. (2010). Minimally invasive measurement of cardiac output during surgery and critical care: a meta-analysis of accuracy and precision. *Anesthesiology, 113*(5), 1220-1235. doi:10.1097/ALN.0b013e3181ee3130

Ripolles-Melchor, J., Casans-Frances, R., Espinosa, A., Abad-Gurumeta, A., Feldheiser, A., Lopez-Timoneda, F., & Calvo-Vecino, J. M. (2016). Goal directed hemodynamic therapy based in esophageal Doppler flow parameters: A systematic review, meta-analysis and trial sequential analysis. *Rev Esp Anestesiol Reanim*. doi:10.1016/j.redar.2015.07.009

Sakka, S. G., Reinhart, K., Wegscheider, K., & Meier-Hellmann, A. (2000). Is the placement of a pulmonary artery catheter still justified solely for the measurement of cardiac output? *J Cardiothorac Vasc Anesth, 14*(2), 119-124.

Sugo, Y., Sakai, T., Terao, M., Ukawa, T., & Ochiai, R. (2012). The comparison of a novel continuous cardiac output monitor based on pulse wave transit time and echo Doppler during exercise. *Conf Proc IEEE Eng Med Biol Soc, 2012*, 236-239. doi:10.1109/embc.2012.6345913

Sugo, Y., Ukawa, T., Takeda, S., Ishihara, H., Kazama, T., & Takeda, J. (2010). A novel continuous cardiac output monitor based on pulse wave transit time. *Conf Proc IEEE Eng Med Biol Soc, 2010*, 2853-2856. doi:10.1109/iembs.2010.5626343

Tsutsui, M., Araki, Y., Masui, K., Kazama, T., Sugo, Y., Archer, T. L., & Manecke, G. R., Jr. (2013). Pulse wave transit time measurements of cardiac output in patients undergoing partial hepatectomy: a comparison of the esCCO system with thermodilution. *Anesth Analg, 117*(6), 1307-1312. doi:10.1213/ANE.0b013e3182a44c87

Yamada, T., Tsutsui, M., Sugo, Y., Sato, T., Akazawa, T., Sato, N., . . . Takeda, J. (2012). Multicenter study verifying a method of noninvasive continuous cardiac output measurement using pulse wave transit time: a comparison with intermittent bolus thermodilution cardiac output. *Anesth Analg, 115*(1), 82-87. doi:10.1213/ANE.0b013e31824e2b6c

Youden, W. J. (1950). Index for rating diagnostic tests. *Cancer, 3*(1), 32-35.

Chapter 4

Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs

---

Sano H, Fujiyama M, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P

School of Veterinary Science, Massey University, Palmerston North, New Zealand

This chapter was published in *Journal of Veterinary Emergency and Critical Care:*

Sano H, Fujiyama M, Wightman P, Cave NJ, Gieseg MA, Johnson CB, Chambers P: Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. *Journal of Veterinary Emergency and Critical Care.* 29(4):391-398, 2019. doi: 10.1111/vec.12860.

This work was also presented as an oral presentation at *12th* World Congress of Veterinary Anaesthesiology, Kyoto, Japan 2015: 166.

This study was Supported by Massey University Research Funds.

PREFACE

Chapter 2 investigated the ability of pulse pressure variation (PPV) and pleth variability index (PVI) to predict fluid responsiveness in mechanically ventilated anaesthetised dogs. However, use of these dynamic parameters requires tightly controlled clinical settings. Therefore, alternative methods for prediction of fluid responsiveness should be investigated in dogs. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. Since **Chapter 3** proved pulse wave transit time (PWTT) can be used as an indicator of SV, **Chapter 4** investigated changes in PWTT induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. The study in **Chapter 4** was conducted concurrently using the same fluid challenge in **Chapter 2**.

This chapter was published in *Journal of Veterinary Emergency and Critical Care* and is formatted for the style of the journal.

ROLES OF EACH OF THE AUTHORS

Sano H: Primarily contributed to the study conception and design, collected and interpreted data with statistical analysis, prepared the manuscript and approved the final version to be published.

Fujiyama M: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Wightman P: Contributed to ultrasound performance and prepared the manuscript.

Cave NJ: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Gieseg MA: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Johnson C: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Chambers P: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

ABSTRACT

Objective: To investigate whether percentage changes in pulse wave transit time (ΔPWTT) induced by mini-fluid challenges predict fluid responsiveness in mechanically ventilated anaesthetised dogs.

Design: Prospective experimental trial.

Setting: University teaching hospital.

Animals: Twelve Harrier hounds.

Intervention: Each dog was anaesthetised with propofol and isoflurane after premedication with acepromazine, mechanically ventilated and had a fluid challenge. This was repeated 4 weeks later. The fluid challenge, 10 mL/kg of colloid administration over 13 minutes, consisted of 3 intermittent mini-fluid challenges (1 mL/kg of each over a minute) with a minute interval, and the remaining colloid administration (7 mL/kg) over 7 minutes.

Measurements and Main Results: Percentage change in velocity time integral of pulmonary arterial flow by echocardiography was calculated as an indication of change in SV. Fluid responsiveness was defined as percentage change in velocity time integral ≥ 15% after 10 mL/kg colloid. Dogs responded on 14 fluid challenges and did not on 10. After 1, 2, 3, and 10 mL/kg of fluid challenge, $\Delta PWTT_{1, 2, 3, 10}$ were measured. Receiver operator characteristic (ROC) curves were generated and areas under ROC curve were calculated for $\Delta PWTT_{1, 2, 3}$. A grey zone approach was used to identify the clinically inconclusive range. The area under the ROC curve for $\Delta PWTT_3$ was 0.91 (p = 0.001). Cutoff value for $\Delta PWTT_3$ was -2.5% (sensitivity: 86%, specificity: 90%). The grey zone for $\Delta PWTT_3$ was identified as between -2.9% and -1.9% for which fluid responsiveness could not be predicted reliably in 6 out of 24 fluid challenges.

Conclusions: In mechanically ventilated anaesthetised dogs given a mini-fluid challenge of 3 mL/kg of colloid, ΔPWTT could predict fluid responsiveness although the grey zone should be considered.

Keywords: Frank-Starling, dogs, fluid therapy, pulse wave analysis, and stroke volume

Abbreviations

SV	stroke volume (mL)
CO	cardiac output (mL/minute)
ΔSV	percentage change in SV (%)
PWTT	pulse wave transit time (msecond)
ECG	electrocardiogram
PPV	pulse pressure variation (%)
PVI	pleth variability index (%)
ΔPWTT	percentage change in PWTT (%)
VTI	velocity time integral (cm)
ΔVTI	percentage change in VTI (%)
$\Delta VTI_{1,2,3,10}$	ΔVTI after 1, 2, 3, and 10 mL/kg colloid administration (%)
$\Delta PWTT_{1,2,3,10}$	ΔPWTT after 1, 2, 3, and 10 mL/kg colloid administration (%)
ROC	receiver operator characteristic
95%CI	95% confidence interval

Chapter 4

INTRODUCTION

Perioperative fluid management is crucial in order to maintain haemodynamics. Hypovolaemia reduces stroke volume (SV) and cardiac output (CO), resulting in a decrease in peripheral blood perfusion to tissues (Murakawa & Kobayashi, 1988), while fluid overload causes tissue oedema without a further increase in SV and CO (Holte, Jensen, & Kehlet, 2003), both of which impair tissue oxygenation. Both hypovolaemia and hypervolaemia can be contributory factors to postoperative complications, prolonged length of hospital stay, organ failure and mortality (Boyd, Forbes, Nakada, Walley, & Russell, 2011; Lobo, Macafee, & Allison, 2006; Murphy et al., 2009; Rosenberg, Dechert, Park, & Bartlett, 2009). Therefore, fluids should only be administered to patients who are likely to respond, but should not for the others.

Prediction of fluid responsiveness has been investigated to improve haemodynamic management in anaesthesia and intensive care unit. In the past, static parameters such as central venous pressure and ventricular end diastolic volume were used to estimate preload for prediction of fluid responsiveness (Dellinger et al., 2004; Mohrman & Heller, 2006), however, numerous studies showed that they are poor predictors for fluid responsiveness in people and dogs (Bandt C BL, 2012; Berkenstadt et al., 2005; Sasaki, Mutoh, Mutoh, Kawashima, & Tsubone, 2016; Taguchi et al., 2011) because the preload is not only determined by cardiac filling pressures but also by the compliance of the heart and venous tone (Marik & Cavallazzi, 2013). Meanwhile, dynamic parameters based on heart-lung interactions during positive ventilation such as pulse pressure variation and SV variation have been shown to predict fluid responsiveness more accurately than the static parameters in dogs (**Chapter 2**) (Sano et al., 2018; Taguchi et al., 2011). But they require a highly controlled

environment in only specific types of patients (Marik & Lemson, 2014), because they are highly likely to be affected by not only the preload but also other factors such as hear rate (De Backer, Taccone, Holsten, Ibrahimi, & Vincent, 2009), respiratory rate (De Backer et al., 2009), pleural pressure (Liu et al., 2016), and tidal volume (Diaz, Erranz, Donoso, Salomon, & Cruces, 2015; H. K. Kim & Pinsky, 2008). These confounding variables may reduce clinical application. Therefore, further investigations for assessment of fluid responsiveness are required.

The mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in SV after a small loading dose of fluid. Theoretically, the change in SV of a heart at the steep portion of the Frank-Starling curve will be greater than at the plateau portion after both the mini-fluid challenge and fluid challenge (Figure 4.1). In addition, the greater the increase in SV after the mini-fluid challenge, the more we can expect a similar increase in SV after the fluid challenge. Therefore, the magnitude of the change in SV after a small fluid loading could predict responsiveness to a larger fluid bolus. Studies have shown that a mini-fluid challenge could predict fluid responsiveness in both mechanically ventilated and spontaneously breathing people (Guinot et al., 2014; Muller et al., 2011).

.

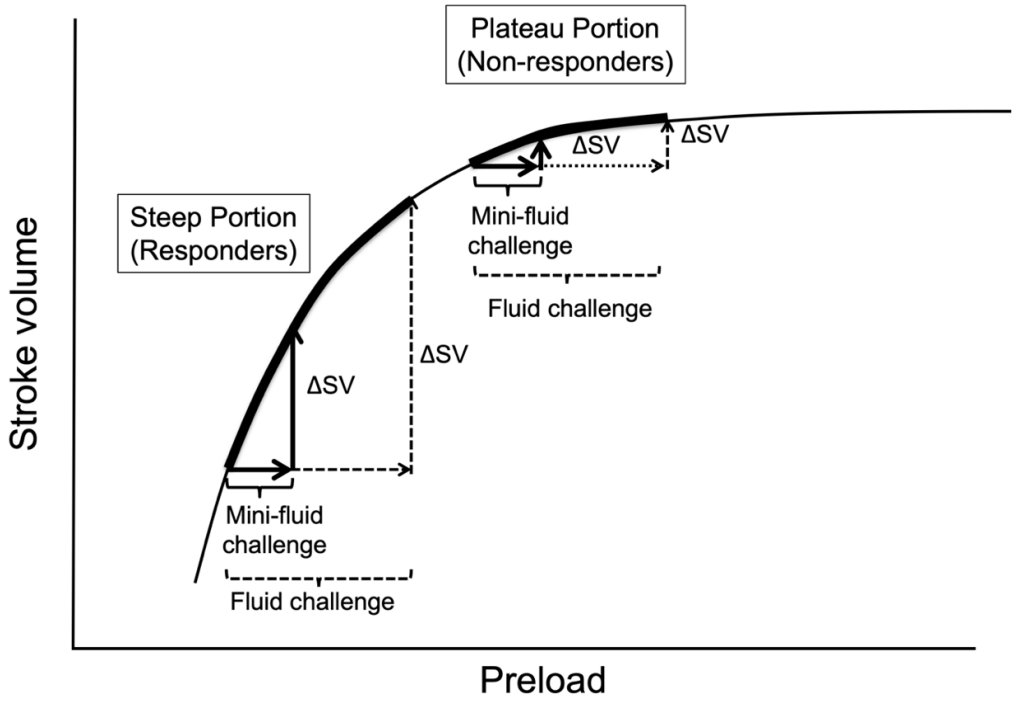

Figure 4.1 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

A good trending ability or accurate method of measuring SV or CO is essential when mini-fluid challenges are performed, because the change in SV induced by the mini-fluid challenge

is smaller than the classical fluid challenge (Figure 4.1). The studies evaluating a mini-fluid challenge in people used either transthoracic echocardiography or thoracic impedance cardiography (Guinot et al., 2014; Muller et al., 2011). Echocardiography has a good trending ability for SV or CO (K. Kim, Kwok, Chang, & Han, 2004; Lorne et al., 2014) although it does not accurately measure the absolute values of CO (K. Kim et al., 2004). Access to this technique is limited in veterinary practices because it needs a trained echocardiography expert with an ultrasound machine and an expensive monitoring device. Therefore, practical and accurate methods of SV or CO measurement are required for the mini-fluid challenge in veterinary practices.

The pulse wave transit time (PWTT) is the time from the electrocardiogram (ECG) R-wave peak to the rise point of the pulse oximeter wave of the same cardiac cycle (Sugo et al., 2010). The rise point of the pulse wave is defined as the point at which the differentiated pulse wave reached 30% of its peak amplitude. PWTT was reported to be inversely proportional to SV and has a strong negative correlation with SV (**Chapter 3**) (Ishihara et al., 2004; Sano & Chambers, 2017; Sugo, Sakai, Terao, Ukawa, & Ochiai, 2012; Sugo et al., 2010). Percentage change in PWTT (ΔPWTT) had a good trending ability to detect changes in SV measured by thermodilution technique over a wide range of CO in isoflurane-anaesthetized dogs (**Chapter 3**)(Sano & Chambers, 2017). Importantly, the PWTT can be easily measured using inexpensive, non-invasive and routine anaesthetic monitoring equipment (ECG and pulse oximeter). Thus, the Δ PWTT could be used for the assessment of a mini-fluid challenge in dogs.

Chapter 4

The aims of this study were 1) to determine what volume of fluid is required for a mini-fluid challenge to reliably predict fluid responsiveness; and 2) to obtain the best cutoff value of Δ PWTT after a mini-fluid challenge in mechanically ventilated anaesthetised dogs. The hypothesis was that ΔPWTT following a mini-fluid challenge could predict fluid responsiveness in mechanically ventilated anaesthetised dogs.

MATERIALS AND METHODS

We have previously described the protocol approved by the animal ethics committee of Massey University (Protocol ID: 14/113) for anaesthetising the dogs, and measuring haemodynamic variables in response to a fluid challenge (**Chapter 2**)(Sano et al., 2018). The study described here was conducted concurrently, using the same fluid challenge.

Briefly, twelve Harrier hounds (7 female, 5 male) with median (range) weight of 24.7 (22.1-31.0) kg and median (range) age of 6 (3-9) years old were used. Their health status was confirmed by history, physical examination, stable bodyweight and basic blood analysis (packed cell volume, total solids, blood glucose, and blood urea nitrogen). After the dogs were premedicated with 0.05 mg/kg of acepromazine (Acezine 2, Ethical Agents Ltd, Manukau, NZ) intramuscularly, anaesthesia was induced with propofol (Repose, Norbrook NZ Ltd, Auckland, NZ) to effect and maintained with an end-tidal isoflurane (Attane, Bayer Animal health New Zealand, Auckland, NZ) concentration of 1.5 ± 0.1% in 100% oxygen (2 L/minute) throughout the study. Dogs were positioned in right lateral recumbency and mechanically ventilated with volume-controlled mode (Excel 80 system with a 7800 Ventilator, Datex-Ohmeda NZ Pty, Auckland, NZ) with a tidal volume of 15 mL/kg at a respiratory rate of 15 breaths/minute.

Basic measurements

Heat rate, oxygen saturation, invasive arterial blood pressure measured via the dorsal pedal artery, central venous pressure measured in the cranial vena cava via the left jugular vein, and rectal temperature were recorded by a multipurpose bedside monitor (Life Scope BSM-

3763, Nihon Kohden, Tokyo, Japan). Transducers (BD DTXPlus: DTX Plus TNF-R, Becton Dickinson Critical Care Systems PTE LTD, Singapore) for the invasive arterial blood pressure and central venous pressure were zeroed to atmospheric pressure and positioned at the level of the manubrium. The end-tidal isoflurane concentration and partial pressure of end-tidal carbon dioxide were measured by a commercial monitor (Anesthesia Gas monitor POET IQ, Criticare systems Inc, Waukesha, WI, USA). Velocity time integral (VTI) of main pulmonary arterial flow was determined using transthoracic echocardiography (Toshiba Xario 200 ultrasound unite with a 2–5 MHz phased array transducer, Toshiba, Tokyo, Japan) by the same ultrasound expert, and percentage change in VTI (ΔVTI) was calculated from VTI before and after fluid administration as an indication of ΔSV described by previous studies (Muller et al., 2011; Sano et al., 2018). Minor changes in transducer position did not influence Doppler frequency shift integral substantially in dogs (Steingart et al., 1980).

PWTT measurement

The PWTT was calculated automatically and averaged over 64 consecutive heart beats by the multi-purpose bedside monitor (Life Scope BSM-3763, Nihon Kohden, Tokyo, Japan). Average PWTT and heart rate were updated every second. Data with a large variability in PWTT (>20 milliseconds) or pulse amplitude deviating from median values (>30%) during calculation were excluded. In addition, the calculation of PWTT was automatically inhibited when >25% of the 64 beats met the following conditions: (1) either ECG or pulse-oximetry pulse wave signal was not obtained; (2) either R wave or the start point of the ascending portion of pulse-oximetry wave was not clearly identified (Sugo et al., 2012). In this study, ΔPWTT was calculated as:

$$\Delta PWTT(\%) = \frac{PWTT\ after - PWTT\ before}{PWTT\ before} \times 100$$

ΔPWTT is a percentage change in PWTT (**Chapter 3**)(Sano & Chambers, 2017) and derived from PWTT before and after fluid administration. In this study, ΔVTIΔ and ΔPWTT after 1, 2, 3 and 10 mL/kg of fluid administration were expressed as $\Delta VTI_{1, 2, 3, 10}$ and $\Delta PWTT_{1, 2, 3, 10}$, respectively.

Fluid challenge and mini-fluid challenge

Each dog was anaesthetised for a fluid challenge [10 mL/kg of colloid (Voluven 6%, Fresenius Kabi Australia Pty Limited, Pymble NSW, AU) over 13 minutes] twice, 4 weeks apart. Thus, a total of 24 fluid challenges were included in this study, as we have previously described (**Chapter 2**)(Sano et al., 2018).

Forty-five minutes after the induction of anaesthesia, 15 minutes was allocated without any stimulation in order to confirm cardiopulmonary stability. Baseline measurements were performed before the colloid administration and every minute for 5 minutes. The highest and lowest values were excluded, and the other three values were averaged to give a representative value. The first mini-fluid challenge (1 mL/kg) was infused over a minute and stopped. Thirty seconds after discontinuation of the first fluid challenge, VTI and PWTT were measured every 10 seconds for 30 seconds and these three values were averaged to give a representative value. The second and third mini-fluid challenges (1 mL/kg each) with respective measurements were repeated as for the first challenge. Lastly the remaining fluid (7 mL/kg) was administered over 7 minutes (total 10 mL/kg over 13 minutes). One minute

after the end of fluid challenge, measurements were made every minute for five minutes as for the baseline measurement. Responders were defined as having more than a 15% increase in ΔVTI after the fluid challenge.

On recovery, dogs received 0.2 mg/kg of meloxicam (Metacam, Boehringer Ingelheim Vetmedica Inc, St Joseph, MO, USA) intravenously.

Statistics

All data are presented as the mean (standard deviation) unless otherwise stated. Normality was assessed with the Shapiro-Wilk test. For each cardiovascular variable, we used a linear mixed model for repeated measurements with time and fluid responsiveness as fixed factors with an interaction term for time and responsiveness. Comparisons of percentage change in variables between responders and non-responders were assessed using an unpaired Student's t-test or a Mann-Whitney U test where appropriate.

Receiver operator characteristic (ROC) curves were generated for $\Delta VTI_{1,2,3}$ and $\Delta PWTT_{1,2,3}$ (Hanley & McNeil, 1983). Clinically acceptable area under ROC curve to predict fluid responsiveness is defined as less than 0.75 (Fan, Upadhye, & Worster, 2006). The best cutoff values to discriminate responders and non-responders were defined as the value closest to the Youden index that maximizes the sum of the sensitivity and specificity minus 1 (Ray, Le Manach, Riou, & Houle, 2010; Youden, 1950). To determine a clinically useful range (below which patients are unlikely to respond, and above which they are expected to respond), the grey zone was calculated (Cannesson et al., 2011) Youden index determination was then

conducted for each bootstrapped population, resulting in a set of 1000 “optimal” values. The mean value of these optimal values and its 95% confidence interval (95%CI) were then estimated. Thus, the grey zone was defined along with the 95%CI (Cannesson et al., 2011) and considered not precise enough for fluid responsiveness prediction. Data were analysed by statistical software (SPSS statistics 22, IBM, San Jose, CA, USA; MedCalc for Windows version 12.5, MedCalc Software, Oostende, Belgium).

RESULTS

Out of 24 fluid challenges, 14 responders and 10 non-responders were obtained as previously described (**Chapter 2**)(Sano et al., 2018). Heart rate did not change after the mini-fluid challenges but heart rate in responders significantly increased after the fluid challenge (Table 4.1). Mean arterial blood pressure in responders after 2 and 3 mL/kg of mini-fluid challenge were significantly higher than in non-responders, but this seems to be clinically undetectable along with central venous pressure. VTI and PWTT significantly changed after the mini-fluid challenges in responders but not non-responders. Responders had significantly higher PWTT before the fluid challenge than non-responders.

Table 4.1 Cardiovascular variables before and after the mini-fluid challenges (1, 2 and 3 mL/kg) and the fluid challenge (10 mL/kg) in responders and non-responders. Data are mean (standard deviation). HR; heat rate, MAP; mean arterial pressure, CVP; central venous pressure, VTI; velocity time integral, PWTT; pulse wave transit time. Some variables are extracted from the concurrent study (**Chapter 2**)(Sano et al., 2018). $^{*}p < 0.05$ between responders (n = 14) and non-responders (n = 10). $^{\dagger}p < 0.05$ between baseline and either 1, 2, 3 or 10 mL/kg.

	Baseline	1 mL/kg	2 mL/kg	3 mL/kg	10 mL/kg
HR (beats/minute)					
Responders	91 (11)	92 (13)	93 (13)	90 (13)	108 (19)*†
Non-responders	95 (12)	96 (13)	96 (11)	96 (12)	99 (12)
MAP (mmHg)					
Responders	57 (6)	60 (9)	60 (9)*	61 (9)*†	66 (11)*†
Non-responders	55 (4)	57 (5)	57 (5)	57 (4)	57 (4)
CVP (mmHg)					
Responders	4 (2)	4 (2)*	4 (2)*	5 (2)*†	6 (2)*†
Non-responders	3 (1)	4 (1)	4 (1)†	4 (1)†	6 (1)†
VTI (cm)					
Responders	12.0 (1.6)	12.5 (1.6)	12.8 (1.6)*†	13.6 (1.8)*†	15.3 (2.1)*†
Non-responders	13.1 (1.5)	13.1 (1.5)	13.4 (1.6)	13.6 (1.4)†	14.0 (1.5)†
PWTT (msec)					
Responders	265 (13)*	261 (13)*†	259 (13)*†	256 (13)*†	238 (15)*†
Non-responders	252 (22)	249 (23)	248 (23)	248 (22)	245 (25)†

After the fluid challenge, $\Delta PWTT_{10}$ in responders was significantly shorter than in non-responders (Table 4.2). After the mini-fluid challenges, $\Delta VTI\Delta_2$ and ΔVTI_3 were significantly greater, and $\Delta PWTT_3$ was lower in responders than non-responders, but ΔVTI_1, $\Delta PWTT_1$, and $\Delta PWTT_2$ were not (Table 4.2).

Table 4.2 Comparison of $\Delta VTI_{1,2,3,10}$ and $\Delta PWTT_{1,2,3,10}$ after the mini-fluid challenges (1, 2 and 3 mL/kg) and the fluid challenge (10 mL/kg) in responders and non-responders. Data are mean (standard deviation). $\Delta VTI_{1,2,3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 1, 2, 3, and 10 mL/kg of fluid; $\Delta PWTT_{1,2,3,10}$, percentage change in pulse wave transit time after 1, 2, 3, and 10 mL/kg of fluid. Some variables are extracted from the concurrent study (**Chapter 2**)(Sano et al., 2018). $^{*}p < 0.05$ between responders (n = 14) and non-responders (n = 10).

	Responders	**Non-responders**	***p*-Value**
ΔVTI_1 (%)	4.6 (4.5)	0.4 (6.6)	0.108
$\Delta PWTT_1$ (%)	-1.4 (1.8)	-1.1 (2.4)	0.122
ΔVTI_2 (%)	7 (6.2)	2.1 (4.9)	0.045*
$\Delta PWTT_2$ (%)	-2.4 (1.6)	-1.2 (1.9)	0.088
ΔVTI_3 (%)	13.4 (5.8)	4.3 (4.5)	< 0.001*
$\Delta PWTT_3$ (%)	-3.7 (1.3)	-1.3 (1.2)	< 0.001*
ΔVTI_{10} (%)	27 (7)	7 (4)	N/A
$\Delta PWTT_{10}$ (%)	-11 (3)	-3 (2)	< 0.001*

Figure 4.2 showed that $\Delta PWTT_3$ has good correlations with ΔVTI_{10} and ΔVTI_3, indicating that $\Delta PWTT_3$ can predict fluid responsiveness and can be a surrogate parameter of ΔSV. $\Delta PWTT_{10}$ also has a good correlation with ΔVTI_{10}.

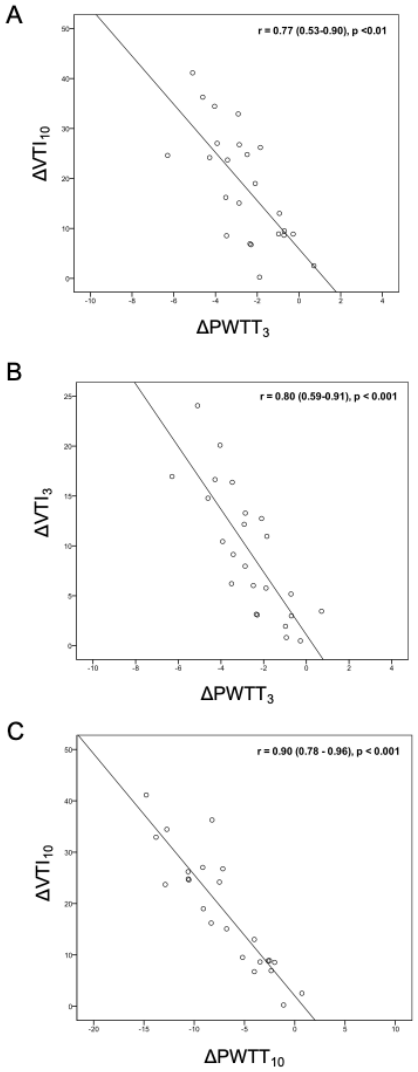

Figure 4.2 Correlation between $\Delta PWTT_3$ and ΔVTI_{10}, ΔVTI_3, and **$\Delta PWTT_{10}$ and ΔVTI_{10}**

$\Delta VTI_{3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 3, and 10 mL/kg of fluid; $\Delta PWTT_{3,10}$, percentage change in pulse wave transit time after 3, and 10 mL/kg of fluid. $^*p < 0.05$

As the cutoff value for Δ VTI_3 [5.8% (sensitivity: 100%, specificity: 90%)] determined by the Youden index (Table 4.3 and Figure 4.3), was within the margin of error for the reported reproducibility of echocardiography (3-8%)(Lewis, Kuo, Nelson, Limacher, & Quinones, 1984; Oren-Grinberg & Park, 2008), a cutoff value of 9.1% (sensitivity: 71%, specificity: 90%) derived from ROC analysis was considered the best value. The cutoff value for $\Delta PWTT_3$ was determined -2.5% (sensitivity: 86%, specificity: 90%) (Table 4.3 and Figure 4.2). With this cutoff value, numbers of true positive, true negative, false negative, and false positive were 11, 9, 3 and 1, respectively. Grey zone values identified for $\Delta PWTT_3$ ranged between -2.9 and -1.9% in 6 out of 24 fluid challenges (Figure 4.4).

Table 4.3 Areas under the receiver operator characteristic (ROC) curves and cutoff values for the prediction of fluid responsiveness. AUC: Area under the curve, 95%CI: 95% confidence interval, $\Delta VTI_{1,2,3,10}$, percentage change in velocity time integral of main pulmonary arterial blood flow after 1, 2, 3, and 10 mL/kg of fluid; $\Delta PWTT_{1,2,3,10}$, percentage change in pulse wave transit time after 1, 2, 3, and 10 mL/kg of fluid. $^{*}p < 0.05$ between AUC and 0.5.

	AUC	95%CI	*p*-Value	Cutoff value (%)	Sensitivity (%)	Specificity (%)
ΔVTI_1 (%)	0.70	0.48 – 0.92	0.08	1.2	64	80
$\Delta PWTT_1$ (%)	0.69	0.46 – 0.92	0.12	-0.8	80	29
ΔVTI_2 (%)	0.75	0.55 – 0.95	0.01	6.1	57	90
$\Delta PWTT_2$ (%)	0.78	0.57 – 0.99	0.02	-0.9	60	71
ΔVTI_3 (%)	0.93	0.79 – 1.00	< 0.01	5.8	100	90
$\Delta PWTT_3$ (%)	0.91	0.80 – 1.00	< 0.01	-2.5	86	90

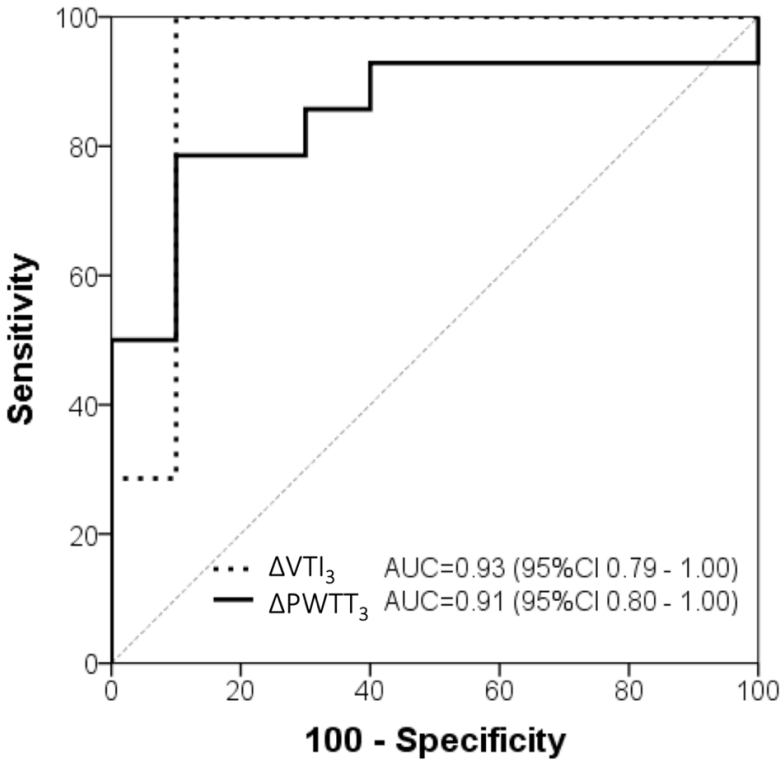

Figure 4.3 Receiver operator characteristic (ROC) curve for ΔVTI_3 and $\Delta PWTT_3$. ΔVTI_3 and $\Delta PWTT_3$, percentage changes in velocity time integral of main pulmonary arterial blood flow and pulse wave transit time after a 3 mL/kg mini-fluid challenge; AUC, area under the curve; 95%CI, 95% confidence interval.

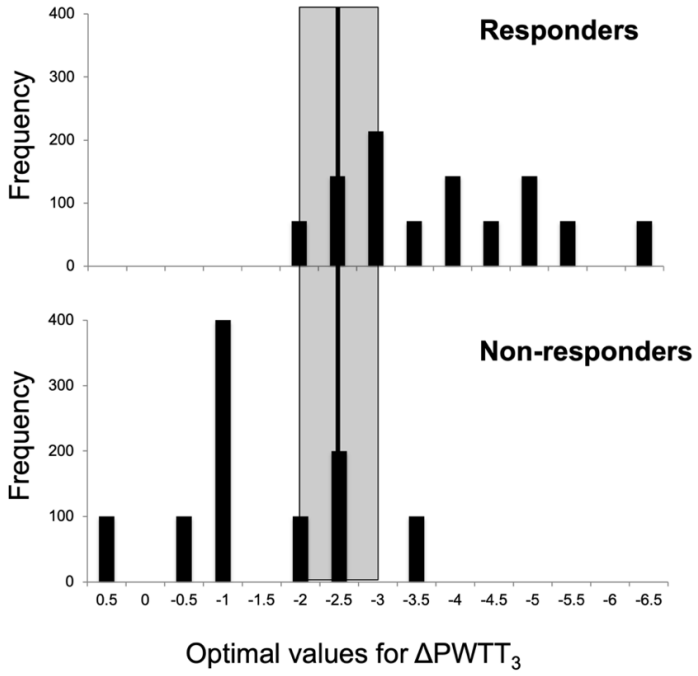

Figure 4.3 The grey zone and the best cutoff value within the optimal cutoff values for $\Delta PWTT_3$. Distribution of the cutoffs for each bootstrapped population (1000 "optimal" values). Grey rectangle (the grey zone), 95% confidence interval for the optimal cutoffs; Black line (the best cutoff value), mean; $\Delta PWTT_3$, percentage changes in pulse wave transit time after a 3 mL/kg mini-fluid challenge.

Discussion

A mini-fluid challenge involves a small amount of fluid administration to assess fluid responsiveness based on ΔSV. The administration of a small amount of fluid is thought to have less adverse effects in non-responders than a typical fluid challenge. This study found the minimal volume of fluid required for a mini-fluid challenge to predict fluid responsiveness reliably was 3 mL/kg. The lowest 95% CI of ΔVTI_2 and $\Delta PWTT_2$ were less than 0.75 although the areas under the ROC curve were more than 0.75, while the areas under the ROC curve of ΔVTI_3 and $\Delta PWTT_3$ met the criteria (Table 4.3 and Figure 4.2). However, clinically significant changes in heart rate, mean arterial pressure, and central venous pressure were not seen after a 3 mL/kg mini-fluid challenge (although some small changes were statistically significant). Thus, these cardiovascular variables were not a good predictor for fluid responsiveness (Muir et al., 2014). The second finding was that the ΔPWTT following a mini-fluid challenge of 3mL/kg can predict fluid responsiveness in mechanically ventilated anaesthetised dogs. A shortening of PWTT of more than 2.5% accurately predicted fluid responsiveness. However, monitoring with other clinical parameters should be taken into account to confirm fluid responsiveness when values of $\Delta PWTT_3$ lie within the grey zone between -2.9 and -1.9% (6/24 of the mini-fluid challenges). This technique can be easily and non-invasively performed with a specific type of anaesthetic monitor, but comprehensive assessment should be considered in clinical practice.

When a mini-fluid challenge with 100 mL fluid administration increased the subaortic VTI by more than 10%, it predicted a 15% increase in ΔVTI after a 500 mL of fluid administration in mechanically ventilated and sedated ICU patients with acute circulatory failure (Muller et al., 2011). Guinot et al measured the SV by thoracic impedance cardiography and

demonstrated that an ΔSV more than 7% after 100 mL of fluid administration was a good predictor of a subsequent more than 15% increase in ΔSV in response to a 500 mL fluid loading in spontaneously breathing patients under spinal anaesthesia (Guinot et al., 2014). Therefore, the mini-fluid challenge can be a clinically useful diagnostic method to predict fluid responsiveness in either mechanically or spontaneously ventilated people. In this study, ΔVTI_3 greater than 9.1% measured by transthoracic echocardiography predicted fluid responsiveness in mechanically ventilated anaesthetised dogs. These results support the ability of the mini-fluid challenge to predict fluid responsiveness in dogs as well as in people. However, the assessment of ΔSV or ΔVTI is challenging in routine practice because it requires echocardiography expertise and special equipment. Therefore, an alternative simple method to assess ΔSV would be useful.

It has been proven that the PWTT has a strong negative correlation with SV in dogs and people (Ishihara et al., 2004; Sano & Chambers, in press; Sugo et al., 2012; Sugo et al., 2010). In a previous study, ΔPWTT showed a good trending ability to detect changes in SV derived from CO measurement by thermodilution with a concordance rate of 95% and a correlation coefficient of -0.86 over a wide range of CO in isoflurane-anaesthetised dogs (**Chapter 3**)(Sano & Chambers, 2017). In the same previous study, a cut off value of ΔPWTT to detect minimum change in SV (more than 15%) measured by thermodilution was -2.7% which is close to the cutoff values of -2.5% to discriminate responders and non-responders in the current study. These results indicate ΔPWTT can be used as a surrogate parameter of ΔSV for a mini-fluid challenge in anaesthetised dogs.

The multi-purpose bedside monitor (Life Scope BSM-3763, Nihon Kohden, Tokyo, Japan) in this study used algorithms to calculate stable PWTT values to exclude artefacts or errors in recording in clinical settings such as a large variability in PWTT or pulse amplitude deviating from median values. Thus, the effects of arrhythmias that cause irregular PWTT variability and amplitude are likely to be excluded. Moreover, if more than 25% of the 64 measured beats were excluded due to arrhythmias or distortion of either ECG or pulse-oximetry pulse wave signals, the calculation of PWTT was automatically stopped. Therefore, this technique may not be useful when either abnormal ECG and pulse-oximetry waveforms, or extremely critical conditions are present.

There were few studies to assess the fluid responsiveness using dynamic parameters based on heart-lung interactions during positive ventilation in dogs. Our previous study showed that more than 11% (sensitivity: 79%, specificity: 80%) of pulse pressure variation and more than 9.3% (sensitivity: 86%, specificity: 70%) of pleth variability index could predict fluid responders in mechanically ventilated isoflurane-anaesthetised dogs prior to a 10 mL/kg of fluid challenge (**Chapter 2**)(Sano et al., 2018). The other study revealed that either more than 4 mmHg or 4.5% of systolic pressure variation could predict fluid responsiveness in mechanically ventilated anaesthetised dogs prior to a 3 mL/kg of crystalloids (Rabozzi & Franci, 2014). However, it is impractical to use these results in veterinary clinical practice because the dynamic parameters require tightly controlled conditions in specific cardiorespiratory settings (Marik & Lemson, 2014). This means that animals need to be anaesthetised and mechanically ventilated. In contrast to those dynamic parameters, ΔPWTT after a mini-fluid challenge is applicable in clinical practice because it needs only ECG and pulse oximeter.

This study had several major limitations described in our previous report (**Chapter 2**)(Sano et al., 2018). First, the 24 individual anaesthetics were not truly independent, but two fluid challenge studies were performed in each dog, in which they responded the same way or changed their responsiveness depending on their haemodynamic and hydration condition at the time of anaesthesia. Therefore, as in our previous study, we considered each study of fluid responsiveness as statistically independent. Second, the small number of measurements is a limitation and calculating a grey zone or ROC with so few values is undesirable. The results should be reassessed in larger studies. Third, the variability for the measurement of ΔVTI by echocardiography may compromise the results because the normal VTI variation is unknown in dogs, although it was reported to be approximately 3–8% in people (Lewis et al., 1984; Oren-Grinberg & Park, 2008; Tavernier, Makhotine, Lebuffe, Dupont, & Scherpereel, 1998). Fourth, the appropriate volume to test fluid responsiveness and definition of fluid responsiveness in dogs are unknown. In adult, 500mL of colloid has been arbitrarily chosen (Michard & Teboul, 2002) and fluid responsiveness in people has been defined as an increase in VTI or SV of up to 15% or an increase in CO of 10 - 20% (Michard & Teboul, 2002). We chose 10mL/kg of colloid and a 15% increase in VTI based on extrapolation from human studies. Fifth, extremely high or low physiological parameters in terms of heart rate, cardiac contractility, arterial stiffness (elastance), blood viscosity and body temperature may influence PWTT, although the study of isoflurane-anaesthetised dogs with wide range of haemodynamics induced by phenylephrine, high isoflurane and dobutamine administration showed ΔPWTT has a good trending ability to detect percentage changes in SV (**Chapter 3**)(Sano & Chambers, 2017). Lastly this result cannot be transferred directly to non-ventilated

and awake dogs, as the dogs were anaesthetised and mechanically ventilated in this study. A further clinical study is required in spontaneously breathing dogs.

In conclusion, a 3 mL/kg of mini-fluid challenge could predict fluid responsiveness based on ΔPWTT in mechanically ventilated anaesthetised dogs. However, monitoring with other clinical parameters should be taken into account to confirm fluid responsiveness when values of $\Delta PWTT_3$ lies within the grey zone between -2.9 and -1.9% (6/24 of the mini-fluid challenges). Since the calculation of $\Delta PWTT_3$ requires only ECG and pulse oximeter data, this technique could be utilized in routine veterinary anaesthetic practice.

ACKNOWLEDGEMENTS

This work was performed at School of Veterinary Science, Massey University, Palmerston North, 4442, New Zealand. This study was supported by a grant from Massey University Research Funds. The authors thank Ms. Karin Weidgraaf and the staff of Massey University nutrition colony research facility for their invaluable assistance in this work. We are also indebted to Dr. Joonbum Seo and Dr. Tjarda Reints Bok of Companion Animal Hospital, Massey University.

REFERENCES

Bandt C BL, B. G., Shih A, Guereiro H, Ricco C, Delaforcade A, Meola D, Thuramalla N. (2012). Static volumetric monitoring of blood volumes in canine hemorrhagic model. *Abstracts from the International Veterinary Emergency and Critical Care Symposium and the European Veterinary Emergency and Critical Care Society Annual Congress*.

Berkenstadt, H., Friedman, Z., Preisman, S., Keidan, I., Livingstone, D., & Perel, A. (2005). Pulse pressure and stroke volume variations during severe haemorrhage in ventilated dogs. *Br J Anaesth, 94*(6), 721-726. doi:10.1093/bja/aei116

Boyd, J. H., Forbes, J., Nakada, T. A., Walley, K. R., & Russell, J. A. (2011). Fluid resuscitation in septic shock: a positive fluid balance and elevated central venous pressure are associated with increased mortality. *Crit Care Med, 39*(2), 259-265. doi:10.1097/CCM.0b013e3181feeb15

Cannesson, M., Le Manach, Y., Hofer, C. K., Goarin, J. P., Lehot, J. J., Vallet, B., & Tavernier, B. (2011). Assessing the diagnostic accuracy of pulse pressure variations for the prediction of fluid responsiveness: a "gray zone" approach. *Anesthesiology, 115*(2), 231-241. doi:10.1097/ALN.0b013e318225b80a

De Backer, D., Taccone, F. S., Holsten, R., Ibrahimi, F., & Vincent, J. L. (2009). Influence of respiratory rate on stroke volume variation in mechanically ventilated patients. *Anesthesiology, 110*(5), 1092-1097. doi:10.1097/ALN.0b013e31819db2a1

Dellinger, R. P., Carlet, J. M., Masur, H., Gerlach, H., Calandra, T., Cohen, J., . . . Levy, M. M. (2004). Surviving Sepsis Campaign guidelines for management of severe sepsis and septic shock. *Crit Care Med, 32*(3), 858-873.

Diaz, F., Erranz, B., Donoso, A., Salomon, T., & Cruces, P. (2015). Influence of tidal volume on pulse pressure variation and stroke volume variation during experimental intra-abdominal hypertension. *BMC Anesthesiol, 15*, 127. doi:10.1186/s12871-015-0105-x

Fan, J., Upadhye, S., & Worster, A. (2006). Understanding receiver operating characteristic (ROC) curves. *Cjem, 8*(1), 19-20.

Guinot, P. G., Bernard, E., Defrancq, F., Petiot, S., Majoub, Y., Dupont, H., & Lorne, E. (2014). Mini-fluid challenge predicts fluid responsiveness during spontaneous breathing under spinal anaesthesia: an observational study. *Eur J Anaesthesiol*. doi:10.1097/eja.0000000000000175

Hanley, J. A., & McNeil, B. J. (1983). A method of comparing the areas under receiver operating characteristic curves derived from the same cases. *Radiology, 148*(3), 839-843. doi:10.1148/radiology.148.3.6878708

Holte, K., Jensen, P., & Kehlet, H. (2003). Physiologic effects of intravenous fluid administration in healthy volunteers. *Anesth Analg, 96*(5), 1504-1509, table of contents.

Ishihara, H., Okawa, H., Tanabe, K., Tsubo, T., Sugo, Y., Akiyama, T., & Takeda, S. (2004). A new non-invasive continuous cardiac output trend solely utilizing routine cardiovascular monitors. *J Clin Monit Comput, 18*(5-6), 313-320.

Kim, H. K., & Pinsky, M. R. (2008). Effect of tidal volume, sampling duration, and cardiac contractility on pulse pressure and stroke volume variation during positive-pressure ventilation. *Crit Care Med, 36*(10), 2858-2862. doi:10.1097/CCM.0b013e3181865aea

Kim, K., Kwok, I., Chang, H., & Han, T. (2004). Comparison of cardiac outputs of major

burn patients undergoing extensive early escharectomy: esophageal Doppler monitor versus thermodilution pulmonary artery catheter. *J Trauma, 57*(5), 1013-1017.

Lewis, J. F., Kuo, L. C., Nelson, J. G., Limacher, M. C., & Quinones, M. A. (1984). Pulsed Doppler echocardiographic determination of stroke volume and cardiac output: clinical validation of two new methods using the apical window. *Circulation, 70*(3), 425-431.

Liu, Y., Wei, L. Q., Li, G. Q., Yu, X., Li, G. F., & Li, Y. M. (2016). Pulse Pressure Variation Adjusted by Respiratory Changes in Pleural Pressure, Rather Than by Tidal Volume, Reliably Predicts Fluid Responsiveness in Patients With Acute Respiratory Distress Syndrome. *Crit Care Med, 44*(2), 342-351. doi:10.1097/ccm.0000000000001371

Lobo, D. N., Macafee, D. A., & Allison, S. P. (2006). How perioperative fluid balance influences postoperative outcomes. *Best Pract Res Clin Anaesthesiol, 20*(3), 439-455. Retrieved from http://ac.els-cdn.com/S1521689606000279/1-s2.0-S1521689606000279-main.pdf?_tid=c45ce5ce-2622-11e5-9f96-00000aab0f26&acdnat=1436436827_cee5abbb13777627cc79d047bf2b6afd

Lorne, E., Mahjoub, Y., Diouf, M., Sleghem, J., Buchalet, C., Guinot, P. G., . . . Dupont, H. (2014). Accuracy of impedance cardiography for evaluating trends in cardiac output: a comparison with oesophageal Doppler. *Br J Anaesth, 113*(4), 596-602. doi:10.1093/bja/aeu136

Marik, P. E., & Cavallazzi, R. (2013). Does the central venous pressure predict fluid responsiveness? An updated meta-analysis and a plea for some common sense. *Crit Care Med, 41*(7), 1774-1781. doi:10.1097/CCM.0b013e31828a25fd

Marik, P. E., & Lemson, J. (2014). Fluid responsiveness: an evolution of our understanding. *Br J Anaesth, 112*(4), 617-620. doi:10.1093/bja/aet590

Michard, F., & Teboul, J. L. (2002). Predicting fluid responsiveness in ICU patients: a critical analysis of the evidence. *Chest, 121*(6), 2000-2008. Retrieved from http://journal.publications.chestnet.org/article.aspx?articleid=1080704

Mohrman, D., & Heller, L. (2006). Central venous pressure: an indicator of circulatory hemodynamics. In D. Mohrman & L. Heller (Eds.), *Lange cardiovascular physiology* (6th ed.). New York, NY: McGraw-Hill.

Muir, W. W., Ueyama, Y., Pedraza-Toscano, A., Vargas-Pinto, P., Delrio, C. L., George, R. S., . . . Hamlin, R. L. (2014). Arterial blood pressure as a predictor of the response to fluid administration in euvolemic nonhypotensive or hypotensive isoflurane-anesthetized dogs. *J Am Vet Med Assoc, 245*(9), 1021-1027. doi:10.2460/javma.245.9.1021

Muller, L., Toumi, M., Bousquet, P. J., Riu-Poulenc, B., Louart, G., Candela, D., . . . Lefrant, J. Y. (2011). An increase in aortic blood flow after an infusion of 100 ml colloid over 1 minute can predict fluid responsiveness: the mini-fluid challenge study. *Anesthesiology, 115*(3), 541-547. doi:10.1097/ALN.0b013e318229a500

Murakawa, K., & Kobayashi, A. (1988). Effects of vasopressors on renal tissue gas tensions during hemorrhagic shock in dogs. *Crit Care Med, 16*(8), 789-792.

Murphy, C. V., Schramm, G. E., Doherty, J. A., Reichley, R. M., Gajic, O., Afessa, B., . . . Kollef, M. H. (2009). The importance of fluid management in acute lung injury secondary to septic shock. *Chest, 136*(1), 102-109. doi:10.1378/chest.08-2706

Oren-Grinberg, A., & Park, K. W. (2008). Assessment of myocardial systolic function by TEE. *Int Anesthesiol Clin, 46*(2), 31-49. doi:10.1097/AIA.0b013e31811f45e7

Rabozzi, R., & Franci, P. (2014). Use of systolic pressure variation to predict the

cardiovascular response to mini-fluid challenge in anaesthetised dogs. *Vet J, 202*(2), 367-371. doi:10.1016/j.tvjl.2014.08.022

Ray, P., Le Manach, Y., Riou, B., & Houle, T. T. (2010). Statistical evaluation of a biomarker. *Anesthesiology, 112*(4), 1023-1040. doi:10.1097/ALN.0b013e3181d47604

Rosenberg, A. L., Dechert, R. E., Park, P. K., & Bartlett, R. H. (2009). Review of a large clinical series: association of cumulative fluid balance on outcome in acute lung injury: a retrospective review of the ARDSnet tidal volume study cohort. *J Intensive Care Med, 24*(1), 35-46. doi:10.1177/0885066608329850

Sano, H., & Chambers, J. P. (2017). Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Vet Anaesth Analg, 44*(5), 1057-1067. doi:10.1016/j.vaa.2016.11.014

Sano, H., & Chambers, P. (in press). Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Veterinary Anaesthesia and Analgesia*.

Sano, H., Seo, J., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2018). Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anesthetized dogs. *J Vet Emerg Crit Care (San Antonio), 28*(4), 301-309. doi:10.1111/vec.12728

Sasaki, K., Mutoh, T., Mutoh, T., Kawashima, R., & Tsubone, H. (2016). Electrical velocimetry for noninvasive cardiac output and stroke volume variation measurements in dogs undergoing cardiovascular surgery. *Vet Anaesth Analg*. doi:10.1111/vaa.12380

Steingart, R. M., Meller, J., Barovick, J., Patterson, R., Herman, M. V., & Teichholz, L. E. (1980). Pulsed doppler echocardiographic measurement of beat-to-beat changes in stroke volume in dogs. *Circulation, 62*(3), 542-548.

Sugo, Y., Sakai, T., Terao, M., Ukawa, T., & Ochiai, R. (2012). The comparison of a novel continuous cardiac output monitor based on pulse wave transit time and echo Doppler during exercise. *Conf Proc IEEE Eng Med Biol Soc, 2012*, 236-239. doi:10.1109/embc.2012.6345913

Sugo, Y., Ukawa, T., Takeda, S., Ishihara, H., Kazama, T., & Takeda, J. (2010). A novel continuous cardiac output monitor based on pulse wave transit time. *Conf Proc IEEE Eng Med Biol Soc, 2010*, 2853-2856. doi:10.1109/iembs.2010.5626343

Taguchi, H., Ichinose, K., Tanimoto, H., Sugita, M., Tashiro, M., & Yamamoto, T. (2011). Stroke volume variation obtained with Vigileo/FloTrac system during bleeding and fluid overload in dogs. *J Anesth, 25*(4), 563-568. doi:10.1007/s00540-011-1175-7

Tavernier, B., Makhotine, O., Lebuffe, G., Dupont, J., & Scherpereel, P. (1998). Systolic pressure variation as a guide to fluid therapy in patients with sepsis-induced hypotension. *Anesthesiology, 89*(6), 1313-1321.

Youden, W. J. (1950). Index for rating diagnostic tests. *Cancer, 3*(1), 32-35. Retrieved from http://onlinelibrary.wiley.com/store/10.1002/1097-0142(1950)3:1<32::AID-CNCR2820030106>3.0.CO;2-3/asset/2820030106_ftp.pdf?v=1&t=ibw1k1ax&s=22721ce6b72e5236ea49fd9f92c5180de57e3ce6

Chapter 5

Investigation of change in pulse wave transit time following mini-fluid challenge predicting fluid responsiveness in spontaneously breathing anaesthetised dogs

Sano H & Chambers P

School of Veterinary Science, Massey University, Palmerston North, New Zealand

This chapter was presented at *13th World Congress of Veterinary Anaesthesiology in Venice, Italy.* as a poster session in September 2018 and accepted by *Veterinary Anaesthesia and Analgesia* in 2020:

Sano H & Chambers JP: Changes in pulse wave transit time following mini-fluid challenge predict fluid responsiveness in dogs. *13th World Congress of Veterinary Anaesthesiology in Venice, Italy.* Poster session, 131, September 2018. The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

PREFACE

As we have seen from **Chapter 4**, mini-fluid challenge using pulse wave transit time has been proved to be a reliable method for fluid responsiveness in mechanically ventilated anaesthetised dogs. However, as mechanical ventilation is not commonly available in veterinary practice, the reproducibility of this technique in spontaneously breathing dogs in a clinical setting required investigation. The purpose of this study was to investigate whether a percentage change in pulse wave transit time following mini-fluid challenge can predict fluid responsiveness in spontaneously breathing anaesthetised dogs undergoing stifle surgery.

This chapter was presented at *13th World Congress of Veterinary Anaesthesiology in Venice, Italy.* as a poster session in September 2018 and accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

ROLES OF EACH OF THE AUTHORS

Sano H: Primarily contributed to the study conception and design, collected and interpreted data with statistical analysis, prepared the manuscript and approved the final version to be published.

Chambers P: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Abstract

Objective: To investigate whether changes in pulse wave transit time (PWTT) following mini-fluid challenge (MFC) could predict fluid responsiveness in spontaneously breathing anaesthetised dogs.

Study design: Prospective clinical trial.

Animals: Forty-five client-owned dogs undergoing stifle surgery.

Methods: Dogs were anaesthetised and breathed spontaneously. Loco-regional blockades were performed prior to the surgery. Haemodynamic variables, velocity time integral (VTI) of the aorta measured by transoesophageal echocardiography, which is a proxy of stroke volume (SV), and PWTT, the time from electrocardiograph R-wave to the pulse oximeter wave, which is inversely proportional to SV, were recorded throughout the study. All dogs received MFC followed by an additional volume expansion when mean arterial pressure was < 70 mmHg. The MFC of 3 mL/kg of colloid was administered over 3 minutes and percentage changes in VTI and PWTT (ΔVTI and ΔPWTT) were calculated. The additional volume expansion of 3 mL/kg of colloid was administered and fluid responsiveness was defined as > 15% change in VTI over the administration of total of 6 mL/kg of colloid. Receiver operator characteristic (ROC) curves were generated for ΔVTI and ΔPWTT to determine cutoff values and grey zones.

Results Nineteen of 45 dogs were responders (42%) and 26 were non-responders (58%). ΔVTI and ΔPWTT predicted fluid responsiveness with areas under ROC curve of 0.91 [95% confidence interval (95%CI) 0.79 to 0.98, $p < 0.001$] and 0.94 [95%CI 0.83 to 0.99, $p < 0.001$], respectively. The cutoff values were 9% and -2.1%, and the grey zones ranging between 6 and 9%, and between -2.7 and - 1.5%, were observed in 9 and 11 out of 45 dogs for ΔVTI and ΔPWTT, respectively.

Conclusion In spontaneously breathing anaesthetised dogs, ΔPWTT following MFC could predict fluid responsiveness non-invasively, although the grey zone should be considered.

Keywords Anaesthesia, cardiac output, fluid therapy, haemodynamic and stroke volume

Abbreviations

ASA	American Society of Anesthesiologists
AUROC	area under the receiver operating characteristic curve
CO	cardiac output (mL/minute)
ΔPWTT	percentage change in PWTT (%)
$\Delta PWTT_{FFC}$	percentage change in PWTT over FFC (%)
$\Delta PWTT_{MFC}$	percentage change in PWTT over MFC (%)
ΔVTI	percentage change in VTI (%)
ΔVTI_{FFC}	percentage change in VTI over FFC (%)
ΔVTI_{MFC}	percentage change in VTI over MFC (%)
$F_E'Iso$	end-tidal isoflurane concentration (%)
FFC	full fluid challenge
f_R	respiratory rate (breaths/minute)
IM	intramuscularly
IV	intravenously
MAP	mean arterial pressure (mmHg)
MFC	mini-fluid challenge
$P_E'CO_2$	end-tidal carbon dioxide tension (mmHg)
PWTT	pulse wave transit time (mm/second)

SV	stroke volume (mL)
TEE	transoesophageal echocardiography
VTI	velocity time integral (cm)
95%CI	95% confidence interval

INTRODUCTION

Fluid therapy is often considered the mainstay of treatment to optimise haemodynamics in people (Cecconi et al., 2015) and dogs (Davis et al., 2013). Although fluid resuscitation can improve outcome in hypovolaemic patients (Nelson, 1976), excess fluid administration can be detrimental in haemodynamically unstable critically ill people (Lobo, Macafee, & Allison, 2006). Volume overload may destroy the endothelial glycocalyx layer and cause tissue oedema, increasing morbidity and mortality (Hippensteel et al., 2019). A meta-analysis of critically ill children who received fluid showed strong and consistent evidence of an association between volume overload and poor outcomes, including worsening respiratory function, development of acute kidney injury, longer paediatric intensive care stay, and death (Alobaidi et al., 2018). However, only half of haemodynamically unstable patients were responsive to a fluid challenge, defined as an increase in stroke volume (SV) or cardiac output (CO) following fluid loading, and other half can be non-responsive, leading to volume overload (Cherpanath, Geerts, Lagrand, Schultz, & Groeneveld, 2013). Therefore, fluid responsiveness should be evaluated in order to prevent either hypo- or hypervolaemia when fluid therapy is considered.

In human medicine, the mini-fluid challenge (MFC) is one of techniques currently available which can be reliably used to determine fluid responsiveness (Guinot et al., 2015; Muller et al., 2011). MFC is an administration of a small amount of fluid, increasing preload. Thus, fluid responsiveness can be assessed based on whether SV increases following MFC according to the Frank-Starling curve of the heart (Figure 5.1). The change in SV of a heart at the steep portion of the Frank-Starling curve will be greater than at the plateau portion after MFC. Therefore, the magnitude of the change in SV after

MFC could predict responsiveness. A recent metanalysis of 21 studies showed that MFC reliably predicted fluid responsiveness in the intensive care unit and operating theatre (Messina et al., 2019). However, in order to determine fluid responsiveness using MFC, accurate measurement or estimation of change in SV is essential.

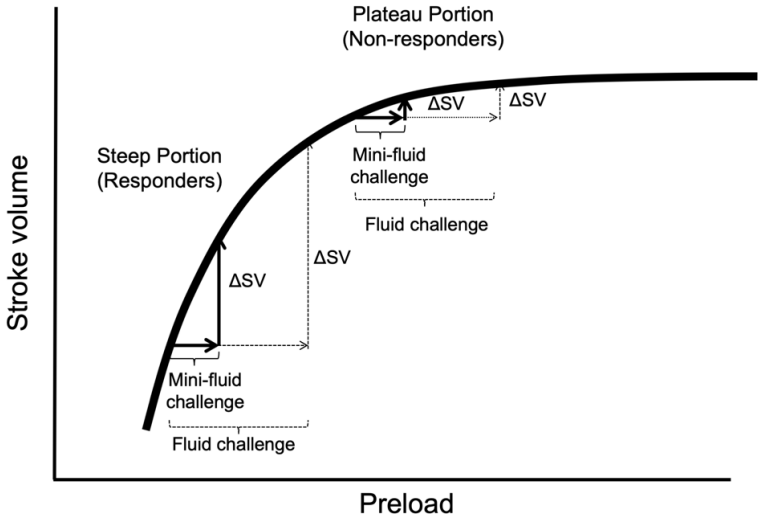

Figure 5.1 The Frank-Starling curve of the heart. Mini-fluid challenge is a strategy to assess fluid responsiveness based on a change in stroke volume (SV) after a small loading dose of fluid. The change in SV at the steep portion of the curve (responders) will be greater than at the plateau portion (non-responders) after both the mini-fluid challenge and fluid challenge. Therefore, the magnitude of the change in SV after the mini-fluid challenge could predict responsiveness to the fluid challenge.

Pulse wave transit time (PWTT), the time from the electrocardiogram R- wave peak to the rise point of the pulse oximeter wave of the same cardiac cycle (Figure 5.2), was reported to be inversely proportional to SV and has a strong negative correlation with SV (Sano & Chambers, 2017; Sugo et al., 2010). A previous study showed that percentage change in PWTT (ΔPWTT) have a good trending ability to detect change in SV measured by thermodilution technique over a wide range of CO in dogs (Sano & Chambers, 2017). A further study revealed ΔPWTT induced by a 3 mL/kg of MFC predicted fluid responsiveness in anaesthetised dogs (Sano et al., 2019). The dogs in this study were mechanically ventilated under a tightly controlled experimental condition. However, in that study, the dogs were mechanically ventilated under tightly controlled experimental conditions. Therefore, reproducibility of this technique in spontaneously breathing dogs in a clinical setting should be investigated.

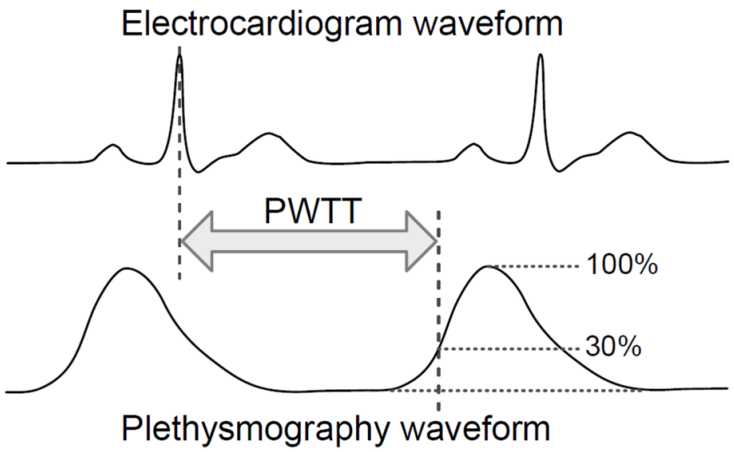

Figure 5.2 Pulse Wave Transit Time (PWTT). PWTT was calculated as the time from the ECG R-wave peak to the rise point of the pulse oximeter wave. The rise point of the pulse wave was defined as the point at which the pulse wave reached 30% of its peak amplitude.

The hypothesis of this study was that a change in PWTT following MFC could predict fluid responsiveness in spontaneously breathing anaesthetised dogs undergoing stifle surgery.

MATERIALS AND METHODS

This study was approved by the animal ethics committee of Massey University (Protocol ID:15/53). Client-owned dogs, weighing between 10 and 40 kg, aged more than 1 year old, and undergoing unilateral stifle surgery (tibial tuberosity advancement or tibial-plateau-leveling osteotomy) were recruited with informed owner consent. Pre-anaesthetic history, physical exam and quick assessment tests (packed cell volume, total solids, blood glucose, and blood urea nitrogen) were evaluated and dogs assessed to be American Society of Anesthesiologists (ASA) physical status I or II and body condition score 3 – 7 (0 - 9) were considered to be eligible for the study. The study was carried out in dogs developing mean arterial pressure (MAP) < 70 mmHg for more than 5 minutes during anaesthesia. Exclusion/withdraw criteria included the requirement of mechanical ventilation [whenever end-tidal carbon dioxide tension ($PE'CO_2$) exceeded 55 mmHg for more than 60 seconds], the failure of loco-regional blocks, and the presence of neurological, respiratory, hepatic, renal, gastrointestinal and cardiac disease including arrhythmias, intractable behaviour, pregnancy, any reasons that would not allow a transoesophageal echocardiography (TEE) probe inserted into the oesophagus, and major perioperative complications such as severe haemorrhage and antibiotic reaction.

Dogs were admitted at least a day before the surgery day. Food was withheld overnight and water was provided *ad libitum*. All dogs were premedicated with acepromazine (0.03 mg/kg; Acezine 2; Ethical agents Ltd, New Zealand) and morphine (0.5 mg/kg; DBL morphine sulfate Injection BP; Hospira NZ Lid, New Zealand) intramuscularly (IM). In thirty minutes, an intravenous catheter (Optiva IV catheter Radiopaque; Smiths Medical International Ltd, UK) was placed into a cephalic vein. Anaesthesia was induced with propofol (Provive MCT-LCT 1%; Claris Lifesciences Australia Pty Ltd, Australia)

intravenously (IV) until orotracheal intubation was achieved and maintained with isoflurane (Isoflurane; Bayer New Zealand Ltd, New Zealand) at an end-tidal isoflurane concentration (F_E´Iso) of 1.0 - 1.4% with 100% oxygen via a rebreathing circuit. All dogs breathed spontaneously and received intravenous fluid [Compound Sodium Lactate (Hartmann's Solution); Baxter Healthcare Ltd, Australia] at a rate of 5 mL/kg/hour for the first hour and then 3 mL/kg/hour for the remainder of anaesthesia. After sterile preparation for loco-regional blockade, ultrasound-guided femoral and sciatic nerve blocks (Toshiba Xario 200 ultrasound unite with a 14 MHz linear transducer, Toshiba, Tokyo, Japan) were carried out on each dog as described elsewhere (Campoy et al., 2010). Each nerve was infiltrated with 0.1 mL /kg of ropivacaine (Ropivacaine Kabi 0.75%; Fresenius Kabi New Zealand Ltd, New Zealand). The dogs were maintained at normal temperature by a warm air blanket (Mistral-Air plus Warming unit; TSCI, Netherlands). After surgery and prior to recovery from anaesthesia, dogs received meloxicam IV (0.2 mg/kg; Metacam; Boehringer Ingelheim Vetmedica, Inc, MO, USA).

During anaesthesia, heart rate, MAP, F_E´Iso, P_E´CO_2, respiratory rate (f_R), haemoglobin oxygen saturation, electrocardiogram (lead II) and rectal temperature were recorded every 5 minutes automatically using a multi-parameter monitor (Life Scope BSM-3763; Nihon Kohden, Tokyo, Japan). MAP was measured via a 20-22 G catheter (Optiva IV catheter Radiopaque; Smiths Medical International Ltd, UK) placed in a dorsal pedal artery after anaesthesia induction using a disposable pressure transducer (BD DTXPlus: DTX Plus TNF-R; Becton Dickinson Critical Care Systems PTE Ltd, Singapore) connected to the multi-parameter monitor. The transducer was positioned at the level of the point of the shoulder and zeroed to atmospheric pressure. All the equipment, including the multi-

parameter monitor, was serviced and maintained at regular intervals by our staff biomechanical engineer.

The PWTT was calculated automatically and averaged over 64 consecutive heart beats by the multi-purpose bedside monitor (Life Scope BSM-3763; Nihon Kohden, Tokyo, Japan). Average PWTT and heart rate were updated every second. Data with a large variability in PWTT (>20 milliseconds) or pulse amplitude deviating from median values (>30%) during calculation were excluded. In addition, the calculation of PWTT was automatically inhibited when >25% of the 64 beats met the following conditions: (a) either electrocardiogram or pulse oximetry pulse wave signal was not obtained; (b) either R wave or the start point of the ascending portion of pulse-oximetry wave was not clearly identified (Sugo, Sakai, Terao, Ukawa, & Ochiai, 2012). ΔPWTT over fluid administration was calculated as ΔPWTT (%) = 100 × (PWTT after fluid administration - PWTT before fluid administration)/(PWTT before fluid administration) (Sano et al., 2019).

The TEE (Toshiba Xario SSA-660A; Toshiba Medical Systems Corporation, Japan) was used to determine velocity time integral (VTI) of aortic blood flow. Percentage change in VTI (ΔVTI) over a fluid challenge was calculated as a proxy for change in SV (Maizel et al., 2007; Muller et al., 2011). The TEE has a clinically acceptable correlation and agreement with the gold standard thermodilution technique in dogs during anaesthesia (Mantovani et al., 2017). It is a minimally invasive CO monitoring tool and has been recommended by the National Institute of Clinical Excellence because of a benefit in guiding fluid therapy in anaesthetized adult and paediatric human patients (Wakeling et al., 2005).

The TEE probe was inserted through a plastic mouth gag into the cranial oesophagus with neutral position and advanced caudally with bent backward position until an image of the heart base was obtained. By setting the array angle to 75-85° and orienting the beam centrally, the longitudinal cranial-oesophageal aorta long-axis-view was obtained. The sample volume of pulsed-wave Doppler was placed in the centre of ascending aorta to measure VTI of the aorta at the end of the expiration in triplicate in three consecutive respiratory cycles. ΔVTI over fluid administration was calculated as ΔVTI (%) = 100 × (VTI after fluid administration - VTI before fluid administration)/(VTI before fluid administration).

MFC, consisting of an intravenous administration of 3 mL/kg of colloid (Voluven 6%, Fresenius Kabi Australia Pty Limited, Pymble NSW, AU) over 3 minutes, was started when MAP < 70 mmHg lasted for more than 5 minutes. All dogs with hypotension received this treatment; dogs which did not develop hypotension were excluded from the study. Measurements were recorded before and one minute after disconnection of MFC. In order to evaluate fluid responsiveness, an additional intravenous administration of 3 mL kg^{-1} of over 3 minutes, was given after the measurement following MFC. The last measurements were recorded one minute after the additional fluid administration. Total fluid administered was 6 mL kg^{-1} of colloid, and defined as full fluid challenge (FFC). During the study, the maintenance fluid was stopped, all clinical settings were kept constant, and dogs were not touched by anybody. The VTI measurement was performed from the stored images in the ultrasound machine by a technician who was unaware of this study after anaesthesia. ΔVTI and ΔPWTT over the MFC or FFC were expressed as ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, and ΔVTI_{FFC} and $\Delta PWTT_{FFC}$, respectively. A responder was defined as having more than a 15% of ΔVTI_{FFC}, and a non-responder as less than 15% of

ΔVTI_{FFC}. After the FFC, the dogs who still had hypotension were given a noradrenaline infusion at a rate of 0.05 μg kg^{-1} $minute^{-1}$.

STATISTICS

Sample size was estimated to detect an area under the receiver operating characteristic curve (AUROC) of 0.75 with null hypothesis (AUROC = 0.5) . At least 42 dogs were required for a study power of 80% and an alpha error of 0.05 and a ratio of sample size in negative/positive groups at 1/2 based on previous similar studies (Goncalves et al., 2020; Sano et al., 2019). Normality was assessed with the Shapiro-Wilk test. Categorical variables were compared using the chi-square or Fisher exact test, as appropriate. Continuous variables were presented as mean (standard deviation) if normally distributed, or as median (interquartile range) if not. Comparisons between responders and non-responders were assessed using Student t test or the Mann–Whitney U test, as appropriate. Comparisons of cardiorespiratory variables during MFC and FFC were assessed using Friedman nonparametric repeated measures comparisons, and post hoc analyses were performed using the Wilcoxon test with Bonferroni adjustment.

Pearson correlations between ΔVTI and ΔPWTT over the fluid administration were also assessed.

Receiver operating characteristic curves were generated for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$ to evaluate the predictive ability of the fluid responsiveness. AUROC with 95% confidence intervals (95%CI) of each variables were created and 95%CI of AUROC > 0.75 were considered a good clinical tool (Coste, Jourdain, & Pouchot, 2006; Ray, Le Manach, Riou, & Houle, 2010). The optimal cutoff value to discriminate responders and non-responders was chosen as the point with the maximal Youden index (sensitivity + specificity - 1). A grey zone was determined to provide a low cutoff value that excludes

positive fluid challenge in 90% of patients, whereas a high cutoff value predicts positive fluid challenge in 90% of cases (Coste et al., 2006; Ray et al., 2010). In order to estimate the grey zone, two steps were required. 1) A bootstrap methodology, which creates 1000 bootstrapped population by randomly drawing instances, with replacement, from the original study population, was used to produce receiver operating characteristic curves in order to determine 95%CI of the optimal thresholds of the 1000 bootstrap population (Carpenter & Bithell, 2000). 2) A threshold related to a sensitivity of 90% and another threshold related to a specificity of 90% using was determined based on receiver operating characteristic curves from the 1,000 bootstrap population. And then the grey zone obtained using these two steps was retained. Statistical analysis was performed using Medcalc (software 11.6; Mariakerke, Belgium) and SPSS (IBM SPSS Statistics Subscription; New York, USA).

RESULTS

A total of 65 dogs were recruited for the study. Among them, 20 were excluded because dogs did not develop MAP < 70 mmHg, the loco-regional blocks seemed not to work well, and adequate TEE images could not be obtained. Characteristics of the remaining 45 dogs are reported in Table 5.1. Dog breeds included 12 cross breed, 10 Huntaway, 8 Labrador Retriever, 6 Golden Retriever, 3 Doberman, 2 Siberian Husky, 1 Rottweiler, 1 German Shepard, 1 Collie, and 1 Weimaraner. Of 45 dogs, 19 were responders and 26 were non-responders. Noradrenaline infusion was given to two dogs in the responders and 19 dogs in non-responders because they still had MAP < 70 mmHg after the FFC.

Table 5.1 Characteristics of 45 dogs [responders (n = 19) and non-responders (n = 26)] before anaesthesia. Data are presented as mean ± standard deviation if the data were normally distributed or median (range) if not. $^{*}p<0.05$ between responders (n = 19) and non-responders (n = 26). f_R, respiratory rate; BUN, blood urea nitrogen; TTA, Tibial tuberosity advancement; TPLO, Tibial plateau leveling osteotomy; NA, non-applicable because BUN was 5-15 mg/dL for all dogs.

	All animals (n = 45)	**Responders** (n = 19)	**Non-responders** (n = 26)	***p*-value**
Age (years)	4.3 ± 1.6	4.4 ± 1.4	4.8 ± 1.5	0.316
Body weight (kg)	26.4 ± 2.9	26.8 ± 2.9	26.0 ± 2.9	0.362
Gender, male/female (n)	20/25	8/11	12/14	0.787
Body condition score (0-9)	7 (6 – 7)	7 (6 – 7)	7 (6 – 7)	0.446
Heart rate (beats/minute)	102 ± 15	104 ± 18	100 ± 13	0.383
f_R (breaths/minute)	26 (22 – 31)	25 (21 – 28)	28 (22 – 31)	0.274
Temperature (°C)	38.5 ± 0.5	38.6 ± 0.6	38.5 ± 0.4	0.618
Packed cell volume (%)	42 ± 4	43 ± 4	42 ± 4	0.299
Total solids (g/dL)	66 ± 7	65 ± 6	66 ± 7	0.437
Blood glucose (mmol/L)	5.4 ± 0.6	5.3 ± 0.6	5.4 ± 0.6	0.730
BUN (mg/L)	5 - 15	5 - 15	5 - 15	NA
TTA/ TPLO	21/24	9/10	12/14	0.936

Cardiorespiratory variables before and after the MFC and the FFC in responders and non-responders are shown in Table 5.2. Heart rate and PWTT decreased, and MAP and VTI increased significantly after the MFC and FFC compared to the baseline, whereas f_R, $P_E'CO_2$ and $F_E'Iso$ were not significantly different over the MFC and FFC. VTI was significantly greater, and PWTT was shorter in responders than those in non-responders. Percentage changes in heart rate, MAP, VTI and PWTT over the fluid administration are presented in Table 5.3. ΔVTI_{FFC}, as a surrogate variable of change in SV to determine fluid responsiveness after the FFC, in responders and non-responders were 22 ± 7% and 7 ± 4, respectively. ΔVTI_{MFC}, $\Delta PWTT_{MFC}$, and $\Delta PWTT_{FFC}$ in responders were more significant than those in non-responders. Significant strong correlations were found between ΔVTI_{MFC} and $\Delta PWTT_{MFC}$ (r = -0.87; 95%CI -0.78 to -0.93, $p < 0.001$), and between ΔVTI_{FFC} and $\Delta PWTT_{FFC}$ (r = -0.94; 95%CI -0.89 to -0.97, $p < 0.001$).

Table 5.2 Cardiorespiratory variables before and after the mini-fluid challenges (MFC: 3 mL/kg) and the full fluid challenges (FFC: 6 mL/kg) in responders and non-responders. Data are presented as median (interquartile range). *$p < 0.017$ compared to baseline using Wilcoxon test with Bonferroni adjustment. †$p < 0.05$ between responders (n = 19) and non-responders (n = 26). HR, heart rate; MAP, mean arterial pressure; VTI, velocity time integral; PWTT, pulse wave transit time; f_R, respiratory rate; $P_E'CO_2$, partial pressure of end-tidal carbon dioxide; F_E'Iso, end-tidal isoflurane concentration.

	Baseline	**After MFC (3 mL/kg)**	**After FFC (6 mL/kg)**	***p*-Value**
HR (beats/minute)				
Responders	73 (67 - 80)	70 (64 - 75) *	70 (64 - 74) *	< 0.00001
Non-responders	76 (70 - 88)	74 (66 - 81) *	74 (69 - 81) *	< 0.00001
MAP (mmHg)				
Responders	66 (65 - 68)	69 (67 - 75) *	71 (70 - 78) *	< 0.00001
Non-responders	66 (65 - 69)	67 (66 - 71) *	68 (67 - 73) *†	< 0.00001
VTI (cm)				
Responders	11.8 (11.1 - 12.0)	13.5 (12.8 - 13.8) *	14.1 (13.6 - 14.5) *	< 0.00001
Non-responders	12.8 (12.4 - 13.2) †	13.6 (13.0 - 13.8) *	13.8 (13.2 - 14.1) *	< 0.00001
PWTT (millisecond)				
Responders	268 (261 - 273)	258 (253 - 262) *	253 (248 - 262) *	< 0.00001
Non-responders	258 (250 - 260) †	254 (248 - 257) *†	254 (247 - 257) *	< 0.00001
f_R (breath/minute)				
Responders	15 (12 - 17)	14 (13 - 16)	14 (12 - 17)	0.721
Non-responders	14 (12 - 16)	15 (12 -16)	14 (12 - 16)	1.000
$P_E'CO_2$ (mmHg)				
Responders	47 (45 - 48)	47 (45 - 48)	47 (45 - 48)	0.607
Non-responders	47 (44 - 48)	46 (44 - 48)	47 (44 - 48)	0.311
F_E'Iso (%)				
Responders	1.4 (1.2 - 1.4)	1.3 (1.3 - 1.4)	1,3 (1.2 - 1.4)	0.472
Non-responders	1.3 (1.3 - 1.4)	1.3 (1.3 - 1.4)	1.3 (1.3 - 1.4)	0.135

Table 5.3 Comparison of percentage changes in velocity time integral (VTI) and pulse wave transit time (PWTT) over the mini-fluid challenges (MFC: 3 mL/kg) and the full fluid challenges (FFC: 6 mL/kg) in responders and non-responders. Data are presented as mean ± standard deviation. $^{*}p < 0.05$ between responders (n = 19) and non-responders (n = 26). ΔHR_{MFC}, ΔMAP_{MFC}, ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage change in heart rate, mean arterial pressure, VTI and PWTT over MFC; ΔHR_{FFC}, ΔMAP_{FFC}, ΔVTI_{FFC} and $\Delta PWTT_{FFC}$, percentage change in heart rate, mean arterial pressure, VTI and PWTT over FFC; NA, non-applicable because the definition of the responders was $15\% > \Delta VTI_{FFC}$.

	Responders (n = 19)	**Non-responders (n = 26)**	***p*-Value**
ΔHR_{MFC} (%)	-3.8 (-5.1 - -3.1)	-3.2 (-6.8 - -2.8)	0.713
ΔMAP_{MFC} (%)	6.0 (1.5 – 9.2)	1.5 (0 – 4.3)	0.029*
ΔVTI_{MFC} (%)	15 ± 7	5 ± 4	< 0.001*
$\Delta PWTT_{MFC}$ (%)	-3.5 ± 1.0	-1.4 ± 0.9	< 0.001*
ΔHR_{FFC} (%)	-5.1 (-7.9 - -4.4)	-4.0 (-7.3 - -2.6)	0.157
ΔMAP_{FFC} (%)	9.2 (6.3 – 13.0)	4.7 (1.5 – 9.2)	0.006*
ΔVTI_{FFC} (%)	22 ± 7	7 ± 4	NA
$\Delta PWTT_{FFC}$ (%)	-5.1 ± 1.4	-1.1 ± 1.0	< 0.001*

The ability of ΔVTI_{MFC} and $\Delta PWTT_{MFC}$ to predict the fluid responsiveness is shown in Table 5.4 and Figure 5.3. ΔVTI_{MFC} and $\Delta PWTT_{MFC}$ predicted fluid responsiveness with AUROC of 0.91 [95% CI0.82 to 0.99, $p < 0.001$] and 0.92 [95% CI0.85 to 0.99, $p < 0.001$], respectively. The cut-off values were 9% and -2.1%, and the grey zones ranging between 5 and 9% and between -1.5 and - 2.7%, were observed in 9 and 11 out of the 45 dogs for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, respectively (Figure 5.4).

No dog showed any complications related to hypotension or fluid administration.

Table 5.4 Ability of percentage change in velocity time integral (ΔVTI_{MFC}) and pulse wave transit time ($\Delta PWTT_{MFC}$) over the mini-fluid challenge to predict increase in stroke volume of 15%. AUROC, area under the receiver operating characteristic curve; 95%CI, 95% confidence interval, Sen; sensitivity, Spe; specificity.

	AUROC (95%CI)	P-value	Cutoff (%)	Sen (%) (95%CI)	Spe (%) (95%CI)	Grey zone (%)
ΔVTI_{MFC}	0.91 (0.79 – 0.98)	< 0.0001	9	89 (67 - 99)	85 (65 - 96)	5 – 9
$\Delta PWTT_{MFC}$	0.94 (0.83 – 0.99)	< 0.0001	-2.1	100 (82 - 100)	73 (52 – 88)	-1.5 – -2.7

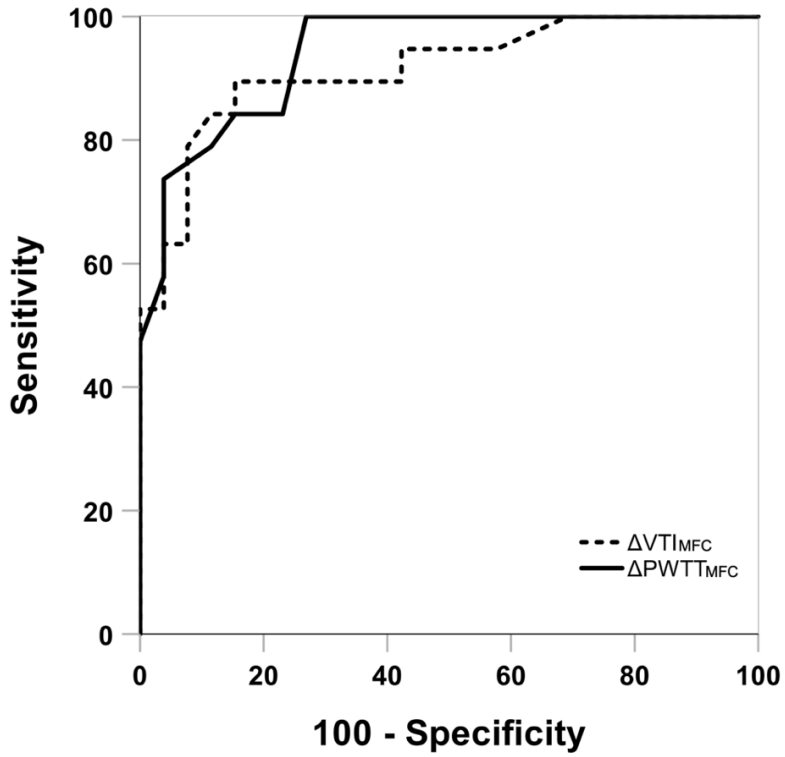

Figure 5.3 Receiver operator characteristic curve for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$. ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage changes in velocity time integral of aortic blood flow and pulse wave transit time after a 3 mL/kg of mini-fluid challenge.

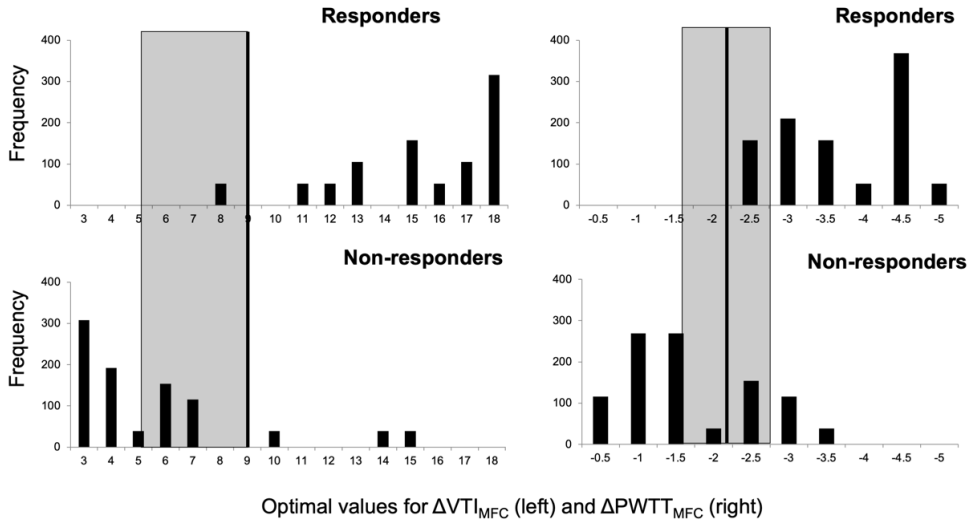

Figure 5.4 The grey zone and the best cutoff value within the optimal values for ΔVTI_{MFC} and $\Delta PWTT_{MFC}$. Distribution of the cutoffs for each bootstrapped population (1000 "optimal" values). Grey rectangle, grey zone; Black line (the best cutoff value); ΔVTI_{MFC} and $\Delta PWTT_{MFC}$, percentage changes in velocity time integral of aortic blood flow and pulse wave transit time after a 3 mL/kg of mini-fluid challenge.

DISCUSSION

This is the first study to demonstrate that fluid responsiveness can be predicted using MFC in spontaneously breathing dogs. The cutoff values of ΔVTI_{MFC} (9%) and $\Delta PWTT_{MFC}$ (-2.1%) can discriminate between responders and non-responders in spontaneously breathing anaesthetized dogs underwent stifle surgery. However, other clinical parameters should be taken into account to evaluate fluid responsiveness when results lie within the grey zones between 5 and 9% (9/45 dogs) for ΔVTI_{MFC} and between -1.5 and -2.7% (11/45 dogs) for $\Delta PWTT_{MFC}$. ΔVTI_{MFC} can be clinically used but requires an expertise technique and expensive equipment. Therefore, $\Delta PWTT_{MFC}$ is clinically more prsctical to assess the fluid responsiveness because this technique can be easily and non-invasively performed with a routine anaesthetic monitor in spontaneously breathing dogs. However, comprehensive assessment should be considered in clinical practice.

The MFC is a technique to assess fluid responsiveness based on a change in SV after a small loading dose of fluid (Marik & Lemson, 2014). Theoretically, the change in SV of a heart at the steep portion of the Frank-Starling curve will be greater than at the plateau portion after both MFC and FFC, and the greater the increase in SV after MFC, the more we can expect a similar increase in SV after FFC. Therefore, the magnitude of the change in SV after MFC could predict responsiveness to FFC. The first study regarding MFC was conducted by Muller et al (Muller et al., 2011). ΔVTI at the aortic outflow tract over colloid infusion of 100 mL was measured in deeply sedated patients on mechanical ventilation without arrhythmias, resulting in excellent accuracy (AUROC = 0.92, 95%CI: 0.78 – 0.98). A study described by Guinot et al. indicated potential use of MFC in spontaneously breathing patients (Guinot et al., 2015), contrary to dynamic indices of fluid responsiveness such as pulse pressure variation. A more than 7% increase in SV

measured by thoracic impedance cardiography over crystalloid administration of 100 mL accurately predicted >15% increase in SV after an administration of 500 mL crystalloid (AUROC = 0.93, 95%CIs: 0.80 – 0.97) in spontaneously breathing patients under spinal anaesthesia. A metanalysis on the reliability of MFC in predicting fluid responsiveness reported that the pooled AUROC for the MFC was 0.91 (95%CIs 0.85 – 0.97) and the pooled sensitivity and specificity were 0.82 (95%CIs: 0.76 – 0.88) and 0.83 (95%CIs: 0.77 – 0.89), respectively, with a best median threshold of 5% (range: 3.0 – 7.0%)(Messina et al., 2019). Therefore, the MFC is a well-tested and effective tool to predict fluid responsiveness under various clinical conditions in people. The current MFC study revealed that ΔVTI_{MFC} accurately predicted the fluid responsiveness with similar result in spontaneously breathing dogs.

The study of MFC in veterinary medicine is relatively new and rare. In order to evaluate the fluid responsiveness using MFC in dogs as well as people, there are several important definitions that need to be considered:

1) The volume of fluid for MFC to predict the increase in SV after FFC: the volume of MFC should be minimal to cause fewer adverse effects in non-responders. However, it must be sufficient to give a greater increase in SV in responders than non-responder. In people, 100 – 150 mL or 1.5 – 3.0 mL/kg of MFC are commonly used. In a previous study on MFC in mechanically ventilated anaesthetised dogs, 3 mL/kg of colloid could predict fluid responsiveness reliably but 1 and 2 mL/kg did not. Therefore, 3 mL/kg of colloid was employed in the current study.

2) The volume of fluid for FFC to distinguish between responders and non-responders: the volume of FFC are around 500 mL or 7 – 15 mL/kg in people, and 3 – 20 mL/kg in dogs. The study described by Rabozzi and Franci showed that 3 mL/kg of crystalloid was

able to discriminate fluid responders (10% increase in MAP and/or >10% decrease in heart rate) (Rabozzi & Franci, 2014). The volume of FFC in the current study used 6 mL/kg of colloid (2 × 3 mL/kg) because 3 mL/kg of MFC could be used repeatedly until dogs become non-responsive in a practice. Therefore, the FFC of 6 mL/kg was thought be more than enough and practical.

3) The type of fluid: Several types of fluid have been used such as crystalloid, colloid, plasma, and albumin. However, one study reported that immediate haemodynamic response to MFC is independent of fluid type (Joosten, Delaporte, Van der Linden, Rinehart, & Hipszer, 2019). The current study used colloid solution because longer intravascular persistence of colloid solution was expected.

4) The definition of the fluid responsiveness and accurate measurement of change in SV after MFC and FFC: Most studied defined the fluid responsiveness as CO > 10-15% or SV/a surrogate > 10-15%. These numbers need to be greater than variability of CO or SV measurement. The current study used PWTT and VTI as surrogate SV measurement. PWTT was calculated by the algorithm of the monitoring machine automatically. Interobserver variability for the measurement of VTI is usually reported at approximately 3–8% (Lewis, Kuo, Nelson, Limacher, & Quinones, 1984; Oren-Grinberg & Park, 2008). Considered to all these factors, 15% was used in the current study.

Assessment of change in CO or SV is challenging in a veterinary practice. Several methods for CO measurement have also been tested in dogs (Fegler, 1954; Kutter, Bettschart-Wolfensberger, Romagnoli, & Bektas, 2016; Mantovani et al., 2017; Morgaz et al., 2014), but are impractical in a routine veterinary practice. In the current study, TEE was used to measure VTI and to estimate SV because it is minimally invasive and provides good agreement with the thermodilution technique in dogs (Mantovani et al.,

2017; Yamashita et al., 2007) and transoesophageal doppler device accurately reflected the direction and magnitude of the changes of CO over time during abrupt haemodynamic changes in dogs (de Figueiredo, Cruz, Silva, & Rocha, 2004). In people, TEE has been used to estimate an increase in SV following MFC for fluid responsiveness (Mukhtar et al., 2019; Muller et al., 2011; Wu et al., 2014). However, the use of TEE is still uncommon in a veterinary practice although TEE may be a good minimally invasive tool to estimate SV. Thus, the PWTT was also used to estimate SV in the current study because the PWTT can be easily measured using inexpensive, non-invasive and routine anaesthetic monitoring equipment (ECG and pulse oximeter). The PWTT has a strong negative correlation with SV in dogs and humans (Ishihara et al., 2004; Sugo et al., 2012; Sugo et al., 2010). In the previous study, ΔPWTT showed an acceptable trending ability to detect changes in SV derived from CO measurement by thermodilution technique with a concordance rate of 95% and a correlation coefficient of -0.86 over a wide range of CO in isoflurane-anaesthetised dogs (Sano & Chambers, 2017). In the current study, the strong correlation between ΔVTI and ΔPWTT was observed. Therefore, ΔPWTT could be used as a surrogate of ΔVTI, thus a change in SV.

Over MFC and FFC in the current study, HR decreased, and MAP increased significantly. However, those changes were subtle and probably clinically insignificant in both responders and non-responders. The only variables that changed statistically and clinically seemed to be VTI and PWTT. Since VTI is challenging to measure due to requirement for an expensive machine and a skilled expert, PWTT may become a useful parameter to assess blood flow in clinical veterinary medicine.

The result of the current study is similar to the previous study showing that ΔPWTT over

MFC of 3 mL/kg predict ΔVTI > 15% after FFC of 10 mL/kg with a cutoff value of -2.4% (grey zone ranging between -1.9 and -2.9%) in mechanically ventilated anaesthetised dogs (Sano et al., 2019). The previous study tested the fluid responsiveness under tightly controlled experimental condition unlike the current clinical study. All dogs were anatomically similar in conformation, and mechanically ventilated with a consistent respiratory setting. In this study, f_R and $P_E'CO_2$ were varied individually although they are within the normal range, thus, effects of individual and respiratory variation may not affect this technique. The most remarkable finding in the current study was that the fluid responsiveness was able to be determined by MFC in spontaneously breathing anaesthetised dogs in a normal clinical setting. Most studies investigating fluid responsiveness used mechanically ventilated dogs (Drozdzynska, Chang, Stanzani, & Pelligand, 2018; Fantoni et al., 2017; Sano et al., 2019). The next stage is to study MFC using PWTT to assess the fluid responsiveness in awake dogs in intensive care.

The current study has limitations. First, dogs recruited in the study were relatively healthy and assessed ASA physical status I or II. Most dogs were assumed to be not hypovolaemic although some may be minimally dehydrated. Extremely high or low physiological parameters in terms of heart rate, cardiac contractility, arterial stiffness (elastance), blood viscosity and body temperature may alter the cutoff number of ΔPWTT. The further study is required to evaluate the feasibility of $\Delta PWTT_{MFC}$ to predict the fluid responsiveness in critically ill dogs or dogs with congestive heart failure. Second, all dogs received the same anaesthetic protocol and similar type of surgery. Vasoactive agent such as alpha 2 agonists may influence the measurement of PWTT (Sano & Chambers, 2017). Therefore, this result should not be extrapolated to dogs with different anaesthetic plans and use of vasoactive agents. Third, Third, the use of VTI to evaluate fluid responsiveness in

conscious spontaneously breathing dogs was reported previously (Oricco, Rabozzi, Meneghini, & Franci, 2019). However, VTI obtained by TEE was less reliable during hypotension although it is still clinically acceptable (Mantovani et al., 2017). Moreover, the trending ability of TEE to measure TVI is unknown in dogs. In people, interobserver variability for the measurement of VTI is usually reported at approximately 3–8% (Lewis et al., 1984; Oren-Grinberg & Park, 2008). In this study, the definition of fluid responsiveness was having more than a 15% of ΔVTI_{FFC}, and the cutoff values of ΔVTI_{MFC} to discriminate between responders and non-responders was 9%. ΔVTI was within the interobserver variability for the measurement of VTI. Fourth, the PWTT is derived from both electrocardiogram and pulse oximetry. Thus, the accuracy of PWTT measurement might be influenced by arrhythmias such as sinus arrhythmia commonly seen in dogs, and the quality of pulse oximetry signal interfered by many factors including hypovolaemia, vascular tone caused by other anaesthetic agents or cold, and measurement location.

In conclusion, a 3 mL/kg of MFC could predict fluid responsiveness based on ΔPWTT in spontaneously breathing anaesthetised dogs. $\Delta PWTT_{MFC}$ may be inconclusive (between -2.7% and -1.5%) in approximately 24% of aneasthetised dogs. Since the measurement of PWTT requires only ECG and pulse oximeter data, this technique could be utilized in routine veterinary practice.

REFERENCES

Alobaidi, R., Morgan, C., Basu, R. K., Stenson, E., Featherstone, R., Majumdar, S. R., & Bagshaw, S. M. (2018). Association Between Fluid Balance and Outcomes in Critically Ill Children: A Systematic Review and Meta-analysis. *JAMA Pediatr, 172*(3), 257-268. doi:10.1001/jamapediatrics.2017.4540

Campoy, L., Bezuidenhout, A. J., Gleed, R. D., Martin-Flores, M., Raw, R. M., Santare, C. L., . . . Wang, A. L. (2010). Ultrasound-guided approach for axillary brachial plexus, femoral nerve, and sciatic nerve blocks in dogs. *Vet Anaesth Analg, 37*(2), 144-153. doi:10.1111/j.1467-2995.2009.00518.x

Carpenter, J., & Bithell, J. (2000). Bootstrap confidence intervals: when, which, what? A practical guide for medical statisticians. *Stat Med, 19*(9), 1141-1164. doi:10.1002/(sici)1097-0258(20000515)19:9<1141::aid-sim479>3.0.co;2-f

Cecconi, M., Hofer, C., Teboul, J. L., Pettila, V., Wilkman, E., Molnar, Z., . . . De Backer, D. (2015). Fluid challenges in intensive care: the FENICE study: A global inception cohort study. *Intensive Care Med, 41*(9), 1529-1537. doi:10.1007/s00134-015-3850-x

Cherpanath, T. G., Geerts, B. F., Lagrand, W. K., Schultz, M. J., & Groeneveld, A. B. (2013). Basic concepts of fluid responsiveness. *Neth Heart J, 21*(12), 530-536. doi:10.1007/s12471-013-0487-7

Coste, J., Jourdain, P., & Pouchot, J. (2006). A gray zone assigned to inconclusive results of quantitative diagnostic tests: Application to the use of brain natriuretic peptide for diagnosis of heart failure in acute dyspneic patients. *Clin Chem, 52*(12), 2229-2235. doi:10.1373/clinchem.2006.072280

Davis, H., Jensen, T., Johnson, A., Knowles, P., Meyer, R., Rucinsky, R., & Shafford, H. (2013). 2013 AAHA/AAFP fluid therapy guidelines for dogs and cats. *J Am Anim Hosp Assoc, 49*(3), 149-159. doi:10.5326/jaaha-ms-5868

de Figueiredo, L. F., Cruz, R. J., Jr., Silva, E., & Rocha, E. S. M. (2004). Cardiac output determination during experimental hemorrhage and resuscitation using a transesophageal Doppler monitor. *Artif Organs, 28*(4), 338-342. doi:10.1111/j.1525-1594.2004.47351.x

Drozdzynska, M. J., Chang, Y. M., Stanzani, G., & Pelligand, L. (2018). Evaluation of the dynamic predictors of fluid responsiveness in dogs receiving goal-directed fluid therapy. *Vet Anaesth Analg, 45*(1), 22-30. doi:10.1016/j.vaa.2017.06.001

Fantoni, D. T., Ida, K. K., Gimenes, A. M., Mantovani, M. M., Castro, J. R., Patrício, G. C. F., . . . Otsuki, D. A. (2017). Pulse pressure variation as a guide for volume expansion in dogs undergoing orthopedic surgery. *Vet Anaesth Analg, 44*(4), 710-718. doi:10.1016/j.vaa.2016.11.011

Fegler, G. (1954). Measurement of cardiac output in anaesthetized animals by a thermodilution method. *Q J Exp Physiol Cogn Med Sci, 39*(3), 153-164. doi:10.1113/expphysiol.1954.sp001067

Goncalves, L. A., Otsuki, D. A., Pereira, M. A., Nagashima, J. K., Ambrosio, A. M., & Fantoni, D. T. (2020). Comparison of pulse pressure variation versus echocardiography-derived stroke volume variation for prediction of fluid responsiveness in mechanically ventilated anesthetized dogs. *Vet Anaesth Analg, 47*(1), 28-37. doi:10.1016/j.vaa.2019.08.047

Guinot, P. G., Bernard, E., Defrancq, F., Petiot, S., Majoub, Y., Dupont, H., & Lorne, E. (2015). Mini-fluid challenge predicts fluid responsiveness during spontaneous breathing under spinal anaesthesia: An observational study. *Eur J Anaesthesiol, 32*(9), 645-649. doi:10.1097/eja.0000000000000175

Hippensteel, J. A., Uchimido, R., Tyler, P. D., Burke, R. C., Han, X., Zhang, F., . . . Schmidt, E. P. (2019). Intravenous fluid resuscitation is associated with septic endothelial glycocalyx degradation. *Crit Care, 23*(1), 259. doi:10.1186/s13054-019-2534-2

Ishihara, H., Okawa, H., Tanabe, K., Tsubo, T., Sugo, Y., Akiyama, T., & Takeda, S. (2004). A new non-invasive continuous cardiac output trend solely utilizing routine cardiovascular monitors. *J Clin Monit Comput, 18*(5-6), 313-320. doi:10.1007/s10877-005-2452-5

Joosten, A., Delaporte, A., Van der Linden, P., Rinehart, J., & Hipszer, B. (2019). Immediate haemodynamic impact response to a mini-fluid challenge is independent of fluid type: A post-hoc analysis of a randomised double blinded controlled trial. *Anaesth Crit Care Pain Med, 38*(6), 669-670. doi:10.1016/j.accpm.2018.10.017

Kutter, A. P., Bettschart-Wolfensberger, R., Romagnoli, N., & Bektas, R. N. (2016). Evaluation of agreement and trending ability between transpulmonary thermodilution and calibrated pulse contour and pulse power cardiac output monitoring methods against pulmonary artery thermodilution in anesthetized dogs. *J Vet Emerg Crit Care (San Antonio), 26*(4), 531-540. doi:10.1111/vec.12439

Lewis, J. F., Kuo, L. C., Nelson, J. G., Limacher, M. C., & Quinones, M. A. (1984). Pulsed Doppler echocardiographic determination of stroke volume and cardiac output: clinical validation of two new methods using the apical window. *Circulation, 70*(3), 425-431. doi:10.1161/01.cir.70.3.425

Lobo, D. N., Macafee, D. A., & Allison, S. P. (2006). How perioperative fluid balance influences postoperative outcomes. *Best Pract Res Clin Anaesthesiol, 20*(3), 439-455. doi:10.1016/j.bpa.2006.03.004

Maizel, J., Airapetian, N., Lorne, E., Tribouilloy, C., Massy, Z., & Slama, M. (2007). Diagnosis of central hypovolemia by using passive leg raising. *Intensive Care Med, 33*(7), 1133-1138. doi:10.1007/s00134-007-0642-y

Mantovani, M. M., Fantoni, D. T., Gimenes, A. M., de Castro, J. R., Flor, P. B., Ida, K. K., & Schwartz, D. S. (2017). Clinical monitoring of cardiac output assessed by transoesophageal echocardiography in anaesthetised dogs: a comparison with the thermodilution technique. *BMC Vet Res, 13*(1), 325. doi:10.1186/s12917-017-1227-9

Marik, P. E., & Lemson, J. (2014). Fluid responsiveness: an evolution of our understanding. *Br J Anaesth, 112*(4), 617-620. doi:10.1093/bja/aet590

Messina, A., Dell'Anna, A., Baggiani, M., Torrini, F., Maresca, G. M., Bennett, V., . . . Cecconi, M. (2019). Functional hemodynamic tests: a systematic review and a metanalysis on the reliability of the end-expiratory occlusion test and of the mini-fluid challenge in predicting fluid responsiveness. *Crit Care, 23*(1), 264. doi:10.1186/s13054-019-2545-z

Morgaz, J., Granados Mdel, M., Munoz-Rascon, P., Dominguez, J. M., Fernandez-Sarmiento, J. A., Gomez-Villamandos, R. J., & Navarrete, R. (2014). Comparison of thermodilution, lithium dilution, and pulse contour analysis for the measurement of cardiac output in 3 different hemodynamic states in dogs. *J Vet Emerg Crit Care (San Antonio), 24*(5), 562-570. doi:10.1111/vec.12219

Mukhtar, A., Awad, M., Elayashy, M., Hussein, A., Obayah, G., El Adawy, A., . . . Teboul, J. L. (2019). Validity of mini-fluid challenge for predicting fluid responsiveness following liver transplantation. *BMC Anesthesiol, 19*(1), 56. doi:10.1186/s12871-019-0728-4

Muller, L., Toumi, M., Bousquet, P. J., Riu-Poulenc, B., Louart, G., Candela, D., . . . Lefrant, J. Y. (2011). An increase in aortic blood flow after an infusion of 100 ml colloid over 1 minute can predict fluid responsiveness: the mini-fluid challenge study. *Anesthesiology, 115*(3), 541-547. doi:10.1097/ALN.0b013e318229a500

Nelson, A. W. (1976). Hypovolemic shock. *Vet Clin North Am, 6*(2), 187-192. doi:10.1016/s0091-0279(76)50028-1

Oren-Grinberg, A., & Park, K. W. (2008). Assessment of myocardial systolic function by TEE. *Int Anesthesiol Clin, 46*(2), 31-49. doi:10.1097/AIA.0b013e31811f45e7

Oricco, S., Rabozzi, R., Meneghini, C., & Franci, P. (2019). Usefulness of focused cardiac ultrasonography for predicting fluid responsiveness in conscious, spontaneously breathing dogs. *Am J Vet Res, 80*(4), 369-377. doi:10.2460/ajvr.80.4.369

Rabozzi, R., & Franci, P. (2014). Use of systolic pressure variation to predict the cardiovascular response to mini-fluid challenge in anaesthetised dogs. *Vet J, 202*(2), 367-371. doi:10.1016/j.tvjl.2014.08.022

Ray, P., Le Manach, Y., Riou, B., & Houle, T. T. (2010). Statistical evaluation of a biomarker. *Anesthesiology, 112*(4), 1023-1040. doi:10.1097/ALN.0b013e3181d47604

Sano, H., & Chambers, J. P. (2017). Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Vet Anaesth Analg, 44*(5), 1057-1067. doi:10.1016/j.vaa.2016.11.014

Sano, H., Fujiyama, M., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2019). Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. *J Vet Emerg Crit Care (San Antonio), 29*(4), 391-398. doi:10.1111/vec.12860

Sugo, Y., Sakai, T., Terao, M., Ukawa, T., & Ochiai, R. (2012). The comparison of a novel continuous cardiac output monitor based on pulse wave transit time and echo Doppler during exercise. *Conf Proc IEEE Eng Med Biol Soc, 2012*, 236-239. doi:10.1109/embc.2012.6345913

Sugo, Y., Ukawa, T., Takeda, S., Ishihara, H., Kazama, T., & Takeda, J. (2010). A novel continuous cardiac output monitor based on pulse wave transit time. *Conf Proc IEEE Eng Med Biol Soc, 2010*, 2853-2856. doi:10.1109/iembs.2010.5626343

Wakeling, H. G., McFall, M. R., Jenkins, C. S., Woods, W. G., Miles, W. F., Barclay, G. R., & Fleming, S. C. (2005). Intraoperative oesophageal Doppler guided fluid management shortens postoperative hospital stay after major bowel surgery. *Br J Anaesth, 95*(5), 634-642. doi:10.1093/bja/aei223

Wu, Y., Zhou, S., Zhou, Z., & Liu, B. (2014). A 10-second fluid challenge guided by transthoracic echocardiography can predict fluid responsiveness. *Crit Care, 18*(3), R108. doi:10.1186/cc13891

Yamashita, K., Ueyama, Y., Miyoshi, K., Igarashi, R., Kushiro, T., Umar, M. A., & Muir, W. W. (2007). Minimally invasive determination of cardiac output by transthoracic bioimpedance, partial carbon dioxide rebreathing, and transesophageal Doppler echocardiography in beagle dogs. *J Vet Med Sci, 69*(1), 43-47. doi:10.1292/jvms.69.43

Chapter 6

Effects of noradrenaline infusion prior to hypotension on anaesthetic management in dogs undergoing ovariohysterectomy

Sano H, Chambers P, Bridges J, Johnson C & McGlade K

School of Veterinary Science, Massey University, Palmerston North, New Zealand

This chapter was presented at *American College of Veterinary Anesthesia and Analgesia Annual Conference in Washington D.C.* as an oral presentation in September 2019 and accepted by *Veterinary Anaesthesia and Analgesia* in 2020:

Sano H, Chambers P, Bridges J: Effects of preemptive norepinephrine infusion prior to hypotension on anesthetic management in dogs undergoing ovariohysterectomy. *American College of Veterinary Anesthesia and Analgesia Annual Conference in Washington D.C., USA*.Oral presentation, 111, September 2019. doi: 10.1016/j.vaa.2019.08.039 The manuscript was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

This study was Supported by Massey University Research Funds.

Chapter 6

PREFACE

As we have seen from **all previous chapters**, PPV, PVI, and MFC using PWTT have been proved to be reliable predicters for fluid responsiveness in anaesthetised dogs. However, those measurements are still limited to be used in veterinary clinical practice because of availability of equipment, difficulty of their interpretation, and a cumbersome process. The main purpose of our investigated parameters is to prevent excessive fluid administration. The time when most practitioners administer a bolus of fluid during anaesthesia is when hypotension is encountered even though the cause of hypotension is not hypovolaemia. Thus, if hypotension can be prevented, excessive fluid administration can be also avoided in normal health dogs. The main cause of perioperative hypotension is anaesthetic agents because they decrease cardiac contractility and cause vasodilation unless obvious hypovolaemia or haemorrhage occur. Therefore, prophylactic noradrenaline, which counteracts cardiovascular adverse effects, would be able to prevent hypotension, and thus excessive fluid loading in anaesthetised dogs.

This chapter was presented in *American College of Veterinary Anesthesia and Analgesia Annual Conference in Washington D.C.* as an oral presentation in September 2019 and was accepted by *Veterinary Anaesthesia and Analgesia* in 2020.

ROLES OF EACH OF THE AUTHORS

Sano H: Primarily contributed to the study conception and design, collected and interpreted data with statistical analysis, prepared the manuscript and approved the final version to be published.

Chambers P: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

Bridges J: Contributed to interpreted data with statistical analysis and prepared the manuscript

Johnson C: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

McGlade K: Contributed to the study conception and design and revised the manuscript critically for important intellectual content.

ABSTRACT

Objective: To investigate whether noradrenaline infusion prior to hypotension improves anaesthetic management in dogs undergoing ovariohysterectomy.

Study design: Prospective, blinded, clinical comparison.

Animals: Fifty client-owned dogs.

Methods: Dogs received either noradrenaline infusion (0.05 µg/kg/minute, $n = 25$) or saline (equal volume, $n = 25$) intravenously during anaesthesia. After intramuscular administration of acepromazine (0.03 mg/kg) and morphine (0.5 mg/kg), anaesthesia was induced with propofol and maintained with isoflurane. The dogs breathed spontaneously and received Hartmann's solution. Hypotension was treated using AAHA guidelines (Bednarski et al., 2011; Davis et al., 2013). Heart rate (HR), mean arterial pressure (MAP), cardiac index (CI), and end-tidal isoflurane ($F_E'Iso$) were recorded every 5 minutes. Lactate was measured before infusion and after anaesthesia. Numbers requiring rescue propofol, fluid bolus, additional noradrenaline and total hypotensive time were recorded. Significance was tested with Mann–Whitney U test for continuous variables and with chi-square test for categorical data. A linear mixed-effects model was fitted to the data.

Results: Data was expressed as median (range). There were no differences in preoperative measurements. Overall, median MAP [86 (range 72-109) *versus* 72 (67-88) mmHg, $p = 0.001$] and $F_E'Iso$ [1.3 (1.2-1.4) *versus* (1.1-1.4) %, $p = 0.001$] were significantly greater and HR [71 (62-114) *versus* 100 (73-118) beats minute^{-1}, $p = 0.001$] was lower in the noradrenaline group, while no differences were detected for CI. Hypotensive time [0 (0-15) *versus* 15 (0-45) minutes, $p = 0.001$], dogs requiring rescue propofol (11/25 *versus* 20/25, $p = 0.019$), fluid boluses (1/25 *versus* 12/25, $p = 0.001$), and additional noradrenaline infusion (0/25 *versus* 10/25, $p = 0.001$), were lower in the noradrenaline group. Lactate did not change.

Conclusions and clinical relevance: Prophylactic noradrenaline infusion provided stable anaesthesia with less fluid administered in dogs undergoing ovariohysterectomy.

Keywords anaesthesia, arterial blood pressure, cardiac output, fluid therapy, haemodynamic

Abbreviations

ASA	American Society of Anesthesiologists
BG	blood glucose (mmol/L)
CDA	clinical depth of anaesthesia
CI	cardiac index (mL/kg/minute)
CO	cardiac output (mL/minute)
$F_E'Iso$	end-tidal isoflurane concentration (%)
f_R	respiratory rate (breaths/minute)
HR	heart rate (beats/minute)
IM	intramuscularly
IV	intravenously
MAP	mean arterial pressure (mmHg)
PCV	packed cell volume (%)
SE	standard error
$P_E'CO_2$	end-tidal carbon dioxide tension (mmHg)
T	temperature (°C)
TEE	transoesophageal echocardiography
TS	total solids (g/L)

Chapter 6

INTRODUCTION

Perioperative blood pressure management is important because most anaesthetic agents have vasodilatory and negative inotropic effects which can increase morbidity and mortality (Brodbelt, 2009; Gaynor et al., 1999). Severe hypotension [mean arterial pressure (MAP) < 60 mmHg or systolic arterial pressure < 80 mmHg] occurs in up to 65% of anaesthetics (Iizuka, Kamata, Yanagawa, & Nishimura, 2013; Redondo et al., 2007) . Hypotension is used as a surrogate measure of inadequate perfusion, and thus probable tissue hypoxia, increasing the incidence of postoperative complications in humans (Monk, Saini, Weldon, & Sigl, 2005; Reich et al., 2005). Hypotension-related postoperative wound infection (Turk, Singh, & Weese, 2015) and anastomotic leakage in intestinal surgery (Snowdon, Smeak, & Chiang, 2016) were reported in dogs. Therefore, treatment for hypotension is essential.

The 2011 American Animal Hospital Association anaesthesia (AAHA) and 2013 AAHA/American Association of Feline Practitioners fluid therapy guidelines suggest treating hypotension by decreasing depth of anaesthesia, giving a bolus of isotonic crystalloid intravenously, considering administration of colloids and inopressors (Bednarski et al., 2011; Davis et al., 2013). In veterinary practice, reducing anaesthetic depth and administering fluid boluses are often considered the first-line treatments as both are simple and cheap.

Decreasing anaesthetic depth is rational because anaesthetics reduce vessel tone and cardiac contractility in a dose-dependent manner, to the point that blood pressure is used as an indication of anaesthetic depth in clinical practice. However, decreasing depth increases the risk of awareness or movement. Administration of fluid to hypovolaemic patients is an appropriate treatment, which can increase cardiac output (CO) by increasing stroke volume

(if the patient is fluid responsive) and thus blood pressure. However, isotonic crystalloid boluses did not improve blood pressure but caused haemodilution in deeply anaesthetised euvolemic hypotensive dogs (Aarnes, Bednarski, Lerche, Hubbell, & Muir, 2009; Valverde, Gianotti, Rioja-Garcia, & Hathway, 2012). In Valverde's study, only reduction of anaesthetic depth improved cardiovascular function. Excessive fluid administration can contribute to postoperative complications, prolonged length of hospital stay, organ failure and mortality (Brodbelt, 2009; Lobo, Macafee, & Allison, 2006). Administration of excess fluid may cause tissue oedema (Holte, Sharrock, & Kehlet, 2002), which impairs oxygen delivery to tissues. Therefore, decreasing anaesthetic depth and fluid administration requires judgement unless anaesthesia is too deep and obvious hypovolaemia or haemorrhage occur.

Since an excessively light plane of anaesthesia and fluid overload can be caused by treatment for hypotension, these can be prevented if hypotension does not occur. Assuming that healthy animals fasted overnight without signs or history of dehydration usually have minimal hypovolaemia (Osugi, Tatara, Yada, & Tashiro, 2011), most hypotension is likely to be due to vasodilatation and/or decreased cardiac contractility produced by anaesthetics. Administration of an inopressor could potentially prevent these effects.

Noradrenaline is a potent α1- and mild β1-adrenergic agonist, and may counteract anaesthesia-induced vasodilatation and thus hypotension (Hiltebrand, Koepfli, Kimberger, Sigurdsson, & Brandt, 2011). It is one of a variety of vasopressors used clinically to treat hypotensive states (Ferguson-Myrthil, 2012). It is associated with a lower mortality, a lower risk of arrhythmias and gastrointestinal reaction in patients with cardiogenic and septic shock compared to dopamine (Rui et al., 2017; Sandifer & Jones, 2012). Treatment of perioperative

hypotension with noradrenaline in abdominal surgery was reported to have no adverse effects on microcirculatory blood flow or tissue oxygen tension in the intestinal tract (Hiltebrand et al., 2011). In experimentally anaesthetised dogs, noradrenaline increased blood pressure but did not change CO and renal blood flow, while in anaesthetised septic dogs, it improved all haemodynamics (Bellomo, Kellum, Wisniewski, & Pinsky, 1999). It was also reported that prophylactic noradrenaline infusion with restrictive fluid therapy reduced complications and hospitalisation time in people (Wuethrich, Burkhard, Thalmann, Stueber, & Studer, 2014).

The aim of this study was to assess the clinical use of routine noradrenaline intravenous infusion prior to any hypotension developing in dogs undergoing ovariohysterectomy. We hypothesised that noradrenaline would reduce the incidence and extent of hypotension, light plane of anaesthesia, and reduce fluid volume administered compared to the controls.

MATERIALS AND METHODS

The study was a prospective, blinded, randomised clinical comparison and approved by the animal ethics committee of Massey University (Protocol ID:15/52). Female dogs with American Society of Anesthesiologists (ASA) physical status I undergoing ovariohysterectomy were included. ASA physical status was determined by an attending anaesthesiologist. Small dogs in which transoesophageal echocardiography (TEE) was not feasible, and obviously pregnant dogs, determined by an attending clinician and their history, were excluded. Withdrawal criteria included intractable behaviour, additional sedatives required, unexpected pregnancy, major perioperative complications such as severe haemorrhage, antibiotic reaction, and additional surgeries required. Based on the assumption that noradrenaline infusion would reduce the incidence of hypotension (percentage of patients in which hypotension was present at some of the recorded time points) from 45% (incidence of hypotension at our facility) to 10%, a sample size of 25 dogs per group was calculated with a type I error of 0.05 and a power of 80%. After informed owner consent was obtained, 56 dogs were recruited from July 2015 to May 2016. Each dog was assigned randomly to one of two groups (noradrenaline or control group) drawing a ballot from a closed envelope after random allocation of ballots for a 1:1 ratio. One ballot for each dog was placed into a closed and opaque envelope by a nurse who was not involved in patient management or data collection. The nurse was responsible for opening the envelope for each dog recruited in the study and for preparing the study drug according to the ballot contained in the envelope. Dog breed, age, body weight, body condition score, preanaesthetic heart rate (HR), respiratory rate (f_R), temperature (T), packed cell volume (PCV), total solids (TS), blood glucose (BG), and blood urea nitrogen were recorded.

Food was withheld overnight and water was provided *ad libitum* until premedication was given. All dogs were premedicated with acepromazine (0.03 mg/kg; Acezine 2; Ethical agents Ltd, New Zealand) and morphine (0.5 mg/kg; DBL morphine sulphate Injection BP; Hospira NZ Ltd, New Zealand) administered intramuscularly (IM). Intravenous catheters (Optiva IV catheter Radiopaque; Smiths Medical International Ltd, UK) were placed in both cephalic veins for routine use and study use. Anaesthesia was induced with propofol (Provive MCT-LCT 1%; Claris Lifesciences Australia Pty Ltd, Australia) intravenously (IV) until orotracheal intubation was achieved and maintained with isoflurane (Isoflurane; Bayer New Zealand Ltd, New Zealand) in 100% oxygen via a re-breathing circuit. All dogs were allowed to breath spontaneously and received intravenous fluid [Compound Sodium Lactate (Hartmann's Solution); Baxter Healthcare Ltd, Australia] via the routine catheter at a rate of 5 mL/kg/hr for the first hour and then 3 mL/kg/hour for the remainder of anaesthesia. After surgical preparation in an induction room, dogs were moved into an operating theatre and placed in dorsal recumbency. The dogs were maintained at normal temperature by a warm air blanket (Mistral-Air plus Warming unit; TSCI, Netherlands). After surgery and prior to recovery from anaesthesia, dogs received meloxicam IV (0.2 mg/kg; Metacam; Boehringer Ingelheim Vetmedica, Inc, MO, USA) and morphine IM (0.3 mg/kg). All dogs were discharged a day after the surgery or later the same day.

During anaesthesia, HR, mean arterial pressure (MAP), end-tidal isoflurane concentration ($F_E'Iso$), end-tidal carbon dioxide tension ($P_E'CO_2$), f_R, haemoglobin oxygen saturation, electrocardiogram and core temperature (T) were recorded every 5 minutes after the induction using a multi-parameter monitor (Cardell Max 12 HD; Midmark Corporation, OH, USA). MAP was measured via a 20-22 Ga catheter (Optiva IV catheter Radiopaque; Smiths

Medical International Ltd, UK) placed in a dorsal pedal artery immediately after anaesthesia induction using a disposable pressure transducer (BD DTXPlus: DTX Plus TNF-R; Becton Dickinson Critical Care Systems PTE Ltd, Singapore) connected to the multi-parameter monitor. The system was calibrated against a mercury manometer using a three-point calibration technique (0, 50 and 150 mmHg) and replaced between patients. The transducer was positioned at the level of the point of the shoulder and zeroed to atmospheric pressure. Clinical depth of anaesthesia (CDA) was judged and recorded every 5 minutes by an experienced anaesthetist using the definitions described by others (Bleijenberg, van Oostrom, Akkerdaas, Doornenbal, & Hellebrekers, 2011). CDA scores of 1, 2 and 3 indicated light anaesthesia, adequate surgical anaesthesia and an excessive depth of anaesthesia, respectively. After moving to the operating theatre, the measurement of CO was started using TEE (Toshiba Xario SSA-660A; Toshiba Medical Systems Corporation, Japan) at 5 minutes intervals. In order to facilitate optimal visualisation of longitudinal cranial-oesophageal aorta long-axis-view, the chest position of dogs was slightly tilted to the right side but did not interfere the surgery. The TEE probe was inserted through a plastic mouth gag into the cranial oesophagus with neutral position and advanced caudally with bent backward position until an image of the heart base was obtained. By setting the array angle to 75-85° and orienting the beam centrally, the longitudinal cranial-oesophageal aorta long-axis-view was obtained. The probe was fixed in position throughout the study. The sample volume was placed in the centre of ascending aorta to measure velocity time integral of the aorta at the end of the expiration. CO was calculated as the following: CO = HR × cross sectional area of the aorta ×velocity time integral. Cardiac index (CI) was also calculated by dividing CO by body weight. CO was measured in triplicate and averaged for the determination of the CO value every 5 minutes. Blood samples were taken from the arterial catheter immediately after the

catheter was placed and immediately after the end of anaesthesia to measure PCV, TS, creatinine, BG and lactate (epoc blood analysis system; Siemens Healthineers, Germany). All the equipment, including the multi-parameter monitor and the syringe pumps used in this study, were serviced, calibrated, and maintained at regular intervals by our staff biomechanical engineer.

Five minutes after induction as soon as the arterial catheter was placed, noradrenaline (Noradrenaline BNM; BNM group, Australia) infusion started at a rate of 0.05 μg/kg/min (0.5 mL/kg/hr)] via the study catheter throughout the anaesthesia in the noradrenaline group, while saline infusion was given at the same rate in the control group via the study catheter. In order to blind anaesthetists, an unlabelled perfusion pump was used to deliver either noradrenaline or saline. When hypertension was detected (MAP > 120 mmHg), noradrenaline or saline infusion was stopped. When hypertension was considered to be caused by light anaesthesia, F_E'Iso was increased by 0.25%. When spontaneous body movements were noticed, propofol (0.5-1.0 mg/kg) was administered IV and F_E'Iso increased by 0.1-0.5%. If hypertension was still present, morphine (0.3 mg/kg; DBL morphine sulphate Injection BP; Hospira NZ Ltd, New Zealand) was administered IV over 3 minutes. When hypotension, defined as MAP < 65 mmHg, was noted, the following actions were taken:

1. Decrease anaesthetic depth by reducing F_E'Iso by 0.25%;
2. Administer glycopyrrolate (0.005 mg/kg; Glycopyrronium Bromide; Martindale Pharmaceuticals Ltd, UK) IV if bradycardia (defined as HR < 80% of the preoperative HR) was present;
3. Provide an IV bolus of Hartmann's solution (5 mL/kg) over 5 minutes and repeat once if hypotension still existed;

4. Slowly administer 6% hydroxyethyl starch (3 mL/kg; Voluven 6%; Fresenius Kabi Australia Pty Ltd, Australia) over 5 minutes if hypertension persisted;
5. Start to administer noradrenaline at a rate of 0.05 μg/kg/min if the response to fluid boluses was inadequate. All these treatments were performed via the routine catheter.

Duration of anaesthesia from induction to extubation, duration of surgery, number of dogs administered anticholinergics, rescue doses of propofol, fluid boluses, and additional noradrenaline infusion, number of hypotensive dogs, total hypotensive time and total perianaesthetic fluid volume were recorded. Any signs of postoperative complications such nausea, vomiting, diarrhoea, arrhythmias, abdominal distension, haemorrhage, wound dehiscence were monitored by an attending clinician immediately after the surgery (which was in the morning), in the evening when dogs were fed, and the next morning unless discharged on the same day as the study.

Statistics

Data were tested for normality by the Shapiro–Wilk test and expressed with medians with ranges for continuous variables or frequencies for categorical ones. HR, MAP, CI, f_R, $P_E'CO_2$, $F_E'Iso$ and T recorded at 5 minutes intervals during anaesthesia were averaged for each dog and used for subsequent analysis. The significance of differences between groups was tested with Mann–Whitney U test for continuous variables and with the Fisher exact or the chi-square test for categorical data. Relative risks and 95% confidence intervals were also calculated.

A linear mixed-effects model, accounting for clustering due to repeated measurements on each dog, was fitted to the data using the lme4 package in R (R version 3.5.3; R Foundation for Statistical Computing, Vienna, Austria). The linear mixed model included the fixed effects of treatment (noradrenaline vs control group), anaesthesia/surgery period and their interaction plus dog as a random effect. The explanatory variable, anaesthesia/surgery period was coded as an ordered factor with T0 (5 minutes after the induction), T1 (10 minutes after the infusion started), T2 (before start of the surgery), T3 (time of skin incision), T4 (time of removal of the first ovary), T5 (time of removal of the second ovary), T6 (time of closure of the abdominal wall), and T7 (immediately after the skin was sutured). Diagnostic residual and random-effects plots were examined to assess adherence to model assumptions of within-group errors that are normally distributed, were centred at zero and had constant variance and random effects that were normally distributed, with mean zero and were independent for different groups. In addition, an analysis of deviance with its accompanying Wald test was carried out to indicate the quality of each model fit to the data. CDA scores were analysed using a cumulative link mixed model from the Ordinal package in R. The data in the model were expressed as means ± standard error (SE). Significance was set at $p < 0.05$.

RESULTS

Of 56 recruited dogs, 50 participated in the study and six were withdrawn: five needed additional sedative drugs such as dexmedetomidine and ketamine in addition to the premedication and one was unexpectedly pregnant. All surgeries were uneventful, and all dogs recovered from anaesthesia unremarkably. No differences between groups were detected for preoperative characteristics of dogs although breeds were varied (Table 6.1).

Table 6.1 Preoperative characteristics of dogs between noradrenaline and control groups. Breed is expressed as breed name (number of dogs). Other values are medians (range). BCS, body condition score; HR, heart rate; f_R, respiratory rate; T, temperature; PCV, packed cell volume; TS, total solids; BG, blood glucose; BUN, blood urea nitrogen. BUN was 5-15 mg/dL for all dogs.

Characteristic	Group		*p*-Value
	Noradrenaline (*n* = 25)	Control (*n* = 25)	
Breed	Cross breed (6) Labrador Retriever (4) Golden Retriever (3) Huntaway (3) Bulldog (2) Mastiff (2) Siberian Husky (2) Collie (1) Poodle (1) Rottweiler (1)	Cross breed (7) Labrador Retriever (4) Huntaway (3) Collie (2) German Shepard (2) Rottweiler (2) Wlsh Corgi (2) Bulldog (1) Australian Kelpie (1) Shar Pei (1)	NA
Age (years)	0.8 (0.3-3.0)	0.6 (0.3-3.0)	0.127
Weight (kg)	23.0 (10.5-34.5)	19 (8.4-35.5)	0.177
BCS (1-9)	6 (4-8)	6 (4-8)	0.406
HR (beats/minute)	99 (68-149)	97 (69-150)	0.915
f_R (breaths/minute)	22 (12-50)	19 (12-62)	0.195
T (°C)	38.8 (37.9-39.8)	38.8 (37.8-39.5)	0.741
PCV (%)	42 (29-50)	40 (33-55)	0.521
TS (g/L)	68 (49-80)	67 (52-75)	0.409
BG (mmol/L)	4.9 (3.9-6.6)	4.8 (4.2-6.9)	0.712
BUN (mg/dL)	5-15	5-15	NA

Median averaged MAP and $F_E'Iso$ were significantly greater and HR was lower in the noradrenaline group than those in the control group, while no differences were detected for CI, f_R, $P_E'CO_2$, CDA, and T between groups (Table 6.2). Durations of anaesthesia and surgery were not different between groups. Number of dogs administered rescue anaesthetics, fluid boluses and additional noradrenaline, total perianaesthetic fluid volume, number of hypotensive dogs and total hypotensive time were significantly lower in the noradrenaline group than those in the control group (Table 6.2). Three out of 10 dogs who received the additional noradrenaline infusion required cessation of the infusion due to hypertension in the control group, but no dogs needed to stop the noradrenaline infusion in the noradrenaline group. No dogs required morphine administration during the study.

Table 6.2 Intraoperative values from dogs undergoing ovariohysterectomy between noradrenaline and control groups. Values are medians (range). HR, heart rate; MAP, mean arterial pressure; CI, cardiac index; f_R, respiratory rate; $P_E'CO_2$, partial pressure of end-tidal carbon dioxide; F_E'Iso, End-tidal isoflurane concentration; CDA, Clinical depth of anaesthesia; T, temperature.

Variables	Group		RRs	95% CI	*p*-Value
	Noradrenaline (*n* = 25)	Control (*n* = 25)			
HR (beats/minute)	71 (62-114)	100 (73-118)			0.001*
MAP (mmHg)	86 (72-109)	72 (67-88)			0.001*
CI (mL/minute/kg)	100 (78-116)	97 (79-146)			0.503
f_R (breaths/minute)	11 (9-20)	13 (9-24)			0.082
$P_E'CO_2$ (mmHg)	43 (37-50)	44 (38-50)			0.907
F_E'Iso (%)	1.3 (1.2-1.4)	1.2 (1.1-1.4)			0.001*
CDA 1(%)	10 (3-100)	17 (9-100)			
2 (%)	82 (0-97)	78 (0-91)			
3 (%)	0 (0-30)	0 (0-14)			
T (°C)	37.7 (36.4-38.3)	37.5 (36.5-38.2)			0.085
Duration of anaesthesia (minute)	170 (115-190)	160 (115-180)			0.521
Duration of surgery (minute)	110 (70-145)	105 (70-135)			0.391
Number of dogs administered anticholinergics	5/25	3/25	1.67	0.45-6.24	0.702
Number of dogs administered rescue-anaesthetics	11/25	20/25	0.55	0.34-0.89	0.019*
Number of hypotensive dogs	5/25	16/25	0.31	0.14-0.72	0.004*
Total hypotensive time (minute)	0 (0-15)	15 (0-45)			0.001*
Number of dogs administered fluid boluses	1/25	12/25	0.08	0.01-0.59	0.001*
Total perianaesthetic fluid volume (mL/kg)	10.5 (7.8-14.5)	11 (7.8-24.0)			0.013*
Number of dogs administered additional noradrenaline	0/25	10/25			0.001*

The initial model included the explanatory variables age, body weight and body condition score of dogs and were removed from subsequent models as found to have no significant effect. Overall, MAP was significantly greater ($p < 0.001$) and HR was significantly lower ($p < 0.001$) in the noradrenaline group than those in the control group, while CI was not different between groups ($p = 0.118$) (Figure 6.1). F_E'Iso in the noradrenaline group was significantly higher than that in the control group ($p < 0.001$) although to account for repeated measures in the CDA data, there was no effect of treatment with a cumulative link mixed model ($p = 0.663$) (Figure 6.2). PCV ($p < 0.001$), TS ($p < 0.001$) and BG ($p = 0.023$) were significantly lower in the control group than those in the noradrenaline group after the surgery, while there was no effect of treatment on lactate ($p = 0.116$) (Figure 6.3). Difference in creatinine between groups almost reached significance ($p = 0.060$) (Figure. 6.3).

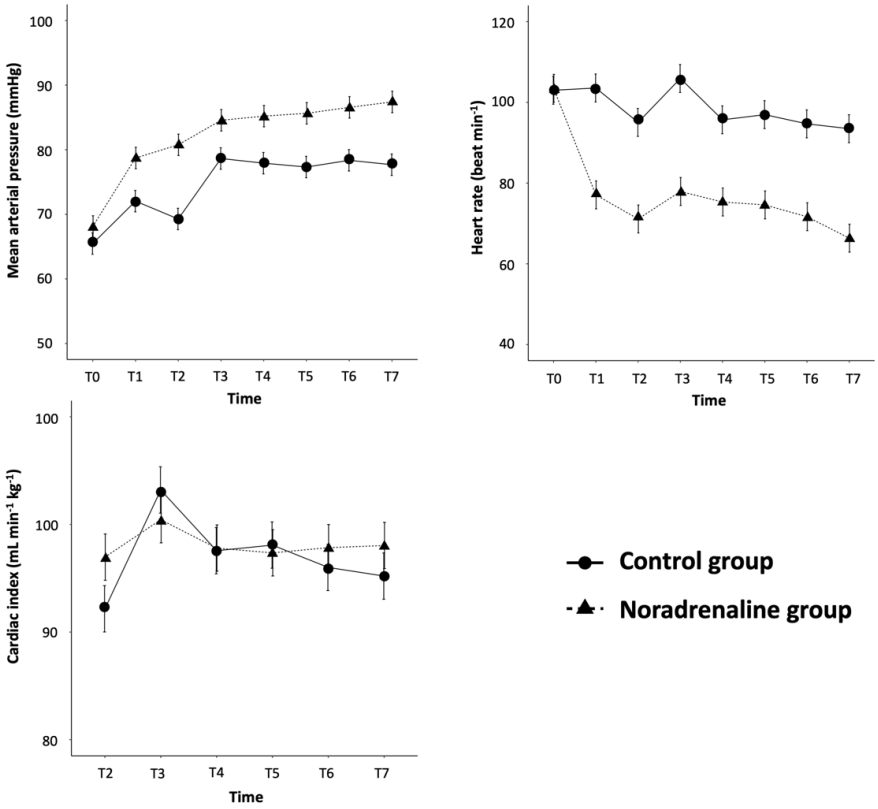

Figure 6.1 Mean ± SE haemodynamic parameters over the time points. T0, 5 minutes after the induction; T1, 10 minutes after the infusion started; T2 ,before start of the surgery; T3 ,time of skin incision; T4 ,time of removal of the first ovary; T5 ,time of removal of the second ovary; T6 ,time of closure of the abdominal wall; T7 ,immediately after the skin was sutured. Overall, mean arterial pressure was significantly greater ($p < 0.01$) and heart rate was significantly lower ($p < 0.01$) in the noradrenaline group than those in the control group, while cardiac index was no significant between groups ($p = 0.12$).

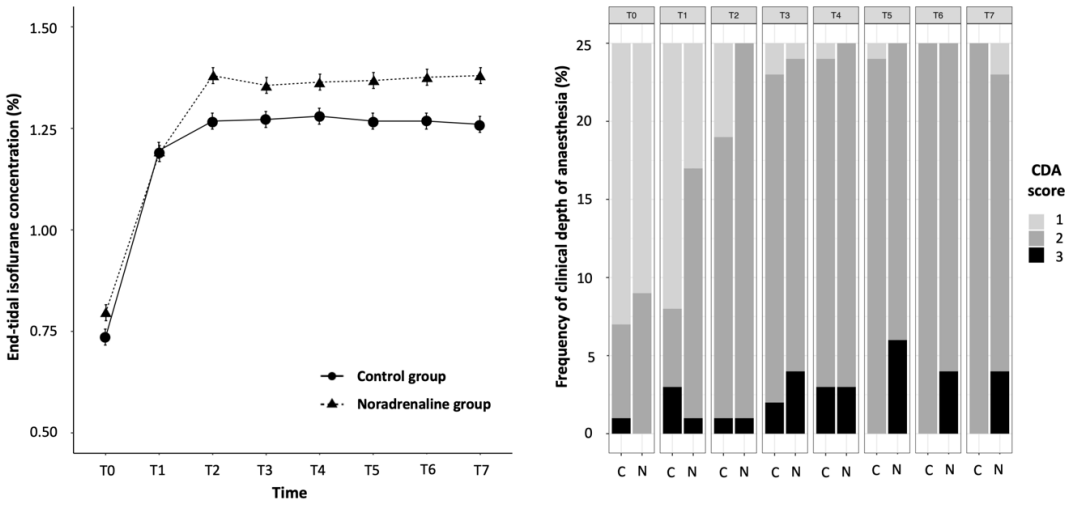

Figure 6.2 Mean ± SE end-tidal isoflurane concentration (Left) and frequency of clinical depth of anaesthesia (CDA) scores (Right) over the time points. T0, 5 minutes after the induction; T1, 10 minutes after the infusion started; T2 ,before start of the surgery; T3 ,time of skin incision; T4 ,time of removal of the first ovary; T5 ,time of removal of the second ovary; T6 ,time of closure of the abdominal wall; T7 ,immediately after the skin was sutured. CDA scores of 1, 2 and 3 indicated light anaesthesia, adequate surgical anaesthesia and an excessive depth of anaesthesia, respectively. C, control group; N. norepinephrine group. End-tidal isoflurane concentration in the noradrenaline group was significantly higher than that in the control group ($p < 0.01$) although to account for repeated measures in the CDA data, there was no effect of treatment with a cumulative link mixed model ($p = 0.66$).

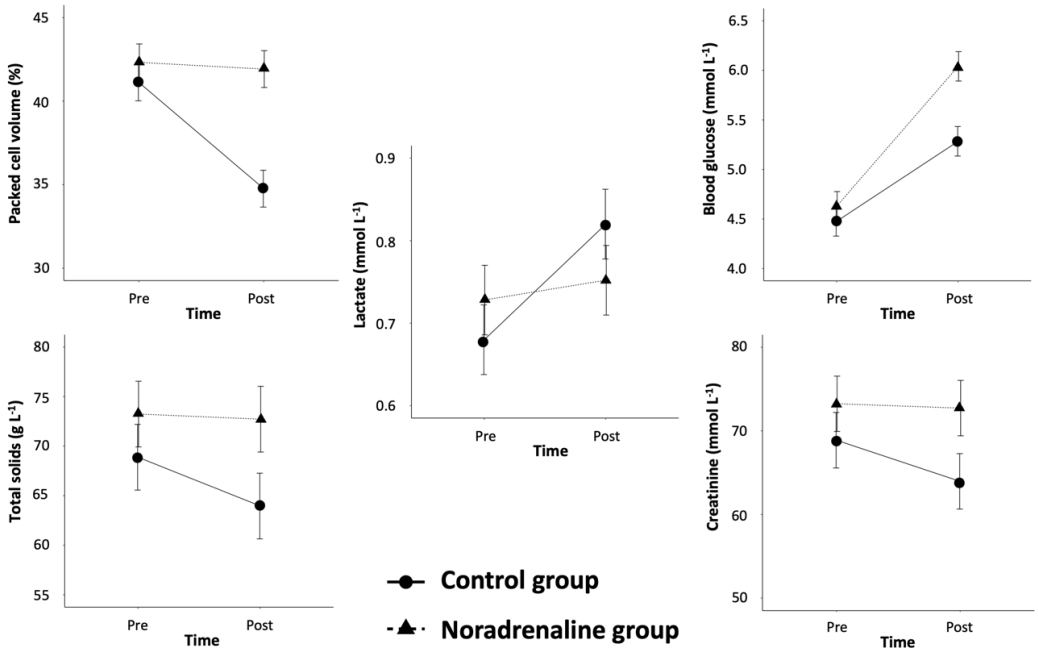

Figure 6.3 Mean ± SE haematology parameters before and after the surgery. Blood samples were taken from the arterial catheter immediately after the catheter was placed (Pre) and immediately after the end of anaesthesia (Post). Packed cell volume ($p < 0.001$), total solids ($p < 0.001$) and blood glucose ($p = 0.023$) were significantly lower in the control group than those in the noradrenaline group after the surgery, while there was no effect of treatment on lactate ($p = 0.116$) and creatinine between groups ($p = 0.06$).

There were no complications reported before discharge in either group in our study. Bradycardia with occasional 1st and 2nd AV block was noted during anaesthesia in 5 dogs of the noradrenaline group. This was not treated because MAP was well maintained and the frequency of the 2nd AV block was less than 2 per minute.

DISCUSSION

The findings in this study are that noradrenaline infusion prior to hypotension provided a lower incidence of hypotension, lower incidence of light plane of anaesthesia and less administered perioperative fluid volume compared to the conventional anaesthetic management, resulting in stable anaesthesia in dogs undergoing ovariohysterectomy.

Noradrenaline works primarily on α1 adrenergic receptors to cause vasoconstriction and weakly on β1 adrenergic receptors to increase cardiac contractility, resulting in an increase in blood pressure and preserved CO (Bellomo et al., 1999). In this study, greater MAP and lower HR were observed in the noradrenaline group than those in the control group. Noradrenaline probably increased systemic vascular resistance, leading to greater MAP and lower HR induced by baroreflex. There were no significant changes or differences between groups in CI over the study. A previous study showed that stroke volume increased as preload increased due to venous vasoconstriction and increased diastolic volume, and increased contractility even though afterload increased due to the vasoconstriction induced by activation of alpha 1 adrenergic receptors (Karzai et al., 1995). In addition, acepromazine, an alpha 1 adrenergic receptor antagonist, given as premedication might attenuate the vasoconstrictive effect, resulting in only a slight increase in afterload and maintained CI in this study. Because there may be reduced gastrointestinal perfusion due to vasoconstriction, the incidence of post-operative nausea, vomiting, diarrhoea and ileus were monitored. However, there were no complications seen before discharge in either group. Any adverse effects on the gastrointestinal tract were likely to be hidden by the effects of morphine. In animals with subclinical heart disease, noradrenaline may decrease CO or increase valvular regurgitation by increasing afterload. In general, veterinary practice, availability of a fluid

pump and cost of drugs may limit the use of this technique. Further studies to investigate potential risks and adverse effect of pre-emptive noradrenaline infusion are necessary in veterinary practices.

Intraoperative awareness and body movement caused by a light plane of anaesthesia is always undesirable. If reducing end-tidal concentration of anaesthetic agents is used to increase blood pressure, there is a risk of awareness. In this study, the noradrenaline group had higher F_E´Iso [1.3 % (1.2-1.4) vs 1.2 % (1.1-1.4)] and lower incidence of additional propofol administration over the anaesthesia period than the control group although there was no difference in CDA between groups. As MAP was maintained in the noradrenaline group, the anaesthetist probably considered it unnecessary to decrease isoflurane concentration to increase blood pressure as often, indicating that anaesthesia in these dogs was slightly deeper but more stable. However, F_E´Iso in the noradrenaline group was not clinically high in this study. Therefore, prevention of hypotension can contribute to high quality of anaesthesia.

Intraoperative fluid therapy is important to improve haemodynamics and thus oxygen delivery. In anaesthetised patients, a bolus of fluid is believed to increase blood pressure by filling additional intravascular space caused by vasodilatation due to anaesthetic agents. However, in dogs with euvolemic isoflurane-induced hypotension, rapid administration of fluid (60 - 80 mL/kg/hr of crystalloid or colloid) did not increase arterial blood pressure and caused haemodilutional effects (Aarnes et al., 2009; Valverde et al., 2012). In this study, intravenous bolus of 5 mL/kg of crystalloid and 3 mL/kg of colloid were administered in addition to the maintenance fluid when hypotension persisted. In the control group, 12 out of 25 dogs needed these fluids, however, 10 out of these 12 dogs also required noradrenaline

infusion. This indicated that a 3-5mL/kg bolus of fluid was ineffective in dogs with presumably euvolemic hypotension. Excess fluid administration can increase complication rate and mortality in human medicine (Ketharanathan et al., 2014; Wang, Jiang, Zhu, Wen, & Xi, 2015; Willson et al., 2013; Wuethrich et al., 2014). A possible reason could be glycocalyx injury, resulting in interstitial or intracellular oedema impeding delivery of oxygen and nutrients. Thus, unnecessary fluid administration should be avoided. In this study, the noradrenaline group had fewer dogs requiring fluid boluses, and less total intraoperative fluid volume than the control group. In the 12 dogs that received a fluid bolus in the control group, the only 2 dogs had improved blood pressure and the others needed noradrenaline infusion.

Finally, PCV and TS were significantly reduced in the control group compared to the noradrenaline group. While there may be some benefits from haemodilution causing reduced viscosity and thus increased flow through capillaries, excessive fluid administration has been shown to increase morbidity presumably by dilution of haematocrit and plasma proteins (Tsui et al., 2010). Therefore, routine noradrenaline infusion could prevent excessive fluid therapy and thus haemodilution.

In terms of effect on blood profiles in this study, PCV and TP significantly decreased in the control group compared to the noradrenaline group. This may have been mainly due to haemodilution, but splenic constriction caused by noradrenaline (Davies, Gamble, & Withrington, 1973) could also have contributed. Blood glucose significantly increased in the noradrenaline group as catecholamines can increase blood glucose (Chu, Sindelar, Neal, & Cherrington, 1996). In people, lower intraoperative and postoperative lactate levels were

shown in patients who received less intraoperative fluid (Forget, Lois, & de Kock, 2010). However, lactate and creatinine were unchanged in both groups. This might be because CI in anaesthetised dogs was maintained and duration and magnitude of hypotension might not have been enough to cause tissue hypoxia.

Similar studies have been conducted in people. One showed that noradrenaline infusion prevented hypotension associated with spinal and epidural anaesthesia (Fu et al., 2020). Another study concluded that noradrenaline infusion combined with lower fluid administration during radical cystectomy and urinary diversion significantly decreased postoperative complication rate, mortality and hospitalisation time (Wuethrich et al., 2014). In Wuethrich's study, whether the noradrenaline infusion reduced the fluid volume administered was not evaluated because the study protocol restricted fluid administration in the treatment group. However, low fluid volume administered was one of factors associated with reduced postoperative complications. In our study, in-hospital complications were not observed in either groups even though the noradrenaline group was given less fluid. Unlike Wuethrich et al. (Wuethrich et al., 2014), MAP was greater, HR was lower in the noradrenaline group and no differences in CI were observed. Our study did not restrict fluid administration in either group, but the control group was given additional fluid boluses. Therefore, CI and MAP were well maintained, and more reflex bradycardia was observed.

This study has several limitations. It was carried out on the assumption that healthy dogs with ASA I were not dehydrated. The pre-emptive administration of noradrenaline infusion for dogs more than ASA 2 should be cautious with blood pressure monitoring and ECG for arrhythmias. Particularly, dehydration or hypovolaemia should be ruled out before this

technique is used though it is difficult to assess hydration status accurately in animals. All dogs in this study were in dorsal recumbency throughout the study. Other positions and changes in position under anaesthesia might contribute to hypotension but this study did not account for positional effects. Noradrenaline infusion prior to the anaesthesia induction might have reduced the incidence of hypotension because hypotension was likely to happen just after induction (Cattai, Rabozzi, Ferasin, Isola, & Franci, 2018), but it was challenging to record all cardiovascular parameters in conscious dogs in the clinical setting. The control group had lower blood pressure than the noradrenaline group. This was because the control group received more propofol than the noradrenaline group (20/25 vs 11/25, $p = 0.019$). However, all dogs who received propofol had spontaneous body movements just because of light plane of anaesthesia. Therefore, noradrenaline could prevent additional propofol administration in the noradrenaline group to prevent hypotension. Postoperative complications and blood profile including lactate and creatinine were unknown as the dogs went home the same or next day, and post-operative monitoring was limited. Sample size was too small to detect any post anaesthetic complication rate. In this study, CO was measured by TEE as it is minimally invasive and provides good agreement with the thermodilution technique in dogs (Mantovani et al., 2017; Yamashita et al., 2007). However, the CO obtained by TEE was less reliable during hypotension although it is still clinically acceptable (Mantovani et al., 2017). Moreover, the trending ability of transoesophageal echocardiography to measure CO is unknown in dogs although a transoesophageal doppler device accurately reflected the direction and magnitude of the changes of CO over time during abrupt haemodynamic changes in dogs (de Figueiredo, Cruz, Silva, & Rocha, 2004). In this study, CI at T0 and T1 were not measured because of limited availability of the ultrasound machine. However overall there was no significant difference between groups.

The definition of hypotension in this study was MAP < 65 mmHg. However, 2020 AAHA Anesthesia and Monitoring Guidelines for Dogs and Cats defines hypotension as MAP < 60 - 70 mmHg (Grubb et al., 2020). Therefore, if the lower figure of 60 mmHg is used, some dogs in this study did not have hypotension. In terms of anaesthetic depth, there was a conflicting result between F_E'Iso and CDA. However, CDA might not be sensitive enough to accurately detect small changes in anaesthetic depth because it is subjective. Even though adequate anaesthetic depth was presumed, dogs sometimes became light unexpectedly.

In conclusion, in this study, noradrenaline infusion prior to hypotension provided a lower incidence of hypotension, lower incidence of light plane of anaesthesia and less perioperative fluid volume administered than the conventional anaesthetic management, resulting in stable anaesthetic management in dogs undergoing ovariohysterectomy. Therefore, low-dose noradrenaline infusion prior to hypotension is likely to be beneficial in fit and healthy young dogs in general veterinary anaesthesia. However, fluid administration or decreasing depth of anaesthesia is still necessary when obvious dehydration, hypovolaemia or profound depth of anaesthesia is recognised.

References

Aarnes, T. K., Bednarski, R. M., Lerche, P., Hubbell, J. A., & Muir, W. W., 3rd. (2009). Effect of intravenous administration of lactated Ringer's solution or hetastarch for the treatment of isoflurane-induced hypotension in dogs. *Am J Vet Res, 70*(11), 1345-1353. doi:10.2460/ajvr.70.11.1345

Bednarski, R., Grimm, K., Harvey, R., Lukasik, V. M., Penn, W. S., Sargent, B., & Spelts, K. (2011). AAHA anesthesia guidelines for dogs and cats. *J Am Anim Hosp Assoc, 47*(6), 377-385. doi:10.5326/jaaha-ms-5846

Bellomo, R., Kellum, J. A., Wisniewski, S. R., & Pinsky, M. R. (1999). Effects of norepinephrine on the renal vasculature in normal and endotoxemic dogs. *Am J Respir Crit Care Med, 159*(4 Pt 1), 1186-1192. doi:10.1164/ajrccm.159.4.9802055

Bleijenberg, E. H., van Oostrom, H., Akkerdaas, L. C., Doornenbal, A., & Hellebrekers, L. J. (2011). Bispectral index and the clinically evaluated anaesthetic depth in dogs. *Vet Anaesth Analg, 38*(6), 536-543. doi:10.1111/j.1467-2995.2011.00651.x

Brodbelt, D. (2009). Perioperative mortality in small animal anaesthesia. *Vet J, 182*(2), 152-161. doi:10.1016/j.tvjl.2008.06.011

Cattai, A., Rabozzi, R., Ferasin, H., Isola, M., & Franci, P. (2018). Haemodynamic changes during propofol induction in dogs: new findings and approach of monitoring. *BMC Vet Res, 14*(1), 282. doi:10.1186/s12917-018-1608-8

Chu, C. A., Sindelar, D. K., Neal, D. W., & Cherrington, A. D. (1996). Direct effects of catecholamines on hepatic glucose production in conscious dog are due to glycogenolysis. *Am J Physiol, 271*(1 Pt 1), E127-137. doi:10.1152/ajpendo.1996.271.1.E127

Davies, B. N., Gamble, J., & Withrington, P. G. (1973). Frequency-dependent differences in the responses of the capsular and vascular smooth muscle of the spleen of the dog to sympathetic nerve stimulation. *J Physiol, 228*(1), 13-25. doi:10.1113/jphysiol.1973.sp010069

Davis, H., Jensen, T., Johnson, A., Knowles, P., Meyer, R., Rucinsky, R., & Shafford, H. (2013). 2013 AAHA/AAFP fluid therapy guidelines for dogs and cats. *J Am Anim Hosp Assoc, 49*(3), 149-159. doi:10.5326/jaaha-ms-5868

de Figueiredo, L. F., Cruz, R. J., Jr., Silva, E., & Rocha, E. S. M. (2004). Cardiac output determination during experimental hemorrhage and resuscitation using a transesophageal Doppler monitor. *Artif Organs, 28*(4), 338-342. doi:10.1111/j.1525-1594.2004.47351.x

Ferguson-Myrthil, N. (2012). Vasopressor use in adult patients. *Cardiol Rev, 20*(3), 153-158. doi:10.1097/CRD.0b013e31824e2294

Forget, P., Lois, F., & de Kock, M. (2010). Goal-directed fluid management based on the pulse oximeter-derived pleth variability index reduces lactate levels and improves fluid management. *Anesth Analg, 111*(4), 910-914. doi:10.1213/ANE.0b013e3181eb624f

Fu, F., Xiao, F., Chen, W., Yang, M., Zhou, Y., Ngan Kee, W. D., & Chen, X. (2020). A randomised double-blind dose-response study of weight-adjusted infusions of norepinephrine for preventing hypotension during combined spinal-epidural anaesthesia for Caesarean delivery. *Br J Anaesth, 124*(3), e108-e114. doi:10.1016/j.bja.2019.12.019

Gaynor, J. S., Dunlop, C. I., Wagner, A. E., Wertz, E. M., Golden, A. E., & Demme, W. C.

(1999). Complications and mortality associated with anesthesia in dogs and cats. *J Am Anim Hosp Assoc, 35*(1), 13-17. doi:10.5326/15473317-35-1-13

Grubb, T., Sager, J., Gaynor, J. S., Montgomery, E., Parker, J. A., Shafford, H., & Tearney, C. (2020). 2020 AAHA Anesthesia and Monitoring Guidelines for Dogs and Cats. *J Am Anim Hosp Assoc, 56*(2), 59-82. doi:10.5326/jaaha-ms-7055

Hiltebrand, L. B., Koepfli, E., Kimberger, O., Sigurdsson, G. H., & Brandt, S. (2011). Hypotension during fluid-restricted abdominal surgery: effects of norepinephrine treatment on regional and microcirculatory blood flow in the intestinal tract. *Anesthesiology, 114*(3), 557-564. doi:10.1097/ALN.0b013e31820bfc81

Holte, K., Sharrock, N. E., & Kehlet, H. (2002). Pathophysiology and clinical implications of perioperative fluid excess. *Br J Anaesth, 89*(4), 622-632. doi:10.1093/bja/aef220

Iizuka, T., Kamata, M., Yanagawa, M., & Nishimura, R. (2013). Incidence of intraoperative hypotension during isoflurane-fentanyl and propofol-fentanyl anaesthesia in dogs. *Vet J, 198*(1), 289-291. doi:10.1016/j.tvjl.2013.06.021

Karzai, W., Reilly, J. M., Hoffman, W. D., Cunnion, R. E., Danner, R. L., Banks, S. M., . . . Natanson, C. (1995). Hemodynamic effects of dopamine, norepinephrine, and fluids in a dog model of sepsis. *Am J Physiol, 268*(2 Pt 2), H692-702. doi:10.1152/ajpheart.1995.268.2.H692

Ketharanathan, N., McCulloch, M., Wilson, C., Rossouw, B., Salie, S., Ahrens, J., . . . Argent, A. C. (2014). Fluid overload in a South African pediatric intensive care unit. *J Trop Pediatr, 60*(6), 428-433. doi:10.1093/tropej/fmu041

Lobo, D. N., Macafee, D. A., & Allison, S. P. (2006). How perioperative fluid balance influences postoperative outcomes. *Best Pract Res Clin Anaesthesiol, 20*(3), 439-455.

Mantovani, M. M., Fantoni, D. T., Gimenes, A. M., de Castro, J. R., Flor, P. B., Ida, K. K., & Schwartz, D. S. (2017). Clinical monitoring of cardiac output assessed by transoesophageal echocardiography in anaesthetised dogs: a comparison with the thermodilution technique. *BMC Vet Res, 13*(1), 325. doi:10.1186/s12917-017-1227-9

Monk, T. G., Saini, V., Weldon, B. C., & Sigl, J. C. (2005). Anesthetic management and one-year mortality after noncardiac surgery. *Anesth Analg, 100*(1), 4-10. doi:10.1213/01.Ane.0000147519.82841.5e

Osugi, T., Tatara, T., Yada, S., & Tashiro, C. (2011). Hydration status after overnight fasting as measured by urine osmolality does not alter the magnitude of hypotension during general anesthesia in low risk patients. *Anesth Analg, 112*(6), 1307-1313. doi:10.1213/ANE.0b013e3182114df4

Redondo, J. I., Rubio, M., Soler, G., Serra, I., Soler, C., & Gomez-Villamandos, R. J. (2007). Normal values and incidence of cardiorespiratory complications in dogs during general anaesthesia. A review of 1281 cases. *J Vet Med A Physiol Pathol Clin Med, 54*(9), 470-477. doi:10.1111/j.1439-0442.2007.00987.x

Reich, D. L., Hossain, S., Krol, M., Baez, B., Patel, P., Bernstein, A., & Bodian, C. A. (2005). Predictors of hypotension after induction of general anesthesia. *Anesth Analg, 101*(3), 622-628, table of contents. doi:10.1213/01.Ane.0000175214.38450.91

Rui, Q., Jiang, Y., Chen, M., Zhang, N., Yang, H., & Zhou, Y. (2017). Dopamine versus norepinephrine in the treatment of cardiogenic shock: A PRISMA-compliant meta-analysis. *Medicine (Baltimore), 96*(43), e8402. doi:10.1097/md.0000000000008402

Sandifer, J. P., & Jones, A. E. (2012). Dopamine versus norepinephrine for the treatment of septic shock EBEM commentators. *Ann Emerg Med, 60*(3), 372-373. doi:10.1016/j.annemergmed.2012.04.012

Snowdon, K. A., Smeak, D. D., & Chiang, S. (2016). Risk Factors for Dehiscence of Stapled Functional End-to-End Intestinal Anastomoses in Dogs: 53 Cases (2001-2012). *Vet Surg, 45*(1), 91-99. doi:10.1111/vsu.12413

Tsui, A. K., Dattani, N. D., Marsden, P. A., El-Beheiry, M. H., Grocott, H. P., Liu, E., . . . Hare, G. M. (2010). Reassessing the risk of hemodilutional anemia: Some new pieces to an old puzzle. *Can J Anaesth, 57*(8), 779-791. doi:10.1007/s12630-010-9329-x

Turk, R., Singh, A., & Weese, J. S. (2015). Prospective surgical site infection surveillance in dogs. *Vet Surg, 44*(1), 2-8. doi:10.1111/j.1532-950X.2014.12267.x

Valverde, A., Gianotti, G., Rioja-Garcia, E., & Hathway, A. (2012). Effects of high-volume, rapid-fluid therapy on cardiovascular function and hematological values during isoflurane-induced hypotension in healthy dogs. *Can J Vet Res, 76*(2), 99-108.

Wang, N., Jiang, L., Zhu, B., Wen, Y., & Xi, X. M. (2015). Fluid balance and mortality in critically ill patients with acute kidney injury: a multicenter prospective epidemiological study. *Crit Care, 19*, 371. doi:10.1186/s13054-015-1085-4

Willson, D. F., Thomas, N. J., Tamburro, R., Truemper, E., Truwit, J., Conaway, M., . . . Egan, E. E. (2013). The relationship of fluid administration to outcome in the pediatric calfactant in acute respiratory distress syndrome trial. *Pediatr Crit Care Med, 14*(7), 666-672. doi:10.1097/PCC.0b013e3182917cb5

Wuethrich, P. Y., Burkhard, F. C., Thalmann, G. N., Stueber, F., & Studer, U. E. (2014). Restrictive deferred hydration combined with preemptive norepinephrine infusion during radical cystectomy reduces postoperative complications and hospitalization time: a randomized clinical trial. *Anesthesiology, 120*(2), 365-377. doi:10.1097/ALN.0b013e3182a44440

Yamashita, K., Ueyama, Y., Miyoshi, K., Igarashi, R., Kushiro, T., Umar, M. A., & Muir, W. W. (2007). Minimally invasive determination of cardiac output by transthoracic bioimpedance, partial carbon dioxide rebreathing, and transesophageal Doppler echocardiography in beagle dogs. *J Vet Med Sci, 69*(1), 43-47.

Chapter 7

General discussion and conclusions

PREFACE

Chapters 2 – 6 presented the results of a series of studies designed to investigate appropriate fluid administration to optimise haemodynamics without fluid overload in anaesthetised dogs. As each of these chapters has been previously published in peer-reviewed journals or presented at major anaesthesia conferences, the results are discussed within the chapter in which they are presented. Within this general discussion in **Chapter 7** the principal findings of the studies are summarised, discussion of important aspects of the results is further developed, general conclusion are drawn, and future directions are suggested.

Abbreviations

AUROC	area under receiver operating characteristic
CO	cardiac output (L/minute)
CVP	central venous pressure (mmHg)
esCO	estimated CO from PWTT (L/minute)
HR	heart rate (beat/minute)
MAP	mean arterial pressure (mmHg)
MFC	mini-fluid challenge
PPV	pulse pressure variation (%)
PVI	pleth Variability Index (%)
PWTT	pulse wave transit time (msecond)
SV	stroke volume (mL)
VTI	velocity time integral (cm)
95%CI	95% confidence interval

PRINCIPAL FINDINGS

The series of studies conducted in this thesis, evolved from the initial question, of how perioperative fluid administration can be performed to optimise haemodynamics without volume overload in dogs, because both hypovolaemia and hypervolaemia are known to cause increased perioperative morbidity and mortality in people (Doherty & Buggy, 2012; Navarro et al., 2015).

The study in Chapter 2 concluded that the pulse pressure variation (PPV) and the pleth variability index (PVI) predicted fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine, and they may be useful to guide fluid administration in veterinary practice (Sano, Seo, et al., 2018). PVI can be obtained non-invasively and simply, although PPV requires invasive blood pressure monitoring. However, the clinical use of PPV and PVI require tightly controlled conditions including mechanical ventilation, which is not commonly used in veterinary practice (Sano, Barker, et al., 2018). Therefore, alternative methods to predict fluid responsiveness should be investigated.

As **Chapter 1** described, the mini-fluid challenge (MFC) involves the assessment of a change in stroke volume (SV) in order to predict fluid responsiveness to a larger volume. A sufficient increase in SV following the MFC indicates that the dog is a fluid responder. However, measurement of SV in clinical veterinary practice is challenging. **Chapter 3** found that the pulse wave transit time (PWTT) has a good trending ability (the trends are similar even if the absolute numbers are slightly different) to detect changes in SV precisely in isoflurane-anaesthetised dogs (Sano & Chambers, 2017). This technique is easy to use, inexpensive, non-invasive and could become part of routine anaesthetic

monitoring. Therefore, ΔPWTT was proved to be a surrogate of ΔSV in clinical practice.

The study in **Chapter 4** was conducted concurrently with that described in chapter 2. This study concluded that 1) a 3 mL/kg of MFC was the minimum required for the assessment of fluid responsiveness, and 2) the MFC could predict fluid responsiveness based on ΔPWTT in mechanically ventilated anaesthetised dogs (Sano, Fujiyama, et al., 2019). Since the calculation of ΔPWTT requires only ECG and pulse oximeter data, this technique could be utilised in routine veterinary anaesthetic practice. However, the study in **Chapter 4** as well as **Chapter 2** were performed under tightly controlled experimental conditions with constant mechanical ventilation. Therefore, reproducibility of this technique in spontaneously breathing dogs in a clinical setting should be investigated before it can be adopted in practice.

Based on the studies in **Chapter 3 and 4**, a clinical trial for the MFC was conducted and reported in **Chapter 5**. The study showed that ΔPWTT following a MFC could predict fluid responsiveness non-invasively in spontaneously breathing anesthetised dogs undergoing stifle surgery. MFC using PWTT was shown to be capable of predicting fluid responsiveness in the clinical setting. An advantage of the MFC technique is that it can be used to determine the appropriate endpoint for fluid administration, minimising the risk of volume overload during anaesthesia. In a hypotensive animal, a MFC of 3 mL/kg can be repeated until there is no further response, at which point the administration of an inopressor is indicated if normotension has not been achieved. Furthermore, use of the MFC and PWTT in critically ill awake dogs could be potentially valuable but requires further study.

In veterinary clinical practice, the use of the MFC and PWTT still has limitations because of the difficulty of interpretation, and a requirement for the monitors to be able to calculate the PWTT in real time. In Chapter 6, I investigated the efficacy of noradrenaline intravenous infusion to prevent hypotension developing in dogs undergoing ovariohysterectomy (Sano, Chambers, & Bridges, 2019). Since the most common treatment for hypotension is fluid administration, prevention of hypotension by prophylactic noradrenaline would minimise fluid volume administered and thus the risk of overload. The conclusion of the study in **Chapter 6** was that a noradrenaline infusion resulted in a lower incidence of hypotension, a lower incidence of a light plane of anaesthesia, and a smaller volume of fluid administered intraoperatively than the conventional anaesthetic management, resulting in stable anaesthetic management in dogs undergoing ovariohysterectomy. Therefore, a prophylactic low-dose noradrenaline infusion is likely to be beneficial in fit and healthy young dogs in general veterinary anaesthesia.

DISCUSSION

The study in Chapter 2 showed that PPV and PVI with the cutoff values of 11% and 9.3% respectively could predict fluid responsiveness more accurately than measuring central venous pressure (CVP) in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine. When this study was conducted, there were no other published studies that had evaluated the ability of PPV and PVI to predict fluid responsiveness in dogs. However, over the last few years, several authors have published findings consistent with mine (Celeita-Rodríguez et al., 2019; Drozdzynska, Chang, Stanzani, & Pelligand, 2018; Fantoni et al., 2017). In those new studies, the cutoff values for PPV and PVI above which patients are considered to respond to the fluid administration are between 13 and 16% (Drozdzynska et al., 2018; Fantoni et al., 2017), and 11% (Celeita-Rodríguez et al., 2019) in mechanically ventilated anaesthetised dogs. The values in this study are comparable to these values.

All these results in dogs are comparable with studies in people, in which the cutoff values for PPV and PVI were reported to be 9.4 - 17% (Cannesson et al., 2011; Cavallaro, Sandroni, & Antonelli, 2008; Lopes et al., 2007), and 9.5 - 17% (D. H. Kim et al., 2018; Loupec et al., 2011; Zimmermann et al., 2010), respectively. In clinical practice, the inconsistent cutoff values for PPV and PVI reported in each study are not helpful particularly when PPV or PPV falls into these ranges. The values are influenced by not only the preload but also other factors such as heart rate (Morgan, Abel, Mullins, & Guntheroth, 1966), respiratory rate (De Backer & Vincent, 2018), pleural pressure (Liu et al., 2016), and tidal volume (Díaz, Erranz, Donoso, Salomon, & Cruces, 2015; H. K. Kim & Pinsky, 2008). In dogs, anatomical conformation, body size, and normal physiological parameters vary widely, and each condition may exacerbate this variation.

Thus, depending on breed and clinical condition, PPV and PVI could fluctuate, which might decrease their clinical utility (Marik & Lemson, 2014). All those factors need to be taken into account when PPV and PVI are used clinically. Therefore, PPV and PVI should not be a primary diagnostic tool yet because when a value might be inaccurate is unknown.

The study in Chapter 3 showed that the agreement between cardiac output (CO) estimated from PWTT (esCO) and reference CO measured by the thermodilution technique was clinically unacceptable in isoflurane-anaesthetised dogs, although the change in PWTT was able to detect trends in SV. The calculation of esCO is derived from a simple assumption that SV would be proportional to pulse pressure (a difference between systolic and diastolic arterial pressure), but it is proposed that this oversimplified assumption caused the disagreement with the reference CO. In contrast, the pulse contour analysis makes the more accurate assumption that the area under the arterial curve during systole, minus the background diastolic area, would be proportional to SV (Scolletta, Romano, Biagioli, Capannini, & Giomarelli, 2005).

However, the other finding of the study in Chapter 3 suggests that ΔPWTT may be clinically useful as a non-invasive method to track ΔSV in anaesthetised dogs using only routine cardiovascular monitoring. This could be a convenient tool to interpret haemodynamic changes in clinical practice. Current routine anaesthetic monitoring can only provide a small amount of haemodynamic information such as heart rate and blood pressure, but not SV, systemic vascular resistance and others (Figure 7.1). However, estimated ΔSV derived from ΔPWTT provide a direction of systemic vascular resistance change, which helps a clinician selecting either vasoconstrictors, inotropes, or fluid

administration. This non-invasive monitoring would be very advantageous to manage haemodynamically unstable patients.

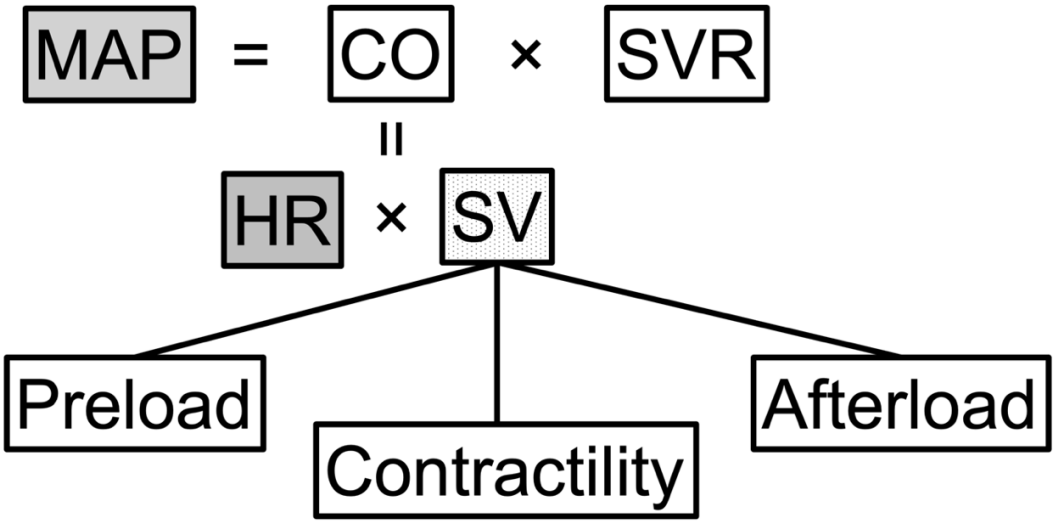

Figure 7.1 Clinical interpretation of haemodynamics. MAP can be calculated by CO and SVR, and CO can be calculated by HR and SV. SV is determined by preload, contractility and afterload. MAP and HR is only parameters that can be obtained clinically, which are not enough to comprehend haemodynamics. MAP; mean arterial pressure, CO; cardiac output, SVR; systemic vascular resistance, HR; heart rate, SV; stroke volume.

However, in Chapter 3, I found that some changes of the PWTT did not follow the same direction of the ΔSV induced by phenylephrine which suggests that vasoconstriction induced by phenylephrine may affect the measurement of the PWTT. Thus, the PWTT may not reflect SV accurately when alpha 2 agonists, which cause vasoconstriction, are used as premedication, or if vasoconstrictors are used in hypotensive patients. Therefore, the interpretation of ΔPWTT should be made with caution in the presence of vasoconstrictors.

The studies in Chapters 4 and 5 showed the feasibility of a MFC to predict fluid responsiveness in both mechanically ventilated and spontaneously breathing dogs, and these are the first studies of the MFC in dogs. The areas under the receiver operating characteristic curves (AUROC) for Δ velocity time integral (VTI) of the aortic or pulmonary blood flow following a MFC were 0.93 [95% confidence interval (95%CI) 0.79 – 1.00] and 0.91 (95%CI 0.79 – 0.98) with cutoff thresholds of 9.1% and 9% in mechanically ventilated and spontaneously breathing dogs, respectively. In people, a meta-analysis on the reliability of a MFC in predicting fluid responsiveness reported that the pooled AUROC for the MFC was 0.91 (95%CI 0.85 – 0.97) with a best median threshold of 5% (range: 3.0 – 7.0%) (Messina et al., 2019). which are comparable to the values in dogs in this thesis. Therefore, a MFC can be adopted in veterinary medicine. However, the measurement of VTI requires either transthoracic or oesophageal echocardiography. Transthoracic echocardiography would be difficult to perform because access to the chest during surgery is challenging or impossible, and variability depends on the expertise of the person doing the ultrasound. Oesophageal echocardiography is an ideal method to evaluate a MFC because a probe can be placed in the oesophagus easily and fixed at the same position during the MFC and is less dependent on individual

technique. However, the cost limits its use in clinical veterinary practice. Thus, it is unlikely that measurement of the VTI to evaluate a MFC will be widely adopted in veterinary practice.

The study in Chapter 3 suggests that ΔPWTT may be clinically useful as a non-invasive method to track ΔSV in anaesthetised dogs using only routine cardiovascular monitoring. Thus, main finding of Chapter 4 and 5 was that a reduction in the PWTT of 2.5% in mechanically ventilated and 2.1% in spontaneously breathing dogs over a MFC can predict fluid responsiveness with 0.91 (95%CI 0.80 – 1.00) and 0.94 (95%CI 0.83 - 0.99) of AUROC, respectively. Although there are no compatible data in people, these results indicated that it is a reliable method to predict fluid responsiveness in anaesthetised dogs.

In clinical practice, periodic intraoperative monitoring of ΔPWTT following a 3 mL/kg of MFC (for instance, every hour) would guide a clinician to assess fluid responsiveness in order to prevent hypo- and hypervolaemia in anaesthetised dogs. If done, routine maintenance fluid (generally 3 ml/kg/hr) is not required, because 3 mL/kg of MFC every hour can be the maintenance fluid. If an animal becomes hypotensive, whether fluid administration is required can be determined depending on fluid responsiveness accessed in the previous hourly MFC. In a hypotensive animal with positive fluid responsiveness, a MFC of 3 mL/kg can be repeated until there is no further response, at which point the administration of an inopressor is indicated if normotension has not been achieved. In a hypotensive animal with negative fluid responsiveness, an inopressor can be administered to improve hypotension. Caution must be taken when fluid responsiveness is interpreted because positive fluid responsiveness does not always indicate hypovolaemia, and negative fluid responsiveness does not necessarily indicate hypervolaemia. The

determination of fluid responsiveness is to assess whether the heart can increase SV in response to fluid administration but does not evaluate circulatory volume in the body. This is particularly important in haemodynamically unstable dogs with severely compromised conditions such as sepsis, haemoabdomen, and shock.

In Chapters 2, 4, and 5, cutoff values of the PPV, PVI, and the ΔPWTT following a MFC in both mechanically ventilated and spontaneously breathing dogs were determined in order to discriminate between fluid responders and non-responders. In addition, the grey zones were also calculated to determine a clinically inconclusive range, below which patients are unlikely to respond, above which they are expected to respond, but within which, it is uncertain. In these studies, around 24 - 33% of dogs fell into the grey zone. In a patient in the grey zone, comprehensive judgement from other clinical information such as history of the patient, underlying diseases, intraoperative information (visually apparent haemorrhage, surgical procedure), blood gas analysis, serum lactate, electrolytes, are required to predict fluid responsiveness. Lastly, disruption of endothelial glycocalyx layer has been shown to alter fluid exchange with tissue and is involved in extravascular oedema and inflammation formation (Alphonsus & Rodseth, 2014). Under those circumstances, PPV, PVI, and the ΔPWTT following a MFC may not be functional, and always indicate non-responder even though circulating volume is depleted because all fluid administered move into out of the vasculature.

The study in Chapter 6 found that a prophylactic noradrenaline infusion reduced the incidence of hypotension, lowered the incidence of inadequate planes of anaesthesia, and reduced intraoperative fluid volume administered compared to the conventional anaesthetic management, resulting in stable anaesthesia in dogs undergoing

ovariohysterectomy. Similar studies have been conducted in people. Novel methods to prevent hypo- and hypervolaemia investigated in previous chapters can be essential in a referral hospital, however, they may be bothersome in a small clinic which usually performs short and minor procedures such as spey and castration. Thus, in healthy dogs, a clinically adaptable simple method of prophylactic noradrenaline infusion may be suitable even for a small practice. The prophylactic noradrenaline infusion is able to get rid of the anxiety of most clinicians during surgery because hypotension is the most common complication of general anaesthesia without volume overload.

CONCLUSION

The result of the research presented in this thesis provided evidence that:

Chapter 2

PPV and PVI predicted fluid responsiveness in mechanically ventilated isoflurane-anaesthetised dogs after premedication with acepromazine.

Chapter 3

PWTT showed a good trending ability to be able to detect 15% changes in SV in isoflurane-anaesthetised dogs, but the agreement between estimated CO using PWTT and reference CO measured by the thermodilution technique was clinically unacceptable in isoflurane-anaesthetised dogs.

Chapter 4

-2.5% ΔPWTT following a 3 mL/kg of MFC could predict ≥ 15% ΔSV in mechanically ventilated anaesthetised dogs.

Chapter 5

-2.1% ΔPWTT induced by a 3 mL/kg of MFC could predict ≥ 15% ΔSV in spontaneously breathing anaesthetised dogs.

Chapter 6

Noradrenaline infusion prior to hypotension provided a lower incidence of hypotension, lower incidence of light plane of anaesthesia and less perioperative fluid volume administered than the conventional anaesthetic management, resulting in stable anaesthetic management in dogs undergoing ovariohysterectomy.

These techniques could be incorporated into veterinary clinical anaesthesia and reduce morbidity associated with anaesthesia.

FUTURE DIRECTIONS

There are many questions that arise from the results presented in this thesis. These questions include:

1. Can PPV and PVI be used for prediction of fluid responsiveness with less tightly controlled condition, for example, spontaneously breathing dogs?

   The study in **Chapter 2** showed the ability of PPV and PVI to predict fluid responsiveness in mechanically ventilated anaesthetized dogs in the experimental setting. However, dynamic indices are affected by not only the preload but also other factors such as heart rate (Morgan et al., 1966), respiratory rate (De Backer, Taccone, Holsten, Ibrahimi, & Vincent, 2009), pleural pressure (Liu et al., 2016), and tidal volume (Díaz et al., 2015; H. K. Kim & Pinsky, 2008). These confounding factors may decrease clinical application (Marik & Lemson, 2014). In fact, dynamic techniques are reported to be less reliable to predict fluid responsiveness in spontaneously breathing patients than in mechanically ventilated patients (Soubrier et al., 2007). However, these factors are already measured routinely or could be easily measured, and if their effects were quantified then their influence could be accounted for and included in any decision algorithm. Therefore, a further study should investigate whether PPV and PVI can predict fluid responsiveness in spontaneously breathing dogs, at various heart and respiratory rates.

2. Can a decrease in PPV and PVI following MFC predict fluid responsiveness?

   Studies in Chapter 2, 4 and 5 showed that PPV, PVI and MFC are reliable tools to predict fluid responsiveness in dogs. PPV at the steep portion of the Frank-Starling curve is greater than PPV at the plateau portion of the curve. Thus, a MFC would decrease PPV as preload move from the steep portion to the plateau portion.

In people, a decrease in PPV after a MFC accurately predicted fluid responsiveness in mechanically ventilated patients with a low tidal volume (Mallat et al., 2015). Therefore, the question is whether decrease in PPV and PVI following a MFC could predict fluid responsiveness in spontaneously breathing anaesthetised dogs.

3. Can MFC using PWTT be used for prediction of fluid responsiveness in awake or sedated dogs?

   For example, postoperative fluid management in a dog underwent splenectomy due to haemoabdomen, is always challenging. Because total blood loss and circulating volume required in the dog are often unknown. The study in **Chapter 5** revealed that, ΔPWTT following 3 mL/kg of a MFC could predict fluid responsiveness non-invasively in spontaneously breathing anesthetised dogs. Therefore, this technique may be able to guide a clinician to manage postoperative fluid management in this dog. This must be applicable for critically ill awake dogs in intensive care unit, where fluid administration is often required.

4. Does measurement of tissue oxygenation improve the fluid management after a MFC in dogs as a guide of fluid responsiveness?

   The ultimate purpose of fluid administration is to improve tissue oxygenation by increasing delivery of oxygenated blood. Appropriate fluid administration would increase tissue oxygenation, while excessive fluid administration would cause oedema and decrease tissue oxygenation or cause no change. However, technique to measure tissue oxygenation have proven unreliable in our hands in several pilot studies connected to this thesis and are still under investigation.

5. Can PPV, PVI and MFC using PWTT described in this thesis be used to predict fluid responsiveness in cats?

This thesis indicated that several techniques for prediction of fluid responsiveness in dogs are feasible although there are limitations for their use. Interestingly, prediction of fluid responsiveness in cats have not been investigated at the moment. A nested case-control study undertaken in 117 UK veterinary centres revealed the four-fold increase in odds associated with receiving fluid therapy compared to no fluid therapy (Brodbelt, Pfeiffer, Young, & Wood, 2007). the author in this study concluded that a component of the increased odds may be related to excessive administration of fluids and fluid overload. Cats are more susceptible to fluid overload because of their relatively small intravascular capacity (Robertson et al., 2018). Therefore, further studies for prediction of fluid responsiveness should be explored in cats.

6. How does a bolus of fluid affect endothelial glycocalyx layer?

   The endothelial glycocalyx layer, consists of a variety of endothelial membrane-bound molecules, including glycoproteins and proteoglycans, and covers the intraluminal surface of the vascular endothelium (Henry & Duling, 1999). The role of the endothelial glycocalyx layer is to maintain the normal fluid homeostasis across the vasculature. It is well known that destruction of the glycocalyx increased capillary permeability (Jacob et al., 2006), which potentially cause oedema. Although the mechanisms of glycocalyx shedding is unknown, it happened during septic shock (Nelson, Berkestedt, Schmidtchen, Ljunggren, & Bodelsson, 2008), after aortic surgery (Rehm et al., 2007) and following a bolus of fluid (Chappell et al., 2014; Guidet & Ait-Oufella, 2014) in people. Therefore, it is interesting whether excessive fluid administration could destroy the endothelial glycocalyx layer in dogs and cats. Furthermore, I would like to investigate whether techniques that we investigated in thesis could prevent the

destruction of the endothelial glycocalyx layer in dogs and cats, by optimising fluid administration.

7. Would PPV, PVI and MFC using PWTT described in this thesis improve morbidity and mortality in veterinary practice?

   It would be difficult to detect any improvement when those techniques are applied for healthy dogs because healthy cardiovascular system may be able to handle moderate volume overload. However, it is interesting to see whether those techniques can provide any benefits in haemodynamically unstable ill dogs.

REFERENCES

Alphonsus, C. S., & Rodseth, R. N. (2014). The endothelial glycocalyx: a review of the vascular barrier. *Anaesthesia, 69*(7), 777-784. doi:10.1111/anae.12661

Brodbelt, D. C., Pfeiffer, D. U., Young, L. E., & Wood, J. L. (2007). Risk factors for anaesthetic-related death in cats: results from the confidential enquiry into perioperative small animal fatalities (CEPSAF). *Br J Anaesth, 99*(5), 617-623. doi:10.1093/bja/aem229

Cannesson, M., Le Manach, Y., Hofer, C. K., Goarin, J. P., Lehot, J. J., Vallet, B., & Tavernier, B. (2011). Assessing the diagnostic accuracy of pulse pressure variations for the prediction of fluid responsiveness: a "gray zone" approach. *Anesthesiology, 115*(2), 231-241. doi:10.1097/ALN.0b013e318225b80a

Cavallaro, F., Sandroni, C., & Antonelli, M. (2008). Functional hemodynamic monitoring and dynamic indices of fluid responsiveness. *Minerva Anestesiol, 74*(4), 123-135.

Celeita-Rodríguez, N., Teixeira-Neto, F. J., Garofalo, N. A., Dalmagro, T. L., Girotto, C. H., Oliveira, G. C. V., & Santos, I. F. (2019). Comparison of the diagnostic accuracy of dynamic and static preload indexes to predict fluid responsiveness in mechanically ventilated, isoflurane anesthetized dogs. *Vet Anaesth Analg, 46*(3), 276-288. doi:10.1016/j.vaa.2018.12.004

Chappell, D., Bruegger, D., Potzel, J., Jacob, M., Brettner, F., Vogeser, M., . . . Rehm, M. (2014). Hypervolemia increases release of atrial natriuretic peptide and shedding of the endothelial glycocalyx. *Crit Care, 18*(5), 538. doi:10.1186/s13054-014-0538-5

De Backer, D., Taccone, F. S., Holsten, R., Ibrahimi, F., & Vincent, J. L. (2009). Influence of respiratory rate on stroke volume variation in mechanically ventilated patients. *Anesthesiology, 110*(5), 1092-1097. doi:10.1097/ALN.0b013e31819db2a1

De Backer, D., & Vincent, J. L. (2018). Should we measure the central venous pressure to guide fluid management? Ten answers to 10 questions. *Crit Care, 22*(1), 43. doi:10.1186/s13054-018-1959-3

Díaz, F., Erranz, B., Donoso, A., Salomon, T., & Cruces, P. (2015). Influence of tidal volume on pulse pressure variation and stroke volume variation during experimental intra-abdominal hypertension. *BMC Anesthesiol, 15*, 127. doi:10.1186/s12871-015-0105-x

Doherty, M., & Buggy, D. J. (2012). Intraoperative fluids: how much is too much? *Br J Anaesth, 109*(1), 69-79. doi:10.1093/bja/aes171

Drozdzynska, M. J., Chang, Y. M., Stanzani, G., & Pelligand, L. (2018). Evaluation of the dynamic predictors of fluid responsiveness in dogs receiving goal-directed fluid therapy. *Vet Anaesth Analg, 45*(1), 22-30. doi:10.1016/j.vaa.2017.06.001

Fantoni, D. T., Ida, K. K., Gimenes, A. M., Mantovani, M. M., Castro, J. R., Patrício, G. C. F., . . . Otsuki, D. A. (2017). Pulse pressure variation as a guide for volume expansion in dogs undergoing orthopedic surgery. *Vet Anaesth Analg, 44*(4), 710-718. doi:10.1016/j.vaa.2016.11.011

Guidet, B., & Ait-Oufella, H. (2014). Fluid resuscitation should respect the endothelial glycocalyx layer. *Crit Care, 18*(6), 707. doi:10.1186/s13054-014-0707-6

Henry, C. B., & Duling, B. R. (1999). Permeation of the luminal capillary glycocalyx is determined by hyaluronan. *Am J Physiol, 277*(2), H508-514. doi:10.1152/ajpheart.1999.277.2.H508

Jacob, M., Bruegger, D., Rehm, M., Welsch, U., Conzen, P., & Becker, B. F. (2006). Contrasting effects of colloid and crystalloid resuscitation fluids on cardiac vascular permeability. *Anesthesiology, 104*(6), 1223-1231.

doi:10.1097/00000542-200606000-00018

Kim, D. H., Shin, S., Kim, J. Y., Kim, S. H., Jo, M., & Choi, Y. S. (2018). Pulse pressure variation and pleth variability index as predictors of fluid responsiveness in patients undergoing spinal surgery in the prone position. *Ther Clin Risk Manag, 14*, 1175-1183. doi:10.2147/tcrm.S170395

Kim, H. K., & Pinsky, M. R. (2008). Effect of tidal volume, sampling duration, and cardiac contractility on pulse pressure and stroke volume variation during positive-pressure ventilation. *Crit Care Med, 36*(10), 2858-2862. doi:10.1097/CCM.0b013e3181865aea

Liu, Y., Wei, L. Q., Li, G. Q., Yu, X., Li, G. F., & Li, Y. M. (2016). Pulse Pressure Variation Adjusted by Respiratory Changes in Pleural Pressure, Rather Than by Tidal Volume, Reliably Predicts Fluid Responsiveness in Patients With Acute Respiratory Distress Syndrome. *Crit Care Med, 44*(2), 342-351. doi:10.1097/ccm.0000000000001371

Lopes, M. R., Oliveira, M. A., Pereira, V. O., Lemos, I. P., Auler, J. O., Jr., & Michard, F. (2007). Goal-directed fluid management based on pulse pressure variation monitoring during high-risk surgery: a pilot randomized controlled trial. *Crit Care, 11*(5), R100. doi:10.1186/cc6117

Loupec, T., Nanadoumgar, H., Frasca, D., Petitpas, F., Laksiri, L., Baudouin, D., . . . Mimoz, O. (2011). Pleth variability index predicts fluid responsiveness in critically ill patients. *Crit Care Med, 39*(2), 294-299. doi:10.1097/CCM.0b013e3181ffde1c

Mallat, J., Meddour, M., Durville, E., Lemyze, M., Pepy, F., Temime, J., . . . Tavernier, B. (2015). Decrease in pulse pressure and stroke volume variations after mini-fluid challenge accurately predicts fluid responsiveness†. *Br J Anaesth, 115*(3), 449-456. doi:10.1093/bja/aev222

Marik, P. E., & Lemson, J. (2014). Fluid responsiveness: an evolution of our understanding. *Br J Anaesth, 112*(4), 617-620. doi:10.1093/bja/aet590

Messina, A., Dell'Anna, A., Baggiani, M., Torrini, F., Maresca, G. M., Bennett, V., . . . Cecconi, M. (2019). Functional hemodynamic tests: a systematic review and a metanalysis on the reliability of the end-expiratory occlusion test and of the mini-fluid challenge in predicting fluid responsiveness. *Crit Care, 23*(1), 264. doi:10.1186/s13054-019-2545-z

Morgan, B. C., Abel, F. L., Mullins, G. L., & Guntheroth, W. G. (1966). Flow patterns in cavae, pulmonary artery, pulmonary vein, and aorta in intact dogs. *Am J Physiol, 210*(4), 903-909. doi:10.1152/ajplegacy.1966.210.4.903

Navarro, L. H., Bloomstone, J. A., Auler, J. O., Jr., Cannesson, M., Rocca, G. D., Gan, T. J., . . . Kramer, G. C. (2015). Perioperative fluid therapy: a statement from the international Fluid Optimization Group. *Perioper Med (Lond), 4*, 3. doi:10.1186/s13741-015-0014-z

Nelson, A., Berkestedt, I., Schmidtchen, A., Ljunggren, L., & Bodelsson, M. (2008). Increased levels of glycosaminoglycans during septic shock: relation to mortality and the antibacterial actions of plasma. *Shock, 30*(6), 623-627. doi:10.1097/SHK.0b013e3181777da3

Rehm, M., Bruegger, D., Christ, F., Conzen, P., Thiel, M., Jacob, M., . . . Becker, B. F. (2007). Shedding of the endothelial glycocalyx in patients undergoing major vascular surgery with global and regional ischemia. *Circulation, 116*(17), 1896-1906. doi:10.1161/circulationaha.106.684852

Robertson, S. A., Gogolski, S. M., Pascoe, P., Shafford, H. L., Sager, J., & Griffenhagen, G. M. (2018). AAFP Feline Anesthesia Guidelines. *J Feline Med Surg, 20*(7), 602-

634. doi:10.1177/1098612x18781391

Sano, H., Barker, K., Odom, T., Lewis, K., Giordano, P., Walsh, V., & Chambers, J. P. (2018). A survey of dog and cat anaesthesia in a sample of veterinary practices in New Zealand. *N Z Vet J, 66*(2), 85-92. doi:10.1080/00480169.2017.1413959

Sano, H., & Chambers, J. P. (2017). Ability of pulse wave transit time to detect changes in stroke volume and to estimate cardiac output compared to thermodilution technique in isoflurane-anaesthetised dogs. *Vet Anaesth Analg, 44*(5), 1057-1067. doi:10.1016/j.vaa.2016.11.014

Sano, H., Chambers, P., & Bridges, J. (2019). Effects of preemptive norepinephrine infusion prior to hypotension on anesthetic management in dogs undergoing ovariohysterectomy. *Veterinary Anaesthesia and Analgesia, 46*(6), 831.e814-831.e815. doi:10.1016/j.vaa.2019.08.039

Sano, H., Fujiyama, M., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2019). Investigation of percentage changes in pulse wave transit time induced by mini-fluid challenges to predict fluid responsiveness in ventilated dogs. *J Vet Emerg Crit Care (San Antonio), 29*(4), 391-398. doi:10.1111/vec.12860

Sano, H., Seo, J., Wightman, P., Cave, N. J., Gieseg, M. A., Johnson, C. B., & Chambers, P. (2018). Evaluation of pulse pressure variation and pleth variability index to predict fluid responsiveness in mechanically ventilated isoflurane-anesthetized dogs. *J Vet Emerg Crit Care (San Antonio), 28*(4), 301-309. doi:10.1111/vec.12728

Scolletta, S., Romano, S. M., Biagioli, B., Capannini, G., & Giomarelli, P. (2005). Pressure recording analytical method (PRAM) for measurement of cardiac output during various haemodynamic states. *Br J Anaesth, 95*(2), 159-165. doi:10.1093/bja/aei154

Soubrier, S., Saulnier, F., Hubert, H., Delour, P., Lenci, H., Onimus, T., . . . Durocher, A. (2007). Can dynamic indicators help the prediction of fluid responsiveness in spontaneously breathing critically ill patients? *Intensive Care Med, 33*(7), 1117-1124. doi:10.1007/s00134-007-0644-9

Zimmermann, M., Feibicke, T., Keyl, C., Prasser, C., Moritz, S., Graf, B. M., & Wiesenack, C. (2010). Accuracy of stroke volume variation compared with pleth variability index to predict fluid responsiveness in mechanically ventilated patients undergoing major surgery. *Eur J Anaesthesiol, 27*(6), 555-561. doi:10.1097/EJA.0b013e328335fbd1