Predicting the occurrence of major adverse cardiac events within 30 days after a patient's vascular surgery: An individual patient-data meta-analysis

By Thuvaraha Vanniyasingam

A Thesis Submitted to the School of Graduate Studies

in Partial Fulfillment of the Requirements

for the Degree: Master of Science

McMaster University

MASTER OF SCIENCE (2013) **McMaster University**

(Statistics) **Hamilton, Ontario**

TITLE: Predicting the occurrence of major adverse cardiac events within 30 days after a patient's vascular surgery

AUTHOR: Thuvaraha Vanniyasingam, B.Sc.

(McMaster University)

SUPERVISOR: Dr. Lehana Thabane

NUMBER OF PAGES: xiii, 111

Abstract

Background: Major adverse cardiac events, MACE – a composite endpoint of cardiac death and nonfatal myocardial infarction (MI) – are severe harmful outcomes that commonly arise after elective vascular surgeries. As current pre-operative risk prediction models are not as effective in predicting post-operative outcomes, this thesis will discuss the key results of an individual patient data meta-analysis that is based on data from six cohort studies of patients undergoing vascular surgery.

Objectives: The purpose of this thesis is to determine optimal thresholds of continuous covariates and create a prediction model for major adverse cardiac events (MACE), within 30 days after a vascular surgery. The goals include exploring the minimum p-value method to dichotomize cutpoints for continuous variables; employing logistic regression analysis to determine a prediction model for MACE; evaluating its validity against other samples; and assessing its sensitivity to clustering effects. The secondary objectives are to determine individual models for predicting all-cause mortality, cardiac death, and nonfatal MI within 30 days of a vascular surgery, using the final covariates assessed for MACE.

Methods: Both B-type naturietic peptide (BNP) and its N-terminal fragment (NTproBNP) are independently associated with cardiovascular complications after noncardiac surgeries, and particularly frequent after noncardiac vascular surgeries. In a previous study, these covariates were dichotomized using the receiver operating

characteristic (ROC) curve approach and a simple logistic regression (SLR) model was created for MACE [1]. The first part of this thesis applies the minimum p-value method to determine a threshold for each natriuretic peptide (NP), BNP and NTproBNP. SLR is then used to model the prediction of MACE within 30 days after a patient's vascular surgery. Comparisons were made with the ROC curve approach to determine the optimal thresholds and create a prediction model. The validity of this model was tested using bootstrap samples and its robustness was assessed using a mixed effects logistic regression (MELR) model and a generalized estimating equation (GEE). Finally, MELR was performed on each of the secondary outcomes.

Results: A variable, ROC_thrshld, was created to represent the cutpoints of Rodseth's ROC curve approach, which identified 116pg/mL and 277.5pg/mL to be the optimal thresholds for BNP and NTproBNP, respectively [1]. The minimum p-value method dichotomized these NP thresholds as BNP: 115.57pg/mL ($p<0.0001$) and NTproBNP: 241.7pg/mL ($p=0.0001$), and MINP_thrshld was the indicator variable of these cutpoints. No study provided data on both NP concentration levels. The prognostic factors of MACE were assessed in a series of SLR models, using odds ratios (OR) and corresponding 95% confidence intervals (CIs) and p-values (p). With MINP_thrshld having a slightly better association with MACE than ROC_thrshld, we proceeded with this indicator variable in our model formation. It was concluded that the final model for MACE contained variables MINP_thrshld (OR=8.5, 95% CI:(5.03, 14.41), $p<0.0001$), the type of surgery (OR=2.5, 95%CI:(1.40, 4.60), $p=0.0022$), and diabetes mellitus (OR=2.1, 95%CI:(1.15, 3.71), p=0. 0151). Our internal validation analysis proved this

model to be accurate, however using MELR and GEE, it was sensitive to methods that accounted for clustering effects. In particular, diabetes was not a statistically significant covariate in both the MELR and GEE models. A points system, ranging from 0 to 5, was also created to assist clinicians in determining individual patient risk. Lastly, we applied the same covariates in our final prediction model of MACE to our secondary outcomes, as they are all cardiovascular related events experienced by vascular surgery patients. MELR models were used to account for clustering effects and MINP_thrshld remained consistently significant ($p<0.05$) in all outcomes, while diabetes mellitus was removed from the models. The type of surgery was a statistically significant covariate for all-cause mortality and nonfatal MI.

Discussion: One key limitation to this thesis is the small sample size received for NTproBNP. Also, determining only one cutpoint for each NP concentration may not be sufficient, since dichotomizing continuous factors can lead to loss of information along with other issues. Further research should be performed to explore other possible cutpoints along with performing reclassification to observe improvements in risk stratification. After validating our final model against other samples, we can conclude that MINP_thrshld, the type of surgery, and diabetes are significant covariates for the prediction of MACE. With the simplicity in only requiring a blood test to measure NP concentration levels and easily learning the status of the other two factors, minimal effort is needed in calculating the points and risk estimates for each patient. Further research should also be performed on the secondary outcomes to examine other factors that may be useful in prediction.

Conclusions: The minimum p-value method produced similar results to the ROC curve method in dichotomizing the NP concentration levels. The cutpoints for BNP and NTproBNP were 115.57pg/mL and 241.7 pg/mL, respectively. Further research needs to be performed to determine the optimality of the final prediction model of MACE, with covariates MINP_thrshld, type of surgery, and diabetes mellitus.

Acknowledgement

I would like to express my most sincere thanks to my supervisor, Dr. Lehana Thabane, for not only providing me with the opportunity to work on this project, but for his patience, continuous guidance, inspiring encouragement, and wealth of knowledge. I could not be more grateful for his mentorship and his enthusiasm in my work during and after our meetings.

I am thankful for Reitze Rodseth for introducing me to this project and for offering his insights, clinical expertise and perspectives. A special thank you to Dr. Ben Bolker for patiently sitting by my side and assisting me through my coding dilemmas, offering advice with my work, and helping me build on my programming skills. Many, many, many thanks to Dr. Roman Viveros-Aguilera for being a great friend and continuous supporter since my undergraduate years. I am also appreciative of Dr. Macdonald and his teachings, willingness to share knowledge, and lovely conversations.

My experience as a Master's student at McMaster University was fulfilling, exciting, and very memorable. The faculty and staff who work in the Department of Mathematics and Statistics provided an enriching, motivating, and friendly learning environment for me to thrive in. I also thank Dr. Thabane's Biostatistics team of students for sharing their experiences, providing advice and comfort during stressful times, and for helping me see a fruitful picture that waits beyond my Master's program. I also extend my gratitude to Sayantee Jana, a fellow student and friend of mine, who stuck by me as we pressed on with our work.

Finally, I cannot thank my family and friends enough for their boundless love and interminable support during this entire process – I love you all dearly and keep you very close to my heart. I am extremely appreciative of your kind words, shared laughs, and continuous encouragement, especially during the stressful periods of this journey.

Table of Contents

Chapter 1

Introduction

1.1 Background

Cardiovascular mortality, myocardial infarction, and cardiac arrest are all major adverse outcomes to commonly arise after elective noncardiac surgeries [28]. With cardiovascular disorders being the chief cause of death in the industrialized world, as individuals experience cardiovascular complications, their risk of death is expected to increase [27]. From vascular surgeries alone, patients are more likely to experience issues such as perioperative mortality and adverse cardiovascular outcomes [29]. A clinician's decision-making process is dependent on his/her own knowledge of the risks of a surgery and current pre-operative cardiovascular management. From selecting the most appropriate type of anesthesia to how to monitor a patient during and after surgery, many critical decisions must be made. As current pre-operative risk stratification models are not as effective in predicting post-operative outcomes for vascular surgery patients, we need to improve upon our perioperative management to reduce such adverse events [10]. By better monitoring the pre-operative period of these patients, we can better determine post-operative risks of cardiac death or myocardial infarction (MI).

Details of the abbreviations and key variables used in this thesis are provided in Tables 1.1.1 and 1.1.2 (Appendix A), along with descriptions of each outcome assessed in the analyses in Table 1.1.3 (Appendix A).

1.1.1 Current Guidelines of Risk Assessment

Pre-operative risk estimation is essential in making decisions on testing and treatment methods [10]. Goldman and his team were the pioneers in developing a cardiac risk index for noncardiac surgeries in 1977 [9, 35]. Detsky applied modifications to this

in 1986 that lead to similar results in risk estimation [10]. Finally, Lee's Revised Cardiac Risk Index (RCRI) was developed and displayed significant improvements. It is now the leading clinical index for pre-operative cardiovascular risk stratification [31-34].

The primary risk factors of the RCRI include a high-risk type of surgery, diabetic insulin therapy, a pre-operative serum creatinine level greater than 177μmol/Litre, and a history of congestive heart failure, ischaemic heart disease, or cerebrovascular disease [31]. Despite its popularity as a population-derived index for classifying patients into risk categories, the RCRI is not only unable to determine individual patient risk, it is also not very accurate in stratifying patients undergoing vascular surgeries [8, 10]. In a prospective observational study of 10,081 vascular surgery patients, the RCRI underestimated their post-operative cardiovascular events [36]. Evaluation of other markers, beyond the RCRI, will improve pre-operative risk stratification models in predicting adverse postoperative outcomes of noncardiac vascular surgery.

1.1.2 Recent Approaches to Vascular Surgery Risk Assessment

Vascular surgeries include surgeries of the arteries and veins. Hormones, BNP and NTproBNP are independent prognostic markers of cardiovascular outcomes in vascular surgery [1, 40]. These natriuretic peptides (NP) are released into the blood by ventricular cardiomyocytes, due to the expansion of the atrial or ventricular wall [1, 41]. Stratification of patients into low and high risk categories can be performed from observing their pre-operative NP concentration levels. Not only will this assist the decision-making process of whether or not a vascular surgery is necessary for each individual patient, it will also allow for a better result in any pre-operative assessment

performed by physicians in determining the best type of perioperative management. Examination of these independent predictors will allow for more focus on the risks unique to each patient and will improve perioperative management to reduce such adverse events.

This study runs parallel to an individual patient data meta-analysis, "*The Predictive Ability of Pre-Operative B-Type Natriuretic Peptide in Vascular Patients for Major Adverse Cardiac Events*", a study conducted by Rodseth and colleagues [1]. Both BNP and NTproBNP were determined to be independently associated with cardiovascular complications after noncardiac surgeries [1]. Such events are particularly frequent among vascular surgery patients, given their high comorbidity burden. Rodseth and his team determined thresholds of the NP concentrations by applying the ROC curve approach. With this method, it was concluded that 116pg/mL is the optimal general cutpoint for BNP values with 66% sensitivity and 82% specificity. The NTproBNP threshold was found to be 277.5pg/mL. After applying these thresholds to reclassification, significant improvement was found in risk prediction. The type of vascular surgery, history of coronary artery disease, congestive cardiac failure, cerebrovascular disease, diabetes mellitus, and renal failure (creatinine >2mg/dl) are all identified, along with the NP thresholds, as covariates of cardiac complications [1, 31]. With BNP and NTproBNP now identified as significant predictors, questions still linger: Are these NP thresholds the most precise cutpoints? What model can we now use to replace the RCRI in predicting adverse outcomes after vascular surgery?

1.2 Objectives

This thesis is an individual patient data meta-analysis that is designed clinically to (i) determine optimal cutpoints for BNP and NTproBNP, (ii) determine a model to discriminate individuals into high and low risk groups of a MACE within 30 days of a vascular surgery, and (iii) produce a scoring system to assist physicians in classifying their patients into risk categories. The statistical objectives are to (i) explore the minimum p-value method to dichotomize BNP and NTproBNP and make comparisons with the ROC curve approach, (ii) employ logistic regression analysis on the prognostic factors for predicting MACE, and (iii) examine the validity and robustness of the prediction model. These statistical objectives are explained in a more detailed process in Table 1.2.1 (Appendix A)

The secondary objectives are to determine individual models for predicting the secondary outcomes – all-cause mortality, cardiac death, and nonfatal MI – within 30 days of a vascular surgery. Details are provided in Table 1.2.2 (Appendix A).

1.3 Scope of the Report

In the following chapters, I will discuss the statistical methods and results for dichotomizing pre-operative NP concentration levels and model formation, for MACE within 30 days after a vascular surgery. These chapters will lead to an analysis of the results and related issues, followed by some concluding statements.

In Chapter 2, I explain the strategies applied to search, select, and assess the quality of the studies used in this thesis. The primary and secondary outcomes along with their covariates can also be found in this section.

Details of the statistical methods are specified in Chapter 3 as they are used to tackle the objectives of this thesis. The minimum p-value method is applied to dichotomize BNP and NTproBNP; simple logistic regression (SLR) is used for modeling the prediction of each outcome; and insight into our validation, sensitivity analysis, and points system are provided.

The results of each stage of our analysis are presented in Chapter 4 with references to tables and figures, in the Appendices, to capture a more descriptive image of our assessments. Comparisons are made with the original thresholds and prediction model concluded from Rodseth and his colleagues' study [1], a final model is determined, multicollinearity is explored, and a points system is finalized.

Lastly, a discussion on the key findings lies in Chapter 5. Interpretations of our results, comparisons to similar studies, limitations and implications for clinical practice, and future research are included in this chapter. Concluding remarks are provided in Chapter 6.

Chapter 2

Methods of Literature Review

2.1 Sources of Literature Search and Search Strategy

This thesis uses data provided by Rodseth and his colleagues [1]. They conducted a literature search using electronic databases, MedLine (July 5, 2010) and Embase (week of June 21, 2010), with key words such as "natriuretic peptides", "surgery or surgical procedures", and various combinations of prognostic and diagnostic terms [2-4]. Only reports containing the largest of sample sizes and with the most thorough follow-ups were included to avoid the collection of overlapping data from various studies. Other exclusions included congress reports, study populations from cardiac surgery, and studies where administration of BNP was utilized for interventional purposes. After applying their exclusion criteria, studies were then selected based on pre-determined eligibility conditions [1].

2.2 Study Selection

As this study focuses on noncardiac vascular surgery patients, only 10 of the 1,648 citations found from Rodseth's electronic search satisfied their eligibility criteria [1]. After three attempts of contacting the investigators, individual patient data was obtained from six studies (N=850; n_{BNP}=632 and $n_{NTproBNP}$=218). Five provided information on pre-operative BNP concentration levels [16-20] and one on pre-operative NTproBNP concentration levels [21]. Data were also collected on age, gender, RCRI components and post-operative outcomes experienced within 30 days – from all studies. The RCRI components included were (i) type of noncardiac surgery performed, (ii) diabetes mellitus, (iii) a history of congestive heart failure, (iv) a history of cerebrovascular disease, (v) a history of coronary artery disease, and (vi) renal failure

(experienced creatinine levels ≥ 2mg/dl). Finally, a master dataset was created compiling patients from all studies.

Our final dataset contained 850 vascular patients of both open and catheter-based vascular surgery. Patient characteristics have been broken down for each outcome: (a) MACE, (b) all-cause mortality, (c) cardiac death, and (d) non-fatal MI and can be found in Tables 2.1.1-2.1.4 (Appendix B).

2.3 Study Quality Assessment

To evaluate the study quality of this meta-analysis, Rodseth modified the Quality Assessment of Diagnostic Accuracy Studies (QADAS) checklist [5] to account for the prognostic nature of our six studies. Adjustments included using "natriuretic peptide concentrations", "all-cause mortality", and "outcome" in replacement of "index test", "target condition", and "reference standard", respectively. Also, Criteria 3, 4, 7, and 13 were removed, as they did not apply to this study. Criterion 9 from the QADAS checklist was excluded for studies of in-hospital all-cause mortality. Rodseth also tested for reporting accuracy by randomly selecting 20% of the individuals and comparing them with their originally provided datasets [1].

2.4 Primary Outcome and Study Variables

The chief outcome of interest for this study is the occurrence of MACE within 30 days after a vascular surgery. The prognostic factors for this composite endpoint include:

- Age of patient (AGE, which is converted to AGE_thrshld)
- Brain-type natriuretic peptide (BNP)

- N-terminal pro-B-type natriuretic peptide (NTproBNP)
- Type of vascular surgery (SURGERY_TYPE)
- History of coronary artery disease (Hx_IHD)
- History of congestive cardiac failure (CCF)
- History of cerebrovascular disease (CEREBRO_VASCULAR_DISEASE)
- Diabetes mellitus (DIABETES)
- Renal failure with creatinine levels >2 mg/dl (HIGH_CREAT)

Details of these variables are found in Table 1.1.2 (Appendix A).

We apply the minimum p-value approach exclusively to continuous variables, BNP and NTproBNP, to determine their threshold values for MACE. From this, an indicator variable (MINP_thrshld) is created to identify whether an individual's pre-operative BNP or NTproBNP concentration levels are greater than the respective cutpoint values. Similarly, ROC_thrshld is created to represent cutpoints of BNP and NTproBNP identified by the ROC curve approach [49], performed by Rodseth [1]. These indicator variables are further explained in Section 3.3.

Using logistic regression analysis, Rodseth assessed the above listed covariates and identified ROC_thrshld, SURGERY_TYPE, and DIABETES as the statistically significant variables for MACE immediately after vascular surgery. This study examines these factors with MACE and considers the other covariates listed above. We also use the final predictors of MACE in our secondary analysis.

2.5 Secondary Outcomes and Study Variables

Secondary outcomes for this study include all-cause mortality, cardiac death, and nonfatal MI – each within 30 days after a patient's vascular surgery. The covariates identified in the final model for primary outcome, MACE, are the only study variables considered for these outcomes. Since all of the events are cardiovascular-related problems, we assumed that the significant factors in determining MACE would also be thresholds and key variables in predicting these secondary outcomes.

Chapter 3

Statistical Methods

3.1 Introduction

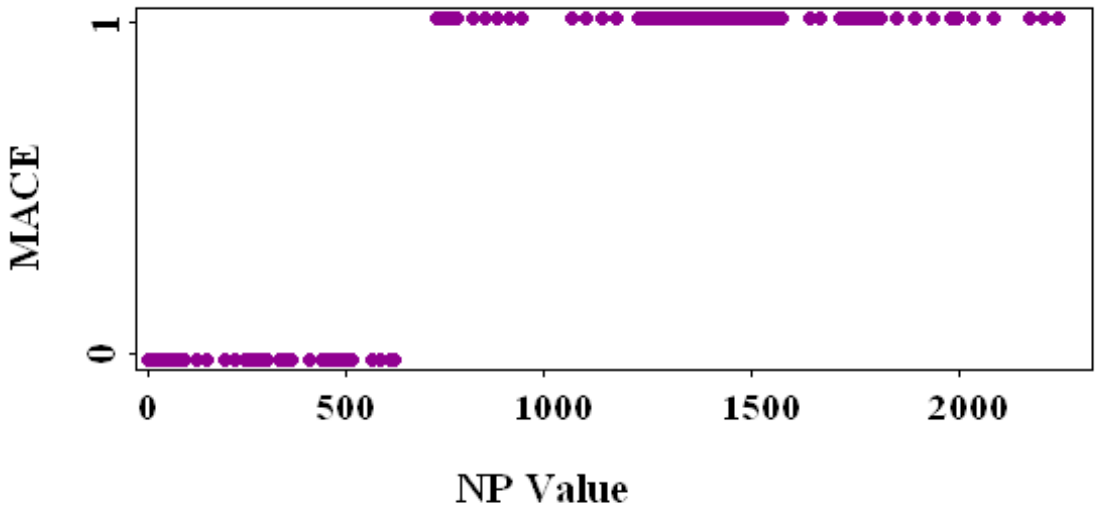
To bring more ease to their decision-making process, categorizing continuous variables assists clinicians in their understanding and interpretation of statistical results. This metamorphosis can be performed using the minimum p-value method -- a systematic approach in determining thresholds for a continuous variable. This chapter describes the initial graphical assessments performed, details of the minimum p-value method, and the features of the adjusted p-value formulae. Our logistic regression analysis, sensitivity analysis, and validation analysis are also provided to explain our approach in finalizing a prediction model for the outcome, MACE. To assist clinicians in interpreting our model, we created a point system and explain how to calculate risk estimates unique to each patient. Finally, our method of exploring our secondary outcomes of all-cause mortality, cardiac death, and non-fatal MI is described. A description of the different methods used in this thesis is provided in Table 3.1.1 (Appendix C) along with flowcharts in Figures 3.1.1-3.1.5 (Appendix C), to provide a more descriptive image of each step of our analysis.

For this thesis, statistical software RStudio 0.96.316 was used to determine these thresholds via the minimum p-value method. Logistic regression analysis, sensitivity analysis, and validation analysis were all performed in SAS 9.3.

3.2 Graphical Assessment

To begin examining the pre-operative concentration levels of BNP and NTproBNP, we used boxplots to observe the range of our data and to search for outliers. Scatter plots of MACE against each NP were also created to see if a step function would

appear in either graph. Since our outcome variable is binary, this step function would ideally display no occurrence of MACE (MACE=0) at low NP concentration levels and all occurrences of MACE (MACE=1) at high NP levels, as demonstrated below.



In this "perfect" scenario, possible noise would only appear around the optimal cutpoints of the graphs. However in our case, and in many real-world situations, such a step function does not exist. To accommodate for our data, a range of potential cut points were systematically evaluated to determine thresholds for BNP and NTproBNP.

3.3.1 The Minimum P-Value Method

For this thesis, we adopted the methodology and utilized the detailed R code provided by Glassman and Mazumdhar [6]. Aside from a portion of extreme values, the minimum p-value method assesses all of the observed data of the covariate as potential threshold values. Evaluation is based on the cutpoint holding the largest chi-squared statistic and corresponding p-value. We evaluate the predictive power of each potential cutpoint, c_i, for dependent, binary variable, MACE. This method is performed twice,

once for determining an optimal cutpoint for pre-operative BNP and once for pre-operative NTproBNP.

Suppose there exists p potential cutpoints in a set $c = \{c_1, c_2, \ldots, c_i, \ldots, c_p\}$, where $i=1, \ldots, p$. For each threshold, two bivariate groups are created:

(1) Patients with NP levels $\leq c_i$ and

(2) Patients with NP levels $> c_i$.

These groups and outcome MACE are evaluated using chi-squared tests to determine the predictive ability of each cutpoint. A table is created, computationally, to determine the test statistic and corresponding p-value of each c_i. The test statistic is evaluated based on a null hypothesis that the cutpoint under evaluation is not well defined -- there is no difference between the two groups. Contingency tables were created for MACE and each potential cutpoint, c_i, as seen below.

		Occurrence of a Major Adverse Cardiac Event	
Cutpoints		**NO MACE** *(k=0)*	**YES MACE** *(k=1)*
c_1=___	$X \leq c_1$ *(j=0)*	n_{00} (Observed number)	n_{01} (Observed number)
	$X > c_1$ *(j=1)*	n_{10}	n_{11}

The chi-square statistic, p-value, and relative risk measure for each potential cutpoint were calculated for both NPs. The cutpoint with the minimum p-value, or corresponding maximum chi-square value, was the best discriminator of patients into groups of high and

low risk. As the chi-squared statistic can be influenced by sample size, it is best to also examine the relative risk for each potential threshold [6].

3.3.2 The Corrected P-Value Approach

The main criticism of the minimum p-value method is the inflation that arises in the Type I error rate [6, 46]; this error occurs when a test is a "false positive". In our case, we are testing for whether or not an individual will experience MACE within 30 days after vascular surgery. Since our meta-analysis is comprised of follow-up studies, we know the outcomes of each patient within this timeframe. The individuals classified as high-risk and who do not experience MACE fall under the Type I error.

To adjust for such inflation and ensure the minimum p-value is significant, a few correction formulae were proposed. Mazumdhar and Glassman first identify Miller and Siegmund, who derived the first formula:

$$p_{ms} = \phi(z)\left(z - \frac{1}{z}\right) log\left(\frac{\varepsilon_{high}(1-\varepsilon_{low})}{(1-\varepsilon_{high})\varepsilon_{low}}\right) + 4\frac{\phi(z)}{z} \ , \qquad \textbf{(1)}$$

where p_{ms} is the adjusted p-value using the Miller and Siegmund formula [15], ϕ is the standard normal probability density function, z is equivalent to the $\left(1 - \frac{p_{min}}{2}\right)^{\text{th}}$ percentile of the standard normal distribution, and p_{min} is the determined minimum p-value. ε_{low} and ε_{high} are the proportions of observed values below the lowest cutpoint and at or below the highest cutpoint considered, respectively.

The second formula, by Altman et al. [23], is considered as a simplification formula. Here, ε is set as $\varepsilon = \varepsilon_{low} = \varepsilon_{high}$ = 5% or 10%. These formulas are useful for small minimum p-values that lie between 0.0001 and 0.1 [6]. The respective formulas are as follows:

For $\varepsilon_{low} = \varepsilon_{high} = 5\%$,

$$p_{alt5} = -3.13 p_{\min}(1 + 1.65 \ln(p_{\min})), \quad \textbf{(2)}$$

For $\varepsilon_{low} = \varepsilon_{high} = 10\%$,

$$p_{alt10} = -1.63 p_{\min}(1 + 2.35 \ln(p_{\min})). \quad \textbf{(3)}$$

The last correction formula we assess is the original Bonferroni correction formula. It multiplies the minimum p-value by the number of potential cutpoints. This formula, however, is not suitable for adjustment since the consecutive test statistics are not independent in the minimum p-value approach [6]. What is considered as appropriate is the revised version of the Bonferroni correction formula. Lausen and Schumaker [48] developed a tailored version of the minimum p-value, p_{modbon}, which considers the relationship between neighbouring cutpoints. Let ε_i be the proportion of observed values at or below the i[th] cutpoint, for p cutpoints, and the modified version is as follows:

$$p_{modbon} = p_{\min} + \sum_{i=1}^{k-1} D(\varepsilon_i, \varepsilon_{i+1}) \quad \textbf{(4)}$$

where $$D(\varepsilon_i, \varepsilon_{i+1}) = \frac{\exp\left(\frac{-z^2}{2}\right)}{\pi}\left[a(\varepsilon_i, \varepsilon_{i+1}) - \left(\frac{-z^2}{4} - 1\right)\left(\frac{a(\varepsilon_i, \varepsilon_{i+1})^3}{6}\right)\right]$$

and $$a(\varepsilon_i, \varepsilon_{i+1}) = \sqrt{\left\{1 - \frac{\varepsilon_i(1-\varepsilon_{i+1})}{(1-\varepsilon_i)\varepsilon_{i+1}}\right\}}.$$

3.4 MINP_thrshld and ROC_thrshld

Once the thresholds have been determined, MINP_thrshld is created to denote whether or not an individual's pre-operative BNP and NTproBNP concentration levels

surpass their respective cutoff values. In this meta-analysis, a total of six studies were used: five contained information on BNP while only one provided data on NTproBNP. Since no studies provided data on both pre-operative NP concentration levels, an indicator variable was created to represent the two cutpoints. MINP_thrshld is described as follows:

$$\text{MINP\_thrshld} = \begin{cases} 0, & \text{if BNP} < BNP\_thrshld \text{ or } NTproBNP < NTproBNP\_thrshld \\ 1, & \text{if BNP} \geq \text{BNP\_thrshld or NTproBNP} \geq \text{NTproBNP\_thrshld} \end{cases},$$

where BNP is a patient's actual BNP concentration level and BNP_thrshld is the BNP cutoff value determined by the minimum p-value method. Similarly, NTproBNP and NTproBNP_thrshld are defined.

The thresholds found using the minimum p-value method will then be compared to those from the ROC curve approach. Since both methods use the same dataset, MINP_thrshld will be compared with ROC_thrshld, the indicator variable for Rodseth's BNP and NTproBNP cutpoint values. The BNP and NTproBNP cutpoints determined by Rodseth are 116pg/mL and 277.5pg/mL, respectively. ROC_thrshld is defined as follows:

$$\text{ROC\_thrshld} = \begin{cases} 0, & \text{if BNP} < 116\text{pg/mL or NTproBNP} < 277.5pg/mL; \\ 1, & \text{if BNP} \geq 116\text{pg/mL or NTproBNP} \geq 277.5\text{pg/mL}. \end{cases}$$

Finally, the outcomes of interest, both primary (MACE) and secondary (all-cause mortality, cardiac death, and non-fatal MI) are all binary, response variables.

3.5 Logistic Regression Analysis

To address one of our statistical objectives of which method is better at dichotomizing continuous variables, the ROC curve method or the minimum p-value method, we performed simple logistic regression analysis. Rodseth has used SLR and identified ROC_thrshld, SURGERY_TYPE, and DIABETES as the significant predictors of his final model. The best way to make comparisons and draw conclusions is to mimic this model and create a similar one that replaces ROC_thrshld with MINP_thrshld. After this was performed, we moved on to determining a final prediction model for MACE within 30 days after a noncardiac vascular surgery.

From an epidemiological standpoint, we want to use logistic regression to determine the probability that a disease/outcome will occur within a set time frame for an individual, where values of the independent covariates of interest are already measured for each patient. From our literature review, we identified BNP and NTproBNP as independent predictors of MACE. The remaining prognostic factors, as listed in Section 2.4, are assumed independent for the purposes of our logistic regression analysis. Our set study period is 30 days after a vascular surgery, with details of our patients' characteristics taken prior to their surgery. Our outcome is MACE and the independent variables initially assessed are MINP_thrshld, SURGERY_TYPE, and DIABETES.

Statistically, the general form of an SLR model can be written as the probability, $\pi(\boldsymbol{X})$, of an event occurring ($E=1$) for an individual, given a set of k independent variables $X=(X_1, X_2, \dots, X_k)$:

$$\pi(\boldsymbol{X}) = P(\mathrm{E} = 1|X_1, X_2, \dots, X_k) = \frac{1}{1+e^{-(\beta_0 + \Sigma_{i=1}^{k} \beta_i X_i)}},$$

where β_0 and β_i are unknown parameters, and estimated based on our sample of patients and variables X_i, with i ranging from 1 to k, covariates. The parameters are estimated using maximum likelihood estimation. In this form, $\pi(\boldsymbol{X})$ provides the estimated probability or predicted risk of a patient experiencing MACE, given the independent variables.

The logit of the probability, $\pi(\boldsymbol{X})$, is a transformation of this form into a linear form:

$$logit\ \pi(\boldsymbol{X}) = \ln\left[\frac{\pi(\boldsymbol{X})}{1-\pi(\boldsymbol{X})}\right] = \beta_0 + \sum_{i=1}^{k}\beta_i X_i,$$

where $\frac{\pi(\boldsymbol{X})}{1-\pi(\boldsymbol{X})}$ is the odds for a specific individual and β_i is the change in the log odds when there is a unit change in X_i. We use PROC LOGISTIC in SAS 9.3 to create five logit models, explore the covariates, and determine a final prediction model of MACE. The ORs and corresponding 95%CIs and p-values of these SLR models (Models A1-A5), were assessed to make comparisons with Rodseth's results and to establish the final, parsimonious model of MACE. Model A1 used Rodseth's final covariates; Model A2 replaced Rodseth's ROC_thrshld with MINP_thrshld; and Model A3, A4, and A5 explored the inclusion of variables AGE_thrshld, HIGH_CREAT, and Hx_IHD.

3.6 Validation Analysis

To determine whether a prediction model and statistical inferences are subject to change with different samples, internal validation can be used. With several methods to measure a model's discriminative ability, calibration, and overall accuracy, a study was conducted to evaluate different methods of internal validation for logistic regression analysis. It was concluded that among those assessed, split-sample validation was the

least efficient approach while bootstrapping provided stable and nearly unbiased estimates for a predictive logistic regression model [14].

Bootstrap re-sampling, in simple terms, is sampling from sampled data. Sampling with replacement takes place on the original dataset to produce a certain number of samples, all possessing the same size as the original sample set. For our analysis, we created 1000 bootstrapped samples using Random-X bootstrapping in SAS 9.3. This created new samples with different dependent (*Y*) and independent variables (*X*) where we fit a new model, each with different error terms [37]. Our random-X bootstrapping method bootstraps the independent and dependent variables together. The ORs were calculated and recorded, along with the 95% CI and p-value for each covariate, in Table 4.6.3 (Appendix E). The average area under the ROC curve was also determined to assess the accuracy of the bootstrap models in dichotomizing patients into high and low risk groups. This can be found under Table 4.7.1 (Appendix E).

3.7 Sensitivity Analysis

As this thesis is a meta-analysis of six combined studies, our data has a multi-level structure: the study level and the patient level. Correlations, due to unobserved properties of studies, clinics, and patients, may exist at either of these levels. Our primary method of analysis uses simple logistic regression, which assumes homogeneity across patients and studies.

The purpose of this sensitivity analysis was to assess the robustness of our final model against methods that accounted for clustering effects. After determining the best-fit model, the final covariates were modeled against MACE using two different methods

to assess for sensitivity. An MELR model (Model B) and a GEE (Model C) were created.

A random effects or mixed effects logistic regression model was produced using the final predictors from our primary analysis. MELR considers clustering that could arise within- or between-studies and uses both fixed and random effects [30]. The fixed effects were the final covariates in our SLR model and STUDY is the random effect. It is assumed that the error term and random effect follow a normal distribution with a mean of zero and PROC GLIMMIX in SAS 9.3 is used to generate this model.

GEE adjusts for clustering within studies, however, it does not take into account correlations between studies [42]. PROC GENMOD is used in SAS to generate this model. In GEE, an exchangeable correlation matrix, which assumes the correlation between responses within a study is constant, is used to test for within study clustering. This correlation structure is assumed across studies. The models are compared based on their ORs, 95% CIs, and p-values.

3.8 Points System

When provided a statistical model, without background statistical knowledge, one may often feel perplexed by either the complexity of it or uncertainty of how to interpret it. To provide a more user-friendly system for both clinicians and patients to determine individual risks, a scoring system can be created. We use a points system described by Sullivan and his colleagues to break down our logistic regression model [7].

Our points system is based on the regression coefficients of our final model. After any continuous prognostic factors are transformed to binary or categorical variables,

a reference value, W_{ij}, is assigned to each level, *j*, of each variable, *i*. A base level (W_{iREF}) of 0 is assigned to the least unhealthy level of each covariate. The points can then be calculated for each j^{th} level of the i^{th} factor as

$$Points_{ij} = \frac{\widehat{\beta_i}(W_{ij} - W_{iREF})}{B}. \quad (5)$$

The constant, *B*, is set as the number of regression units that represent one point in this scoring system. Once a point is calculated for each level, different combinations are explored to determine all of the possible point totals for patient risk factor profiles. Finally, a risk estimate is calculated for each point total using the logistic regression formula, where we can approximate $\sum_{i=1}^{p} \beta_i X_i$ from the logistic regression formula with $B(Point\ Total)$ and p covariates. The approximation is shown below:

$$p = \frac{1}{1 + e^{-(\beta_0 + \sum_{i=1}^{p} \beta_i X_i)}}$$

$$\approx \frac{1}{1 + e^{-(\widehat{\beta_0} + \sum_{i=1}^{p} \widehat{\beta_i} W_{iREF} + B(Point\ Total))}}.$$

The details of these calculations are found in Tables 4.9.1-4.9.3 (Appendix E). Since all of our final covariates are binary, we set $W_{iREF}=0$ for all *i*. Thus the following formula will be used to determine our risk estimates for each point total:

$$p \approx \frac{1}{1 + e^{-(\widehat{\beta_0} + B(Point\ Total))}}. \quad (6)$$

3.9 Secondary Analysis

Our secondary analysis explores all-cause mortality, cardiac death, and non-fatal MI individually as they are all vascular surgery-related outcomes. Since this is a meta-analysis and clustering may arise among the data (and we are not making comparisons to other methods), we decided to create MELR models to predict each outcome within 30 days after vascular surgery. We assessed each model using a forest plot, ORs, and the corresponding 95% CI and p-values.

Chapter 4

Results

4.1 Key Demographics

Bivariate analyses using t-tests for continuous variables and Pearson chi-squared (P. Chi) tests for categorical variables were performed on the individual covariates and MACE with details provided in Table 2.1.1 (Appendix B). We identified age, RCRI class, coronary artery disease, congestive cardiac failure, diabetes, and renal insufficiency as individual significant predictors of MACE (α=0.05). We continued this assessment for each of the secondary outcomes.

For an all-cause mortality outcome, variables RCRI Class, congestive heart failure, diabetes mellitus, and creatinine were identified as significant factors (Table 2.1.2, Appendix B). While exploring relationships with cardiac death, the noted prognostic factors were AGE, RCRI Class, congestive heart failure, and renal insufficiency (Table 2.1.3, Appendix B). Lastly AGE, RCRI class, coronary artery disease, and congestive heart failure were identified as significant covariates for non-fatal MI (Table 2.1.4, Appendix B). It was no surprise that the RCRI class was significant for each outcome. In fact, this was expected, as it is currently the index used for risk stratification of vascular surgery patients.

4.2 Preliminary Assessment of BNP and NTproBNP

Preliminary assessment of the spread of the NP concentration levels is shown in the form of graphical displays in Figures 4.2.1 – 4.2.9 (Appendix D). The boxplots revealed 10.8% (68 out of 632) patients as outliers for patients with BNP≥161.25pg/mL, and 9.2% (20 out of 218) for patients with NTproBNP≥1522pg/mL. The individual scatter plots of the NP concentration levels reveal how close each concentration level is

to each other. An ideal situation would reveal a step function in Figure 4.2.7 (Appendix D), indicating no incidence of MACE for low levels of NP and all occurrences of MACE at high levels. In such circumstances where no step function or range of potential thresholds is revealed (Figures 4.2.8 and 4.2.9, Appendix D), a systematic approach of the minimum p-value can become quite useful.

4.3 Dichotomization of BNP and NTproBNP Using the Minimum P-Value Method

With the exception of 5% of outliers and values of 0pg/mL, to be set as extreme values, a total of 303 potential thresholds were considered for BNP (0pg/mL<BNP <2322.49pg/mL) and 204 for NTproBNP (21.5pg/mL<NTproBNP<1572pg/mL). Graphical displays of the chi-square statistics, p-values, and relative risks corresponding to each NP threshold value are shown in Figures 4.3.1-4.3.6 (Appendix D). From these graphs we can see a more distinct BNP cutpoint, with an obvious kink in Figure 4.3.2, than in NTproBNP (Figure 4.3.5).

Results of the minimum p-value method in dichotomizing the NP concentration levels are found in Table 4.3.7 (Appendix D). A BNP cutpoint of 115.57pg/mL possessed the smallest p-value (4.39x10^-21), and the largest chi-square value of 88.79, with a relative risk (RR) of 2.09. This RR is not the maximum of all of the potential cutpoints, however it does suggest that individuals with BNP levels beyond 115.57pg/mL are more likely to experience MACE than those with a lower value. The p-value adjustment formulae all indicate the BNP cutpoint of 115.57pg/mL is statistically significant ($p<0.05$).

For NTproBNP, the optimal threshold was determined as 241.7pg/mL. This held the smallest p-value of 0.001, possessed a maximum chi-square statistic of 10.98 from all of the assessed thresholds, and an associated RR of 3.33. It indicates that a patient with a pre-operative NTproBNP level beyond 241.7pg/mL, is over three times more likely to experience MACE within 30 days after vascular surgery than if it was below this cutpoint. With the exception of p_{alt510} at 10%, the remaining adjusted p-values show the determined NTproBNP threshold to be significant (α=0.05).

4.4 Comparisons of the Minimum P-value Method and the ROC Curve Approach

As we were unable to find studies that compared the minimum p-value method and ROC curve approach, we decided to make it one of our statistical goals to analyze the differences between the results of the two methods. To begin drawing comparisons, a few definitions are in order. Sensitivity is a measure of the precision of a diagnostic test in correctly detecting an outcome, while specificity measures how well individuals without an outcome are identified [22]. Accuracy, an overall measure of the ability of a diagnostic test in identifying individuals, is the ratio of the total number of correctly classified persons over all those assessed. The sensitivity and specificity for MINP_thrshld (64.0% and 81.5%, respectively) is fairly close to the ROC_thrshld (61.3% and 83.4%, respectively). The mere 8.8% (75 out of 850) of patients in the entire dataset who experience MACE explains these low true positive and high true negative rates. Since MINP_thrshld incorrectly classifies 1.7% more (14 out of 850) individuals as experiencing MACE than ROC_thrshld, there is only a minimal accuracy level difference (80% and 81.4%, respectively). The variation is due to the higher NTproBNP cutpoint obtained from the minimum p-value method.

Overall, the ROC curve method and minimum p-value method provide very similar results in threshold determination. Further comparisons of MINP_thrshld and ROC_thrshld and their associations with MACE are made in our logistic regression analysis.

4.5 Assessing Prognostic Factors and Studies for MACE

The individual patient data obtained from six studies were assessed by predetermined covariates [1, 10] for each outcome of MACE, all-cause mortality, cardiac death, and non-fatal MI, respectively reported in Tables 2.1.1-2.1.4 (Appendix B).

The third level of SURGERY_TYPE, the "not specified" category, was removed prior to regression analysis as it contained only 0.5% (4 out of 850) of the data. No patient found in this level experienced MACE and we felt that there was not enough information for this one level. Also, Table 4.5.1 (Appendix E) displays the breakdown of each study and outcome. A similar situation existed for individual patient data obtained from Study 5 [20], as it contained only three individuals. Two out of its three patients (67%) experienced MACE, a much higher percentage than any of the other studies. Removing Study 5 along with converting AGE into a binary variable (AGE_thrshld), with a cutpoint at 65 years, were in agreement with Rodseth's methods prior to his logistic regression analysis. Our exclusion of the third level in SURGERY_TYPE may cause slightly different results from his findings.

We continued this investigation with only the variables identified as statistically significant for MACE from Section 4.1. RCRI Class, however, was not considered for model formation as it uses its own RCRI Index to classify individuals into low, medium, and high-risk groups. Since discrimination using the RCRI Index is poorer for vascular

surgery patients than other types of noncardiac operations [10], only the statistically significant components of RCRI were assessed.

With BNP and NTproBNP having been identified as independent prognostic markers of cardiovascular outcomes in vascular surgery patients, literature has described a strong association between congestive cardiac failure (CCF) and these NP concentration levels [13, 41]. Strong correlations between covariates can cause inflation in the variances of the parameter estimates [26]. We assessed for collinearity between MINP_thrshld and CCF using the variation inflation factor (VIF) and tolerance level (TOL). Evaluation was based on a TOL level of less than 0.01 and VIF of greater than 10 to imply high multicollinearity. TOL is the degree to which an independent covariate will vary that is not explained by the other independent factors [24]. The reciprocal of its value is the VIF, which is a measure of how much multicollinearity between covariates has affected the inflation of the variance of each coefficient [26]. A regression model with explanatory variables, CCF and MINP_thrshld, was created with outcome MACE. The VIF and TOL between the two factors were 0.98 and 1.02, respectively. Despite this low association between the two covariates for this dataset, we decided not to include CCF in the prediction model to agree with the literature found.

4.6 Determining a Prediction Model for MACE

For more of a "birds-eye view" of the logistic regression analysis performed in this thesis, a breakdown is provided in Table 4.6.1 (Appendix E) to better envision the processes that took place and their associated variables.

A total of five SLR models (Models A1-A5) were first created to determine the most parsimonious model, with details provided in Table 4.6.2 (Appendix E). We used the Akaike information criteria (AIC) and the area under the ROC curve (AUC) as the goodness-of-fit statistics; where the lower the AIC and the higher the AUC, the better a model fits our data. The ORs and corresponding p-values were also used to assess the association of each covariate with MACE. We begin by comparing Model A1 and A2 to draw comparisons between ROC_thrshld and MINP_thrshld.

At a first glance at Table 4.6.2 (Appendix E), we can see that there are no major differences between Model A1 and A2. A closer look shows MINP_thrshld as not only slightly improving the model fit, but also strengthening the association between the other prognostic factors and MACE. The AIC is lower, AUC is higher, and the ORs of each covariate are higher in Model A2 than in Model A1. With this slight improvement, we decided to continue our SLR analysis with MINP_thrshld instead of ROC_thrshld. We explore the inclusion of AGE_thrshld, HIGH_CREAT, and Hx_IHD individually in Models A3, A4, and A5. From assessing the p-values in each of these models, we see that none of these additional covariates are statistically significant (α=0.05). We can now conclude that Model A2 is our final model with prognostic factors MINP_thrshld, SURGERY_TYPE, and DIABETES.

4.7 Results of Internal Validation Analysis with Bootstrapping

Once our final model was determined, internal validation was performed to assess its accuracy in discriminating patients into high and low risk groups of MACE, within 30 days after a noncardiac vascular surgery. To begin, we implemented a bootstrapping method to generate a total of 1000 samples, each with a size of 843 individuals (Study 5 and SURGERY_TYPE=2 continued to be excluded). We then performed SLR on each of the 1000 samples, using covariates from Model A2, and recorded the average of the results under Model F. The average of the ORs for each covariate were recorded in Table 4.6.3 (Appendix E), under Internal Validation Analysis, along with 95%CI and p-values. From here, the bootstrapped ORs, 95% CIs, and p-values are very close to those of the original model, indicating the regressors are statistically significant and the model is accurate against other samples. The average of the goodness-of-fit statistic, AUC, was also calculated (Table 4.8.1, Appendix E) and also very similar to the AUC of Model A2. Overall, Model F produces close results to Model A2.

4.8 Results of Sensitivity Analysis

As in any meta-analysis, clustering effects can always arise and affect one's analysis. To assess the robustness of Model A2 against such effects, we made comparisons between two different cluster-specific methods, MELR (Model B) and GEE (Model C). The details are presented in the form of ORs, 95% CIs, and p-values in Table 4.6.3 (Appendix E).

ORs were used to determine the effectiveness and association of each predictor and MACE in each model. The high OR for MINP_thrshld agrees with the literature [1, 16-19] that BNP and NTproBNP are important biomarkers of MACE. The 95% CIs for

Models B and C are larger than for Model A2 as they both take into account clustering effects. Overall, there is some variation in the ORs, however the association of each effect to MACE is relatively close. The p-values have indicated that the significance of DIABETES may be subject to clustering effects, with a p-value>0.05 for both Model B and C. Figure 4.6.4 (Appendix E) displays a forest plot corresponding to the results in Table 4.6.3(Appendix E). This plot illustrates the variation in ORs between the models by each covariate. From here, it can be seen that the confidence intervals of Models B and C for DIABETES intersect the vertical line, the line of no effect, also implying that this factor is not significant (α=0.05). The MELR and GEE methods identify the existence of clustering effects and the influence on DIABETES in Model A2. Since our final model, an SLR model, assumes homogeneity across both the patient and the study level, it identified DIABETES as a significant factor.

An ROC curve was created for Model B and the AUC of 0.776 is recorded in Table 4.7.1 (Appendix E). Despite clustering effects and DIABETES not being significant, there is minimal impact on the fit of the model with an AUC for Model A2 as 0.777.

4.9 The Point System

A scoring system was developed using the estimates from Model A2 for risk factors MINP_thrshld, DIABETES and SURGERY_TYPE. The final SLR model possessed baseline levels of (i) less than the NP (BNP<115.57pg/mL or NTproBNP<241.7pg/mL) thresholds, (ii) infrainguinal vascular surgery, and (iii) no diabetes. Details of the formation of the point system are presented in Tables 4.9.1-4.9.3

(Appendix E). Constant "B" was set as the lowest regression coefficient (0.7262) and represents the number of regression units relating to a single point [7]. As the different point totals were gathered, ranging from 0 to 5, their associated risks were calculated using (6) and recorded in Table 4.9.3 (Appendix E).

4.10 Results of Secondary Analysis

As previously mentioned, the data used for this thesis is based on patients who have undergone vascular surgery. As all-cause mortality, cardiac death, and non-fatal MI are all cardiovascular related outcomes; the final covariates in Model A2 were used to produce prediction models for each. Our sensitivity analysis exposed clustering effects among our data and so MELR was used to perform this secondary analysis. DIABETES was removed, as it was not a significant variable (α=0.05) in any of the three models.

Details of the final models in the form of AUC, ORs, 95% CIs and p-values are displayed in Table 4.10.1 (Appendix F), with a forest plot of these ORs and 95% CIs in Figure 4.10.1 (Appendix F). The high ORs and low p-values of MINP_thrshld indicate that it is also a significant covariate for these three outcomes. The range in ORs of SURGERY_TYPE is consistent with the OR from Models A2 (with for MACE). It is not, however, statistically significant for a cardiac death outcome. The AUCs of each model (Models G-I) are greater than 70%, demonstrating a moderate level of goodness-of-fit.

Chapter 5

Discussions

5.1 Summary of Key Findings

The purpose of this individual patient data meta-analysis was to determine the optimal NP thresholds and create a parsimonious statistical model for the prediction of MACE, within 30 days after a vascular surgery. The minimum p-value method identified a BNP cutpoint of 115.57pg/mL and an NTproBNP cutpoint of 241.7pg/mL. In comparison to Rodseth's ROC curve approach, this was a minimal difference of <0.5pg/mL between BNP thresholds and was a slightly lower NTproBNP cutpoint. Overall, there were no major differences in results between the two methods for dichotomizing continuous variables. The NP cutpoints were very similar and possessed close sensitivity, specificity, and accuracy levels.

SLR was set as the primary type of logistic regression analysis. It was consistent with Rodseth's methods and allowed for easy comparisons in threshold effects between the minimum p-value method and the ROC curve approach. Assessment of each model (Models A1-A5) was conducted using ORs, 95% CIs, p-values to measure the strength of association between each covariate and MACE. AUC and AIC statistics were used to determine the goodness-of-fit of each model. It was concluded that MINP_thrshld fit the SLR models slightly better than ROC_thrshld. With such close measures, it was difficult to select one method as more optimal than the other. With this being said, we continued with our final prediction model of MACE as Model A2, which contained prognostic factors MINP_thrshld (OR: 8.5, 95% CI: (5.03, 14.41)), SURGERY_TYPE (OR:2.6, 95% CI: (1.40, 1.70)) and DIABETES (OR: 2.5, 95%CI: (1.15, 3.71). This model was then validated using bootstrap samples.

After assessing Models F and A2, the ORs and p-values indicated that MINP_thrshld was strongly associated with the outcome, MACE, and confirmed

SURGERY_TYPE and DIABETES were key covariates. Based on their AUCs, they were also able to produce a very close goodness-of-fit level.

As our data were subject to clustering effects from studies and patients in this individual patient data meta-analysis, we explored Model A2's sensitivity using MELR and GEE. Slight variations in estimates and ORs were expected between the models since Models B and C accounted for clustering effects, while Model A2 did not. Sensitivity analysis of these models suggested that clustering effects did exist among the data and affected the influence of DIABETES in our final model. In other words, since Model A2 assumes homogeneity within and across studies, DIABETES appeared as statistically significant (α=0.05), while our MELR and GEE models suggested otherwise. However, upon comparison of AUC measures between the MELR and SLR models, there was minimal difference (0.1%) in their goodness of fit.

The prognostic factors finalized in Model A2 were applied to each secondary outcome, as they are all cardiovascular-related events. Table 4.10.2 (Appendix F) displays a combined 2x2 contingency table for each secondary outcome and MINP_thrshld while Table 4.10.3 (Appendix F) provides details of the sensitivity, specificity, and accuracy measures. These tables suggest that MINP_thrshld does not appear to be an accurate discriminant of all-cause mortality, cardiac death, and non-fatal MI. The sensitivity and specificity for each outcome indicate that the NP thresholds, which make up indicator variable MINP_thrshld, are too low.

Overall, the significance level of MINP_thrshld on each outcome is very high ($p<0.0001$). MELR was used for our secondary analysis to account for the clustering effects in our data. DIABETES was not significant in any of the outcomes, including MACE. The ORs and p-values in Table 4.10.1 (Appendix F) suggest SURGERY_TYPE

as having a similar effect on the secondary outcomes as it did on MACE, except for cardiac death where it is not statistically significant. Further research needs to be conducted, alongside the inclusion of other clinically and statistically significant prognostic factors, to determine the best-fit model for each secondary outcome.

5.2 Assessing the Impact of Study Quality

A meta-analysis is the synthesis of results from different studies to explore patterns or relationships that may arise in a larger, combined study than in a smaller individual one. The more diverse a merged dataset is, the more difficult it becomes to make comparisons and draw valid conclusions. To avoid this issue, we assessed the study quality by examining the heterogeneity among the six studies. In a meta-analysis with individual patient-data from different centers, a multi-level structure is formed where correlation can arise among observations within a center and between centers. Mixed effects logistic regression was performed during our sensitivity analysis with STUDY as the random effect, and MINP_thrshld, SURGERY_TYPE, and DIABETES as the fixed effects. We can assess the effect size of the STUDY variable to determine the heterogeneity that exists in our combined dataset. With an estimate of 0.3749, we found that some heterogeneity does exist among the data, which explains the variation in the significance of the DIABETES variable between models.

Another issue that may arise in model formation is multicollinearity. This occurs when there exists a strong, linear relationship among covariates [26]. High correlations between explanatory variables can result in unstable estimates, larger standard errors, and misleading results in determining which variables are statistically significant. We

assessed possible multicollinearity among the variables in Models A1-A5 using PROC REG in SAS 9.3. Since multicollinearity arises among prognostic factors and does not include the response variable, PROC REG in SAS 9.3 can still be used [26]. We set a VIF measure of greater than 10 and TOL level of less than 0.01, for each variable, as the indicators of high multicollinearity [26]. In this study, the TOL for each explanatory variable in Models A1-A5 was greater than or equal to 0.9 while the VIFs were found to be less than 1.2, suggesting that multicollinearity among the final covariates was low.

5.3 Comparison of Findings with Similar Works

With 200 million noncardiac surgeries occurring annually all over the world, pre-operative BNP and NTproBNP have been identified as having clinical significance in risk stratification of patients [39]. Considerable research has been performed on the relationship between elevated pre-operative BNP and NTproBNP concentration levels and post-operative MACE [1, 12, 40]. The main hurdle in using these NPs as prognostic factors is in defining thresholds that classify patients into low and high-risk categories of post-operative MACE. What is considered a cutpoint for one type of surgical procedure may not be the same for another.

Cardiac troponin has been found to determine post-operative predictions of mortality, both short- and long-term after vascular surgery [44, 47]. However, to improve perioperative management, pre-operative assessment is very useful. A meta-analysis was performed to determine which of six different pre-operative tests was the most accurate in predicting a post-operative MACE, within 30 days for vascular surgery patients. Ambulatory electrocardiography (ECG), exercise radionuclide ventriculography,

myocardial perfusion scintigraphy, dipyridamole stress echocardiography and dobutamine stress echocardiography (DSE), were all evaluated and it was concluded that DSE provided more precise predictions [45]. Another study suggested pre-operative BNP levels was just as, if not more, predictive as dobutamine stress echocardiography [12]. The advantages of using BNP and NTproBNP levels are the cost-effectiveness, time-efficiency, and ease in measuring their concentration levels from a simple blood test.

The minimum p-value method is a common systematic method in determining cutpoints of continuous, prognostic factors. The primary disadvantage of this approach lays in the issue of the type I error rate [46], which arises as a result of multiple testing. The 'optimal' cutpoint, which is based on the lowest p-value among the assessed potential thresholds, may lead to what appears to be a highly relevant prognostic dichotomous variable. This conclusion, however, may be misguided due to the inflation of the type I error rate. Our research implements the three correction formulae to assist in determining the accuracy of our BNP and NTproBNP cutpoints. Other methods that utilize this maximum chi-square method to determine thresholds are the two-fold cross-validation approach [44] and the split sample approach [43].

The minimum p-value method was applied to a study with an outcome of breast cancer and continuous predictor variable, sun protection factor (commonly known as SPF). It demonstrated that cutpoints may vary based on the assay and one particular threshold cannot be considered "optimal". This meta-analysis evaluates the accuracy of the cutpoints using the internal validation via bootstrapping. The predictive ability of Model F was very similar to Model A2.

5.4 Key Limitations of the Study and Further Research

As BNP and NTproBNP have been found to be significant biomarkers in predicting MACE for vascular surgery patients, one study has found that NTproBNP is possibly a better indicator than BNP for outcomes such as mortality, morbidity, hospitalization due to heart failure, left ventricular abnormality or impairment, and coronary artery disease. Its more stable composition suggests it to possibly be less sensitive to sudden haemodynamic shifts [11, 12]. With only one study (n=218) containing pre-operative NTproBNP concentration levels and only 19 (8.72%) of these individuals who experience MACE, the thresholds determined for NTproBNP could be misleading. Only 75 of 850 (8.82%) of individuals in total experienced MACE. The small sample size and low number of individuals with an outcome of MACE limit the results of this analysis. Further research needs to be performed on NTproBNP with more outcomes of MACE to ensure the robustness of the determined threshold value.

Another limitation of this meta-analysis is that it contained information on either BNP or NTproBNP. There were no studies that provided data on both NP concentration levels. Exploration in studies containing information on both pre-operative NP concentration levels will be useful in determining which is a better predictor (BNP or NTproBNP) or if both, together, will improve model accuracy.

Thirdly, a study has identified the level of renal dysfunction (HIGH_CREAT) as being highly correlated with NTproBNP concentration levels -- the more severe a patient's renal function is, the lower the specificity is of NTproBNP for adverse cardiac outcomes [12]. This thesis did not explore the effects of HIGH_CREAT and MINP_thrshld together nor did it assess the interaction of the two variables in the model. Further research could also be performed to determine how large of an impact the

association between HIGH_CREAT and NTproBNP has on the final prediction outcome of MACE.

Also, as in many analyses the issue of measurement error may arise. Since this is a meta-analysis, different centers may have measured and recorded BNP and NTproBNP concentration levels differently than others.

5.5 Implications for Clinical Practice

Appropriate perioperative preventative measures can be taken with the ability to determine immediate post-surgical adverse events. Pre-operative risk stratification of patients for MACE has many advantages. Firstly, both patients and physicians are more aware of the risks prior to surgery. They can make better-informed decisions based on the urgency or necessity of the operation, the benefits versus the risks to be taken, and what type of post-operative care is needed. With risk stratification, doctors can focus more attention on the high-risk patients and decide if alternative methods or supplementary interventions will improve the post-operative health of these individuals.

Another key advantage of our final model is that it only incorporates BNP or NTproBNP, the type of vascular surgery to be undergone, and whether or not a patient has diabetes mellitus. The only measurements to be taken are for NP concentration levels, which can be obtained from a simple blood test. Alongside this, the type of vascular surgery to be undergone is already known and a clinician can easily determine the diabetes status of a patient. With patient information that is so easily obtained, without any major machines to be used, this model is at an advantage. The affordability and accessibility of tools, paired with the simplicity of determining an individual patient's point total, allows for an easy assessment of pre-operative risk using the scoring system

developed in this thesis. Its systematic ease will help improve clinical practice in pre-operative risk stratification of post-operative MACE.

Based on our statistical findings, our final model is not robust since the significance of DIABETES is sensitive to clustering effects. Since this is a meta-analysis, we need to explore a larger dataset with more information on NTproBNP and also discuss with clinicians about the importance of DIABETES in the prediction model. The area under the ROC curve does imply that Model A2 is a good fit. Until a larger dataset is obtained, our final model can be explored to see if it improves risk stratification of patients. Although it cannot be concluded that this is the optimal prediction model of MACE, it is quite beneficial in its ease of classifying patients.

Chapter 6

Conclusions

6.1 Conclusions

As previously mentioned, several studies have identified the association of elevated levels of pre-operative BNP and NTproBNP concentration levels with major adverse cardiac events, such as non-fatal MI and cardiac death. These NPs were examined and their cutpoints were determined for MACE within 30 days after vascular surgery. We assessed the prognostic factors in prediction models for this outcome and secondary outcomes all-cause mortality, cardiac death, and non-fatal MI within 30 days after vascular surgery. Statistically, we explored the minimum p-value method and a variety of modeling approaches including SLR, MELR, and GEE.

6.1.1 Statistical Conclusions

The systematic procedure of the minimum p-value method highlighted a BNP cutpoint of 115.57pg/mL and 241.7pg/mL for NTproBNP. The SLR model assumed homogeneity between and within studies for this meta-analysis. The fit statistics exhibited the final model as a good fit with MINP_thrshld, the type of surgery, and diabetes mellitus as the statistically significant prognostic factors of MACE.

By performing internal validation using 1000 bootstrap re-samples, the average ORs of the SLR models created were similar to that of the final model. Comparisons of the ORs, p-values, and AUC for our final model were also made with MELR and GEE models. It was found that DIABETES was not a statistically significant covariate when clustering effects were accounted for. The MELR model did, however, produce similar

AUC measures as our final SLR model. We concluded Model A2, had a modest fit of the data and was sensitive to methods that accounted for clustering effects.

6.1.2 Clinical Conclusions

Our first clinical goal was to determine optimal cutpoints for BNP and NTproBNP. We were successfully able to dichotomize BNP and NTproBNP using the minimum p-value approach. Since our results were similar to a previous ROC curve method that had already been performed on our dataset, it was difficult to determine which cutpoints were the most optimal. Further research using a larger dataset with more NTproBNP data will assist in determining the most optimal cutpoints. The ORs, p-values, and fit statistics identify MINP_thrshld, SURGERY_TYPE, and DIABETES as the key covariates in predicting MACE within 30 days after vascular surgery. Using these variables a point system was created, ranging from 0 (2.5% risk) to 5 (49% risk). This scoring system provides ease for clinicians and patients to determine the risks of undergoing their surgery and improves their decision-making process and perioperative management.

References

[1] Rodseth RN, Buse GA, Bolliger D et al. The Predictive Ability of Pre-Operative B-Type Natriuretic Peptide in Vascular Patients for Major Adverse Cardiac Events, An Individual Patient Data Meta-Analysis. *Journal of the American College of Cardiology* 2011; *58* (5), 522:529.

[2] Haynes RB, Wilczynski NL. Optimal search strategies for retrieving scientifically strong studies of diagnosis from Medline: analytical survey. *BMJ* 2004;1:5.

[3] Wilczynski NL, Haynes RB. EMBASE search strategies for identifying methodologically sound diagnostic studies for use by clinicians and researchers. *BMC Medicine - BioMed Central* 2005; *3* (7), 1:6.

[4] Wilczynski NL, Haynes RB, Lavis JN et al. Optimal search strategies for detecting health services research studies in MEDLINE. *Canadian Medical Association Journal 2004*;*171* (10), 1179:1185.

[5] Whiting P, Rutjes AW, Reitsma JB et al. The development of QUADAS: a tool for the quality assessment of studies of diagnostic accuracy included in systematic reviews. *BMC Medical Research Methodology – BioMed Central* 2003; *3* (25), 1:13.

[6] Glassman JR, Mazumdar M. Tutorial in Biostatistics - Categorizing a Prognostic Variable: Review of Methods, Code for Easy Implementation and Applications to Decision-Making about Cancer Treatments. *Statistics in Medicine* 2000; *19* (1), 113:132.

[7] Sullivan LM, Massaro JM, D'Agostino Sr. RB. Tutorial in Biostatistics, Presentation of multivariate data for clinical use: The Framingham Study risk score functions. *Statistics in Medicine* 2004; *23*, 1631:1660.

[8] Ford KM, Beattie WS, Wijeysundera DN. Systematic Review: Prediction of Perioperative Cardiac Complications and Mortality by the Revised Cardiac Risk Index. *Annals of Internal Medicine* 2010; *152* (1), 26:35.

[9] Prateek GK, Himani G, Abhishek S et al. Development and Validation of a Risk Calculator for Prediction of Cardiac Risk After Surgery. *Circulation* 2011; 124, 381:387.

[10] Biccard BM, Rodseth RN. Utility of clinical risk predictors for preoperative cardiovascular risk prediction. *British Journal of Anaesthesia* 2011; *107* (2), 133:143.

[11] Rodseth RN. B type natriuretic peptide - a diagnostic breakthrough in peri-operative cardiac risk assessment? *Journal of the Association of Anaesthetists of Great Britain and Ireland* 2008; *64* (2), 165:178.

[12] Rodseth RN, Padayachee L, Biccard BM. A meta-analysis of the utility of pre-operative brain natriuretic peptide in predicting early and intermediate-term mortality and major adverse cardiac events in vascular surgical patients. *Journal of the Association of Anaesthetists of Great Britain and Ireland* 2008; *63* (11), 1226:1233.

[13] Maisel A. B-Type Natriuretic Peptide Levels: Diagnostic and Prognostic in Congestive Heart Failure: What's Next? *Circulation* 2002; *105* (20), 2328:2331.

[14] Steyerberg EW, Harrell Jr. FE, Borsboom GJ et al. Internal validation of predictive models: Efficiency of some procedures for logistic regression analysis. *Journal of Clincial Epidemiology* 2001; *54* (8), 774:781.

[15] Miller R, Siegmund D. Maximally Selected Chi Square Statistics. *International Biometric Society* 1982; *38* (4), 1011:1016.

[16] Bolliger D, Seeberger M, Lurati Buse GA et al. A Preliminary Report on the Prognostic Significance of Preoperative Brain Natriuretic Peptide and Postoperative Cardiac Troponin in Patients Undergoing Major Vascular Surgery. *Anesthesia Analgesia* 2009; *108* (4), 1069:1075.

[17] Biccard BM, Naidoo P. The role of brain natriuretic peptide in prognostication and reclassification of risk in patients undergoing vascular surgery. *Journal of the Association of Anaesthetists of Great Britain and Ireland* 2011; *66* (5), 379:385.

[18] Cuthbertson BH, Amiri AR, Croa BL et al. Utility of B-type natriuretic peptide in predicting perioperative cardiac events in patients undergoing major non-cardiac surgery. *British Journal of Anaesthesia* 2007; *99* (2),170:176.

[19] Gibson SC, Payne CJ, Byrne DS et al. B-type natriuretic peptide predicts cardiac morbidity and mortality after major surgery. *British Journal of Surgery* 2007; *94* (7), 903:909.

[20] Leibowitz D, Planer D, Rott D et al. Brain natriuretic peptide levels predict perioperative events in cardiac patients undergoing noncardiac surgery: a prospective study. *Cardiology* 2008; *110* (4), 266:270.

[21] Mahla E, Baumann A, Rehak P et al. N-terminal pro-brain natriuretic peptide identifies patients at high risk for adverse cardiac outcome after vascular surgery. *Journal of the Association of Anaesthetists of Great Britain and Ireland* 2007; *106* (6), 1088:1095.

[22] Zhu W, Zeng N, Wang N. Sensitivity, specificity, accuracy, associated confidence interval and ROC analysis with practical SAS® implementations. *NorthEast SAS Users Group proceedings: Health Care and Life Sciences* 2010; 1:9.

[23] Altman DG, Lausen B, Sauerbrei W. COMMENTARY - Dangers of Using "Optimal" Cutpoints in the Evaluation of Prognostic Factors. *Journal of the National*

Cancer Institute 1994; *86* (11), 829:835.

[24] Schroeder, M. Diagnosing and Dealing with Multicollinearity. *Western Journal of Nursing Research* 1990; *12* (2), 175:187.

[25] Li B, Lingsma HF, Steyerber EW et al. Logistic random effects regression models: a comparison of statistical packages for binary and ordinal outcomes. *BMC Medical Research Methodology - BioMed Central* 2011; *11* (77), 1:11.

[26] Allison, PD. *Logistic Regression Using SAS: Theory and Application, Second Edition.* Cary, NC, USA: SAS Institute Inc 1999; 60:67.

[27] Shapiro PA, Wulsin LR. (2009). Cardiovascular Disorders. In B. J. Sadock, V. A. Sadock, & P. Ruiz (Eds.), Kaplan and Sadock's Comprehensive Textbook of Psychiatry (9th Edition) (Vol. II). United States: Lippincott Williams & Wilkins.

[28] Devereaux PJ, Goldman L, Cook DJ et al. Perioperative cardiac events in patients undergoing noncardiac surgery: a review of the magnitude of the problem, the pathophysiology of the events and methods to estimate and communicate risk. *Canadian Medical Association Journal* 2005; *173* (6), 627:634.

[29] Noordzij PG, Poldermans D, Schouten O et al. Postoperative mortality in The Netherlands: a population-based analysis of surgery-specific risk in adults.

Anesthesiology 2010; *112* (5), 1105:1115.

[30] Moonseong H and Leon AC. Performance of a Mixed Effects Logistic Regression Model for Binary Outcomes With Unequal Cluster Size. *Journal of Biopharmaceutical Statistics* 2012; *15*(3), 513:526.

[31] Lee TH, Marcantonio ER, Mangione CM et al. Derivation and Prospective Validation of a Simple Index for Prediction of Cardiac Risk of Major Noncardiac Surgery. *Circulation* 1999; *100* (10), 1043:1049.

[32] Boersma E, Kertai MD, Schouten O et al. Perioperative cardiovascular mortality in noncardiac surgery: Validation of the Lee cardiac risk index. *The American Journal of Medicine* 2005; *118* (10), 1134:1141.

[33] Fleisher LA, Beckman, JA, Brown, KA et al. ACC/AHA 2007 Guidelines on Perioperative Cardiovascular Evaluation and Care for Noncardiac Surgery: Executive Summary: A Report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing Committee to Revise the 2002 Guidelines on Perioperative Cardiovascular Evaluation for Noncardiac Surgery). *Circulation* 2007; *116*, 1971:1996.

[34] Poldermans D, Bax JJ, Boersma E et al. Guidelines for pre-operative cardiac risk assessment and perioperative cardiac management in non-cardiac surgery: The Task

Force for Preoperative Cardiac Risk Assessment and Perioperative Cardiac Management in Non-cardiac Surgery of the European Society of Cardiology (ESC) and endorsed by the European Society of Anaesthesiology (ESA). *European Heart Journal* 2009*; 30*, 2769:2812.

[35] Goldman L, Caldera D, Nussbaum S et al. Multifactorial index of cardiac risk in noncardiac surgical procedures. *The New England Journal of Medicine* 1977; *297* (16), 845:850.

[36] Bertges DJ, Goodney PP, Zhao Y et al. The Vascular Study Group of New England Cardiac Risk Index (VSG-CRI) predicts cardiac complications more accurately than the Revised Cardiac Risk Index in vascular surgery patients. *Journal of Vascular Surgery* 2010; *52* (3), 674:683, 683e1:683e3.

[37] Fox J. Bootstrapping Regression Models. Appendix to An R and S-Plus Companion to Applied Regression. *Sage Publications* 2002.

[38] Steyerber EW, Harrell FE, Borsoom GJ et al. Internal validation of predictive models: Efficiency of some procedures for logistic regression analysis. *Journal of Clinical Epidemiology* 2001; *54* (8), 774:781.

[39] Fox AA, Body SC. Assessment of Preoperative B-Type Natriuretic Peptide in Adult Surgeries: Is It Useful? *International Anesthesia Research Society* 2011*; 112* (5), 1005:1007.

[40] Karthikeyan G, Moncur RA, Levine O et al. Is a Pre-Operative Brain Natriuretic Peptide or N-Terminal Pro–B-Type Natriuretic Peptide Measurement an Independent Predictor of Adverse Cardiovascular Outcomes Within 30 Days of Noncardiac Surgery? A Systematic Review and Meta-Analysis of Observational Studies. *Journal of the American College of Cardiology* 2009; *54* (17), 1599:1606.

[41] Harrison A, Morrison LK, Krishnaswamy P et al. B-type natriuretic peptide predicts future cardiac events in patients presenting to the emergency department with dyspnea. *Annals of Emergency Medicine* 2002; *39* (2), 131:138.

[42] Neuhaus JM, Kalbfleisch JD and Hauck WW. A Comparison of Cluster-Specific and Population-Averaged Approaches for Analyzing Correlated Binary Data. *International Statistical Review* 1991; *59*(1), 25:35.

[43] Mazumdar M, Smith A, Bacik J. Methods for categorizing a prognostic variable in a multivariable setting. *Statististics in Medicine* 2003; *22* (4), 559:571.

[44] Faraggi, D., & Simon, R. A Simulation Study of Cross-Validation for Selecting an Optimal Cutpoint in Univariate Survival Analysis. *Statistics in Medicine* 1996; *15* (20), 2203:2213.

[45] Kertai M, Boersma E, Bax J, Heijenbrok-Kal M et al. A meta-analysis comparing the prognostic accuracy of six diagnostic tests for predicting perioperative cardiac risk in patients undergoing major vascular surgery. *Heart* 2003*; 89* (11), 1327:1334.

[46] Altman DG, Berthold L, Sauerbrei W, Schumacher, M. Dangers of Using "Optimal" Cutpoints in the Evaluation of Prognostic Factors. *Journal of the National Cancer Institute* 1994; *86* (11), 829:835.

[47] Landesberg G, Mosseri M, Shatz V et al. Cardiac Troponin After Major Vascular Surgery - The Role of Perioperative Ischemia, Preoperative Thallium Scanning, and Coronary Revascularization. *Journal of the American College of Cardiology* 2004; *44* (3), 569:575.

[48] Lausen B, & Schumacher M. Evaluating the effect of optimized cutoff values in the assessment of prognostic factors. *Computational Statistics & Data Analysis* 1995; *21* (3), 307:326.

[49] Pencina M, D'Agostino Sr R, D'Agostino Jr R, Vasan R. Evaluating the added predictive ability of a new marker: From area under the ROC curve to reclassification and beyond. *Statistics in Medicine* 2008*; 27* (2), 157:172

Appendix A

Tables of Acronyms, Variables, Outcomes, and Models

Table 1.1.1: Description of Acronyms

Variable/ Term	**Description**
AGE	Patient's age prior to surgery
AGE_thrshld	Age threshold of 65 years old
AIC	Akaike Information Criteria
AUC	Area under the ROC curve; also referred to as the c-statistic
BNP	B-type natriuretic peptide concentration level (in pg/mL)
BNP_thrshld	Final BNP cutpoint determined by the minimum p-value method
CCF	Congestive cardiac failure
CI	Confidence interval
df	Degrees of freedom
GEE	Generalized estimating equations
MACE	Major adverse cardiac event
MELR	Mixed effects logistic regression
MI	Myocardial infarction
NLMIXED	Nonlinear mixed models
NP	Natriuretic peptides: BNP and NTproBNP
NTproBNP	N-terminal pro-B-type natriuretic peptide
NTproBNP_thrshld	Final NTproBNP cutpoint determined by the minimum p-value method
OR	Odds ratio
p	p-value
P.Chi	Pearson Chi Square statistic
QADAS	Quality assessment of diagnostic accuracy studies
RCRI	Revised cardiac risk index
ROC	Receiver operator characteristic curve
RR	Relative risk
SAS	Statistical analysis software
SD	Standard deviation
SLR	Simple logistic regression (using MINP_thrshld for NP cutpoints)
SLR_RODSETH	Simple logistic regression (using ROC_thrshld for NP cutpoints)
TOL	Tolerance level
VIF	Variance inflation factor

Table 1.1.2: Description of Variables

Variable	Type of Variable	Description
AGE	Continuous	Patient's prior to surgery
AGE_thrshld	Binary	Age threshold of 65 0=Age less than 65 1=Age greater than or equal to 65
BNP	Continuous	Measure of patient's pre-operative B-type natriuretic peptide concentration level (in pg/mL)
CCF	Binary	Patient has experienced congestive cardiac failure 0 = no, 1 = yes
CEREBRO_ VASCULAR_DISEASE	Binary	Patient has experienced cerebrovascular disease 0 = no, 1 = yes
DIABETES	Binary	Patient has diabetes mellitus 0 = no, 1 = yes
HIGH_CREAT	Binary	Patient has experienced renal failure (with creatinine levels>2mg/dl) 0 = no, 1 = yes
Hx_IHD	Binary	Patient has a history of coronary artery disease 0 = no, 1 = yes
MACE	Binary	Major adverse cardiac event 0= no, 1 = yes
MINP_thrshld	Binary	Patient NP levels exceed respective threshold values; thresholds determined by minimum p-value method 0 = no, 1 = yes
ROC_thrshld	Binary	Patient NP levels exceed respective threshold values; thresholds determined by ROC curve method [1] 0 = no, 1 = yes
STUDY	Nominal	Identifies which study a patient's information was obtained from 1= study with BNP data [16] 2= study with BNP data [17] 3= study with BNP data [18] 4= study with BNP data [19] 5= study with BNP data [20] 6= study with NTproBNP data [21]
SURGERY_TYPE	Nominal	Identifies the type of vascular surgery patient has undergone (infrainguinal, aortoiliac, and not specified) 0=Infrainguinal 1= Aortoiliac 2=not specified*

*Patients with SURGERY_TYPE=2 are only used for the minimum p-value method to determine thresholds for BNP and NTproBNP; they are not included in any regression analysis.
**NP levels= pre-operative BNP and NTproBNP concentration levels

Table 1.1.3: Description of Outcomes

Outcome	**Description**
MACE	-Major Adverse Cardiac Event -Composite endpoint of cardiac death and non-fatal myocardial infarction within 30 days after vascular surgery. 0= no, 1 = yes
All-Cause Mortality	-Patient experiences cardiac death within 30 days after vascular surgery 0= no, 1 = yes
Cardiac Death	-Patient experiences cardiac death within 30 days after vascular surgery 0= no, 1 = yes
Non-fatal MI	-Patient experiences a non-fatal myocardial infarction within 30 days after vascular surgery 0= no, 1 = yes

Table 1.2.1: Summary of Primary Objectives and Analysis

CLINICAL OBJECTIVE:
1. Determine optimal cutpoints for BNP and NTproBNP in predicting MACE.
STATISTICAL OBJECTIVE:
1. Employ the minimum p-value method to dichotomize NP levels and compare with the ROC curve approach

PROCESS	OUTCOME	PREDICTORS	STUDIES	SIZE OF DATA	METHODS
Determine threshold values for BNP and NTproBNP	MACE (Binary)	- BNP - NTproBNP	BNP: 5 NTproBNP: 1 Total: 6	BNP: 632 NTproBNP: 218 Total: 850	Minimum P-Value Method

CLINICAL OBJECTIVE:
2. Determine a prediction model for predicting MACE within 30 days after a vascular surgery.
STATISTICAL OBJECTIVE:
2. Determine and validate a prediction model with a MACE outcome
3. Examine the robustness final prediction model

PROCESS	OUTCOME	PREDICTORS	STUDIES	SIZE OF DATA	METHODS
STEP 1: Create a prediction model based on covariates used in Rodseth's final model [1].	MACE (Binary)	-MINP_THRSHLD -SURGERY_TYPE* -DIABETES*	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	SLR
STEP 2: Compare results from Minimum p-value method and ROC curve approach.	MACE (Binary)	-MINP_THRSHLD -SURGERY_TYPE◆ - DIABETES◆ -ROC_THRSHLD◆	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	SLR
STEP 3: Determine if inclusion/ removal of any covariates will create a better-fit model.	MACE (Binary)	-MINP_THRSHLD -SURGERY_TYPE◆ -DIABETES◆ -AGE_THRSHLD -HIGH_CREAT -CCF -Hx_IHD -CEREBRO_ VASCULAR_ DISEASE	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	SLR
STEP 4: Validate model	MACE (Binary)	-MINP_THRSHLD -SURGERY_TYPE◆ -DIABETES	**	BNP: 624 607 NTproBNP: 218 393 Total: 843 000	SLR
STEP 5: Determine sensitivity of final model	MACE (Binary)	-MINP_THRSHLD -SURGERY_TYPE◆ -DIABETES◆ -STUDY	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	MELR GEE

*All variables are defined as covariates for MACE among literature; variables identified with '◆' are statistically significant predictors under Rodseth's analysis [1]; MACE=major adverse cardiac event; SLR=simple logistic regression; GEE= generalized estimating equations; MELR=mixed effects logistic regression

**1000 bootstrap samples were generated using simple random sampling with replacement on the updated dataset with four studies containing BNP data and 1 study with NTproBNP information. Study 5 [20] and SURGERY_TYPE=2 patients were not included in these bootstrap samples.

Table 1.2.2: Summary of Secondary Objectives and Analysis

Secondary Objectives:

1. Determine individual prediction models for outcomes: all-cause mortality, cardiac death, non-fatal MI within 30 days of vascular surgery

OBJECTIVE	OUTCOME	PREDICTORS	STUDIES	SIZE OF DATA	METHODS
To determine a model for predicting all-cause mortality within 30 days of vascular surgery	All-cause mortality (Binary)	- MINP_THRSHLD □ -SURGERY_TYPE □ - DIABETES□ - STUDY -AGE_THRSHLD -HIGH_CREAT -CCF -Hx_IHD - CEREBRO_ VASCULAR_DISEASE	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	MELR
To determine a model for predicting cardiac death within 30 days of vascular surgery	Cardiac death (Binary)	- MINP_THRSHLD □ -SURGERY_TYPE □ - DIABETES□ - STUDY -AGE_THRSHLD -HIGH_CREAT -CCF -Hx_IHD - CEREBRO_ VASCULAR_ DISEASE	BNP: 4 NTproBNP: 1 Total: 5	BNP: 625 NTproBNP: 218 Total: 843	MELR
To determine a model for predicting non-fatal MI within 30 days of vascular surgery	Non-fatal MI (Binary)	- MINP_THRSHLD □ -SURGERY_TYPE □ - DIABETES□ - STUDY -AGE_THRSHLD -HIGH_CREAT -CCF -Hx_IHD - CEREBRO_ VASCULAR_ DISEASE	BNP: 625 NTproBNP: 218 Total: 843	BNP: 625 NTproBNP: 218 Total: 843	MELR

*Variables listed in the above table were considered as covariates for each secondary outcome as they are all cardiovascular-related events; covariates with a ' □' are statistically significant in our final model for predicting MACE and will be the only factors used in this secondary analysis. MELR=mixed effects logistic regression; STUDY will be the random effect in each MELR model.

Appendix B

Tables of Patient Characteristics for Each Outcome

Table 2.1.1: Patient Characteristics for MACE

Variable	Total (n=850)	MACE (n=75)	NO MACE (n=775)	P-Value	Test Statistic	Test
Age (yrs): mean(SD)	65.4 (12.1)	69.4 (8.8)	65.0 (12.3)	0.002	3.07	T-Test
Sex (men): n(%)	391 (46.0)	36 (48.0)	355(45.8)	0.696	0.15	P. Chi
+missing	218 (26.0)					
Type of Vascular Surgery: n(%)				0.229	2.95	P. Chi
a) Infrainguinal	629 (74.0)	50 (66.7)	579 (74.7)			
b) Aortoiliac	217 (25.5)	25 (33.3)	192 (24.8)			
c) Not specified	4 (0.5)	0	4 (0.5)			
RCRI Class: n(%)				0.002	12.50	P. Chi
a) Low (RCRI 0)	320 (37.6)	19 (25.3)	301 (38.8)			
b) Intermediate (RCRI 1 or 2)	476 (56.0)	45 (60.0)	431 (55.6)			
c) High (RCRI 3)	54 (6.4)	11 (14.7)	43 (5.5)			
RCRI Components: n(%)						
Coronary artery disease	327 (38.5)	42 (56.0)	285 (36.8)	0.001	10.68	P. Chi
Congestive heart failure	64 (7.5)	14 (18.7)	50 (6.5)	<0.001	14.65	P. Chi
Cerebrovascular disease	145 (17.1)	8 (10.7)	137 (17.7)	0.123	2.38	P. Chi
Diabetes mellitus	204 (24.0)	25 (33.3)	179 (23.1)	0.048	3.93	P. Chi
Creatinine (≥2 mg/dl)	28 (3.3)	6 (8.0)	22 (2.8)	0.017	5.72	P. Chi

*yrs=years; SD=Standard Deviation; P. Chi=Pearson chi-square test; RCRI=Revised Cardiac Risk Index

Table 2.1.2: Patient Characteristics for All-Cause Mortality

Variable	Total (n=850)	All-Cause Mortality (n=30)	No All-Cause Mortality (n=820)	P-Value	Test Statistic	Test
Age (yrs): mean(SD)	65.4 (12.1)	69.1 (11.4)	65.2 (12.1)	0.0809	1.75	T-Test
Sex (men): n(%)	391 (46.0)	22 (29.3)	369 (47.6)	0.112	2.52	P. Chi
+missing	218 (26)					
Type of Vascular Surgery: n(%)				0.307*	2.14	P. Chi
a) Infrainguinal	629 (74)	19 (25.3)	610 (78.7)			
b) Aortoiliac	217 (25.5)	11 (14.7)	206 (26.6)			
c) Not specified	4 (0.5)	0	4 (0.5)			
RCRI Class: n(%)				0.00150	13.05	P. Chi
a) Low (RCRI 0)	320 (37.6)	5 (6.7)	315 (40.6)			
b) Intermediate (RCRI 1 or 2)	476 (56.0)	19 (25.3)	457 (59.0)			
c) High (RCRI 3)	54 (6.4)	6 (8.0)	48 (6.2)			
RCRI Components: n(%)						
Coronary artery disease	327 (38.5)	15 (20.0)	312 (40.3)	0.186	1.75	P. Chi
Congestive heart failure	64 (7.5)	7 (9.3)	57 (7.4)	0.005*	11.16	P. Chi
Cerebrovascular disease	145 (17.1)	5 (6.7)	140 (18.1)	0.954	0.0034	P. Chi
Diabetes mellitus	204 (24.0)	12 (16.0)	192 (24.8)	0.0367	4.36	P. Chi
Creatinine (≥2 mg/dl)	28 (3.3)	5 (6.7)	23 (3.0)	0.0022*	17.46	P. Chi

'*'Variables with 25% of the cells having expected counts less than 5. (Asymptotic) Chi-Square may not be a valid test, and so the Exact p-value for the chi-square statistics is used.
**SD=Standard Deviation; P. Chi=Pearson chi-square test; RCRI=Revised Cardiac Risk Index

Table 2.1.3: Patient Characteristics for Cardiac Death

Variable	Total (n=850)	Cardiac Death (n=75)	NO Cardiac Death (n=775)	P-Value	Test Statistic	Test
Age (yrs): mean(SD)	65.4 (12.1)	71.5 (8.8)	65.2 (12.1)	0.0254	2.24	T-Test
Sex (men): n(%)	391 (46.0)	13 (17.3)	378 (48.8)	0.359	0.84	P. Chi
+missing	218 (26.0)					
Type of Vascular Surgery: n(%)				0.350*	1.37	P. Chi
a) Infrainguinal	629 (74.0)	12 (16.0)	617 (79.6)			
b) Aortoiliac	217 (25.5)	7 (9.3)	210 (27.1)			
c) Not specified	4 (0.5)	0	4 (0.5)			
RCRI Class: n(%)				0.0098	9.25	P. Chi
a) Low (RCRI 0)	320 (37.6)	3 (4.0)	317 (40.9)			
b) Intermediate (RCRI 1 or 2)	476 (56.0)	12 (16)	464 (59.9)			
c) High (RCRI 3)	54 (6.4)	4 (5.3)	50 (6.5)			
RCRI Components: n(%)						
Coronary artery disease	327 (38.5)	11 (14.7)	316 (40.8)	0.0784	3.098	P. Chi
Congestive heart failure	64 (7.5)	5 (6.7)	59 (7.6)	0.0105*	9.851	P. Chi
Cerebrovascular disease	145 (17.1)	2 (2.7)	143 (18.5)	0.556*	0.586	P. Chi
Diabetes mellitus	204 (24.0)	7 (9.3)	197 (25.4)	0.274*	1.757	P. Chi
Creatinine (≥2 mg/dl)	28 (3.3)	4 (5.3)	24 (3.1)	0.0026*	19.25	P. Chi

'*'Variables with 25% of the cells having expected counts less than 5. (Asymptotic) Chi-Square may not be a valid test, and so the Exact p-value for the chi-square statistics is used.
**SD=Standard Deviation; P. Chi=Pearson chi-square test; RCRI=Revised Cardiac Risk Index

Table 2.1.4: Patient Characteristics for Non-Fatal MI

Variable	Total (n=850)	Non-Fatal MI (n=75)	NO Non-Fatal MI (n=775)	P-Value	Test Statistic	Test
Age (yrs): mean(SD)	65.4 (12.1)	68.7 (8.7)	65.1 (12.3)	0.0305	2.17	T-Test
Sex (men): n(%)	391 (46.0)	23 (30.7)	368 (47.5)	0.861	0.031	P. Chi
+missing	218 (26.0)					
Type of Vascular Surgery: n(%)				0.443*	1.62	P. Chi
a) Infrainguinal	629 (74.0)	38 (50.7)	591 (76.3)			
b) Aortoiliac	217 (25.5)	18 (24.0)	199 (25.7)			
c) Not specified	4 (0.5)	0	4 (0.5)			
RCRI Class: n(%)				0.0384	4.97	P. Chi
a) Low (RCRI 0)	320 (37.6)	16 (21.3)	304 (39.2)			
b) Intermediate (RCRI 1 or 2)	476 (56.0)	33 (44)	443 (57.2)			
c) High (RCRI 3)	54 (6.4)	7 (9.3)	47 (6.1)			
RCRI Components: n(%)						
Coronary artery disease	327 (38.5)	31 (41.3)	296 (38.2)	0.0072	7.22	P. Chi
Congestive heart failure	64 (7.5)	9 (12.0)	55 (7.1)	0.0193*	6.28	P. Chi
Cerebrovascular disease	145 (17.1)	6 (8.0)	139 (17.9)	0.192	1.71	P. Chi
Diabetes mellitus	204 (24.0)	18 (24.0)	186 (24)	0.140	2.18	P. Chi
Creatinine (≥2 mg/dl)	28 (3.3)	2 (2.7)	26 (3.4)	>0.999*	0.015	P. Chi

'*'Variables with 25% of the cells having expected counts less than 5. (Asymptotic) Chi-Square may not be a valid test, and so the Exact p-value for the chi-square statistics is used.
**SD=Standard Deviation; P. Chi=Pearson chi-square test; RCRI=Revised Cardiac Risk Index

Appendix C

Figures and Flowcharts of Statistical Methods

Table 3.1.1: Description of Key Methods of Analysis

Method of Analysis	Description	Statistical Software and Procedures
Minimum P-Value Method	Used to dichotomize BNP and NTproBNP and to determine thresholds for MINP_thrshld with a MACE outcome	-Macros obtained from (Glassman & Mazumdar, 2000). -RStudio 0.96.326
MELR	Mixed effects logistic regression Random effect=STUDY	-PROC GLIMMIX -SAS 9.3
GEE	Generalized estimating equations	-PROC GENMOD -SAS 9.3
SLR	Simple logistic regression	-PROC LOGISTIC -SAS 9.3

*MELR= mixed effects logistic regression; GEE=generalized estimating equations; SLR=simple logistic regression

Figure 3.1.1: Flow Chart of Determining Threshold Values
for BNP and NTproBNP

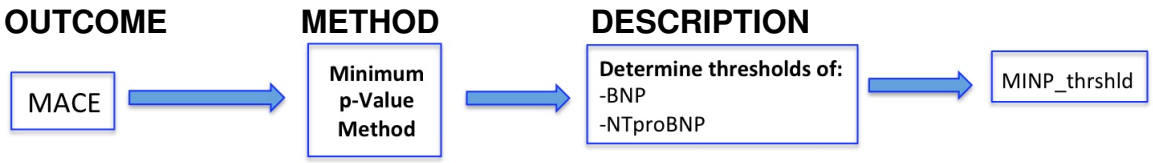


Figure 3.1.2: Flow Chart of Primary Analysis

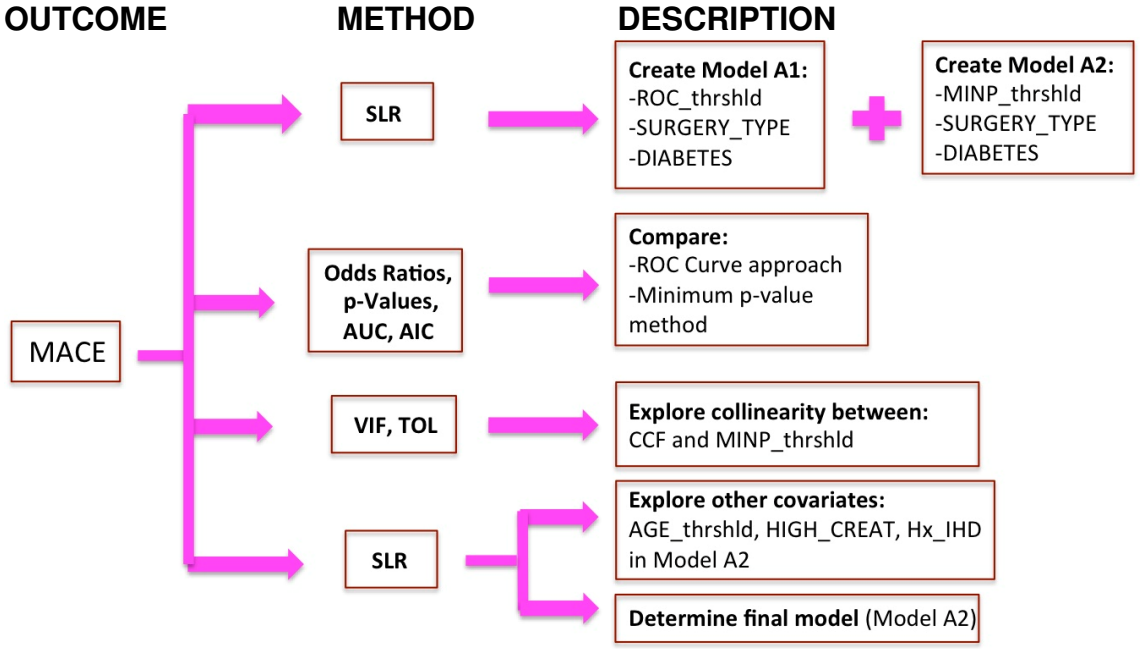


Figure 3.1.3: Flow Chart of Validation Analysis

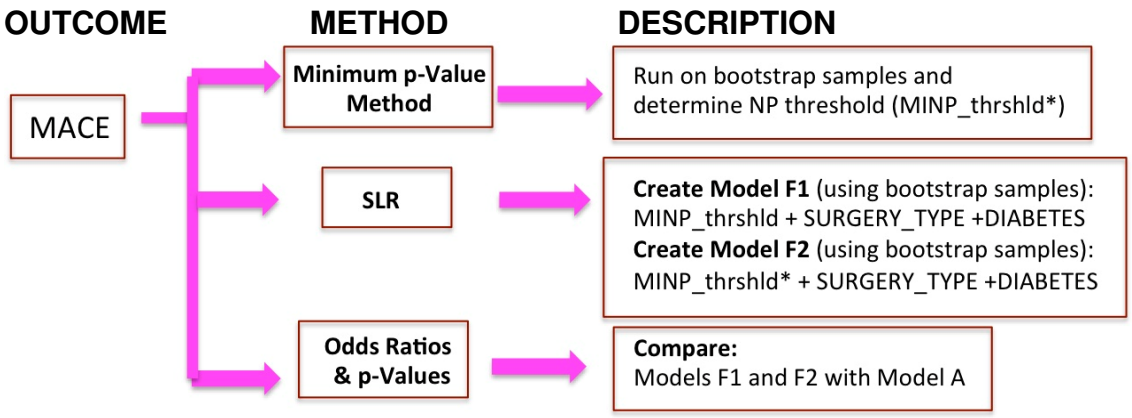


Figure 3.1.4: Flow Chart of Sensitivity Analysis

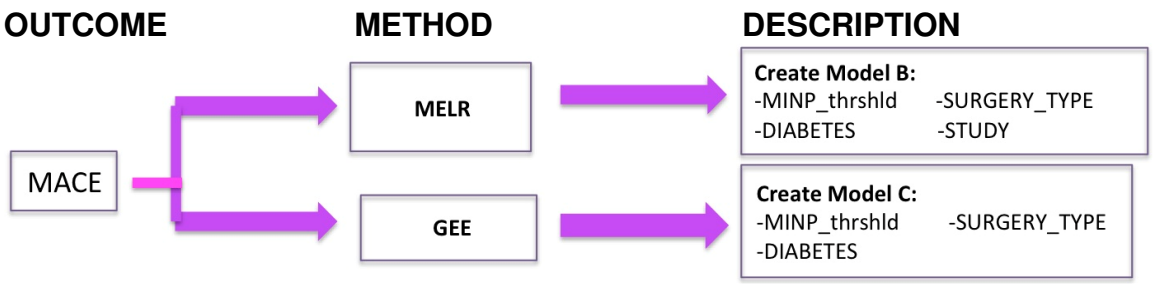
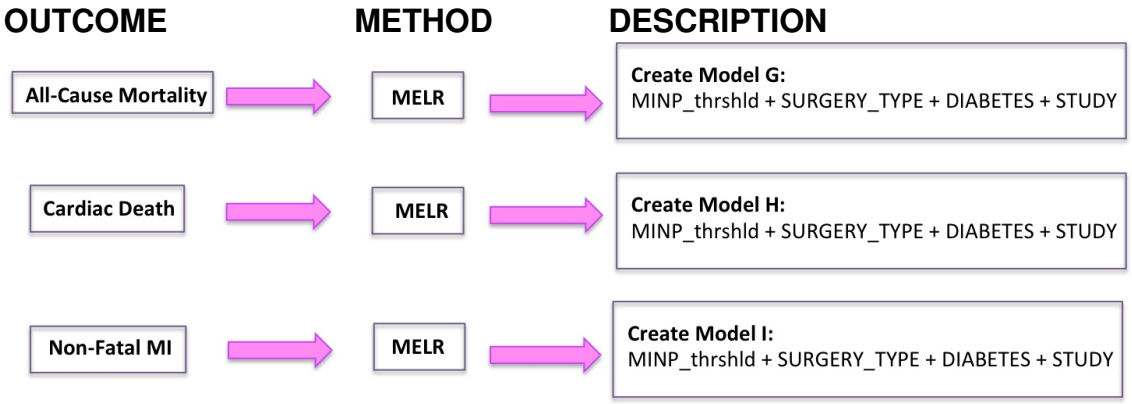


Figure 3.1.5: Flow Chart of Secondary Analysis



Appendix D

Plots and Tables of Preliminary Analysis and the Minimum P-Value Method

Figure 4.2.1: Boxplots of BNP Values

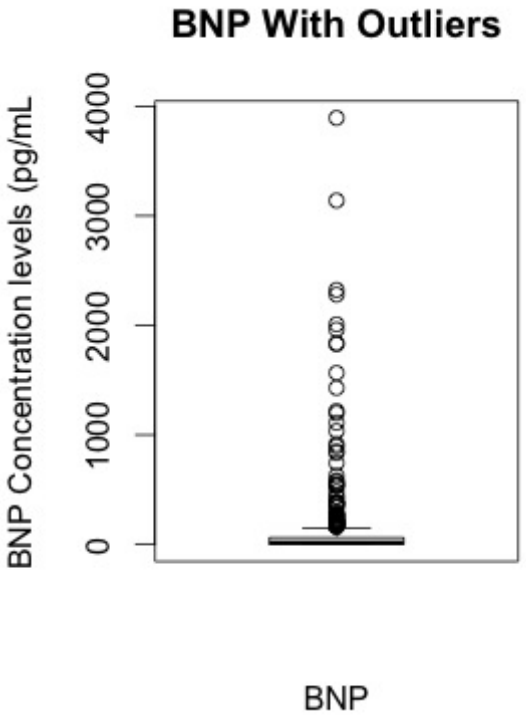


Figure 4.2.2: Boxplots of BNP Values, with Outliers Removed

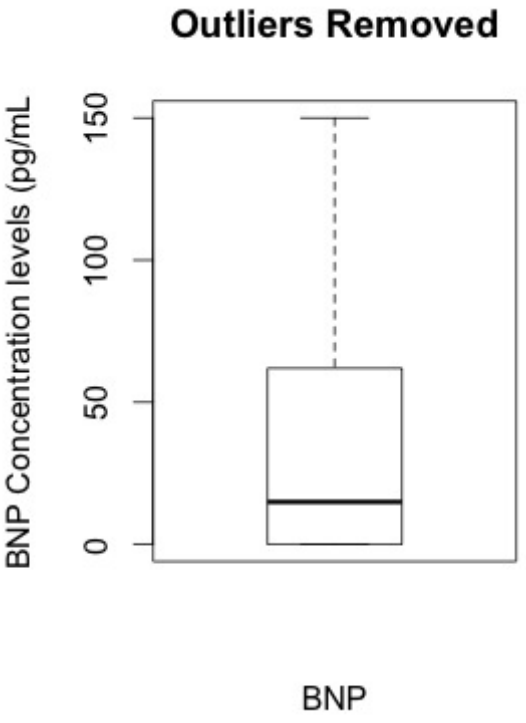


Figure 4.2.3: Boxplots of NTproBNP Values

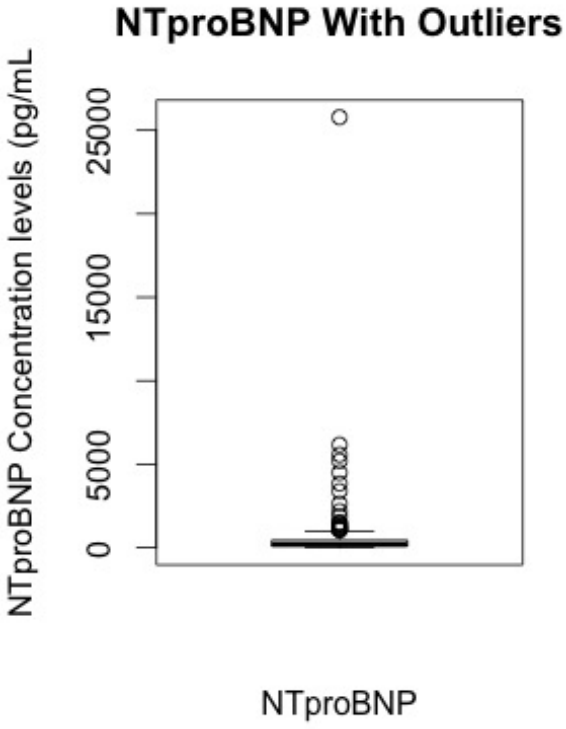


Figure 4.2.4: Boxplots of NTproBNP Values, with Outliers Removed

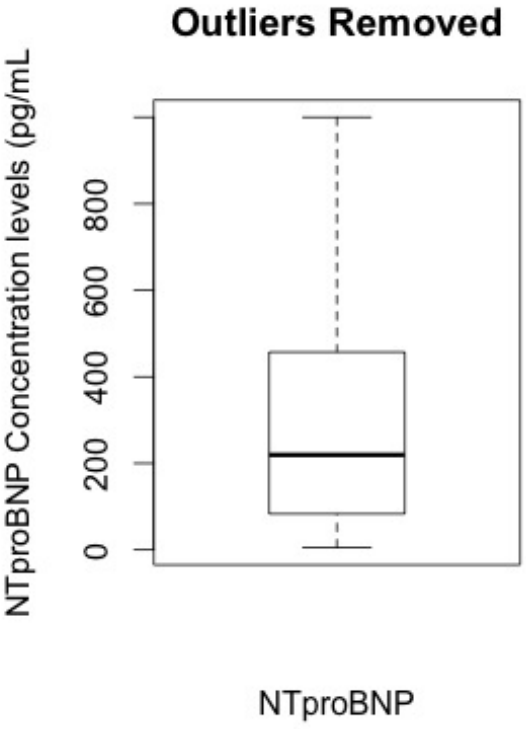


Figure 4.2.5: Observed Pre-Operative BNP Concentration Levels

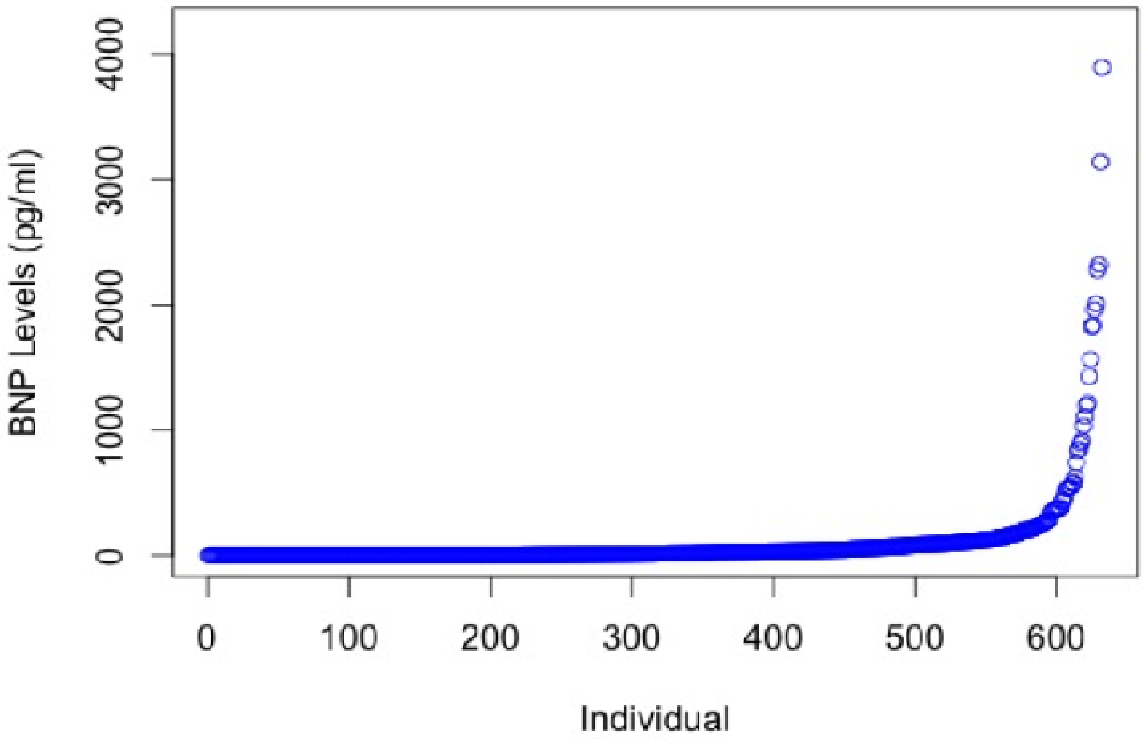


Figure 4.2.6: Observed Pre-Operative NTproBNP Concentration Levels

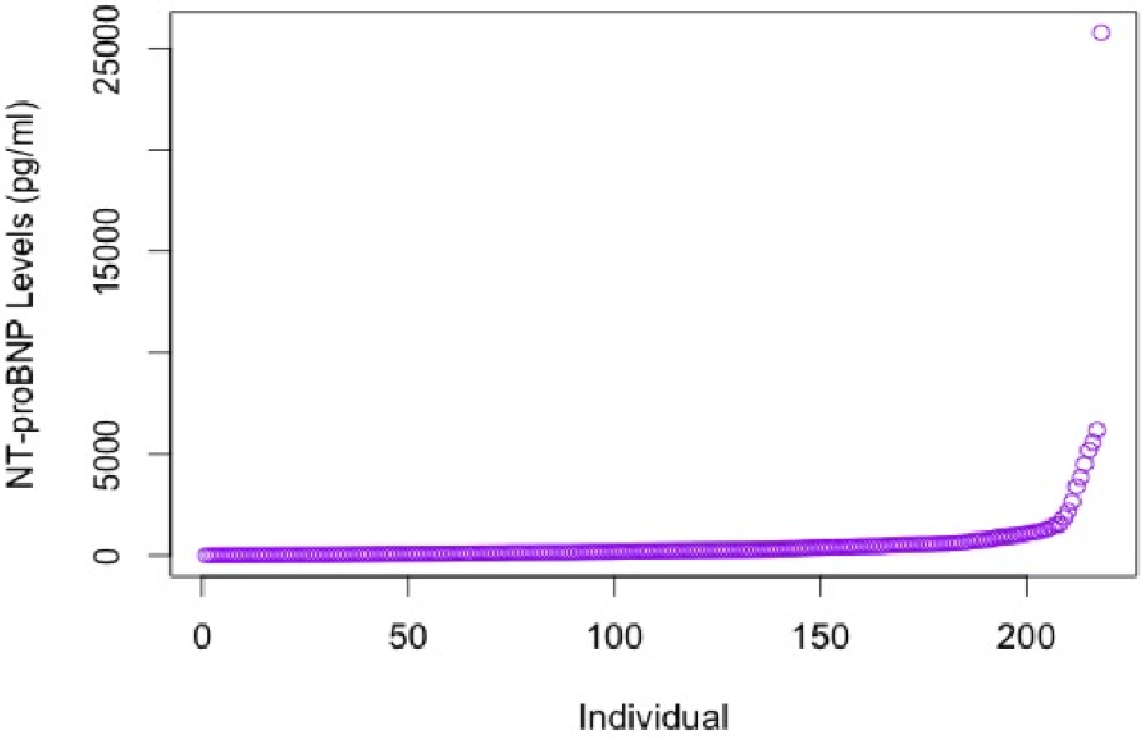


Figure 4.2.7: Ideal Relationship between BNP Concentration Levels and MACE

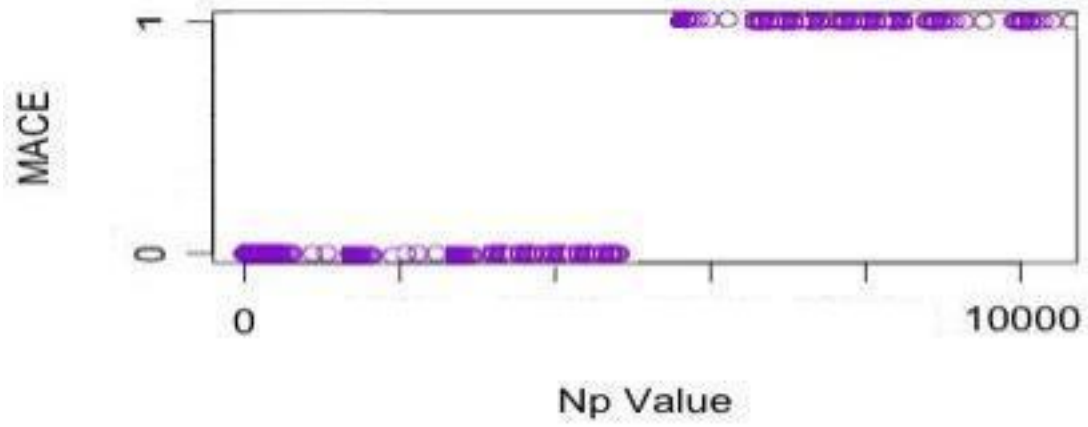


Figure 4.2.8: Actual Relationship between BNP Concentration Levels and MACE

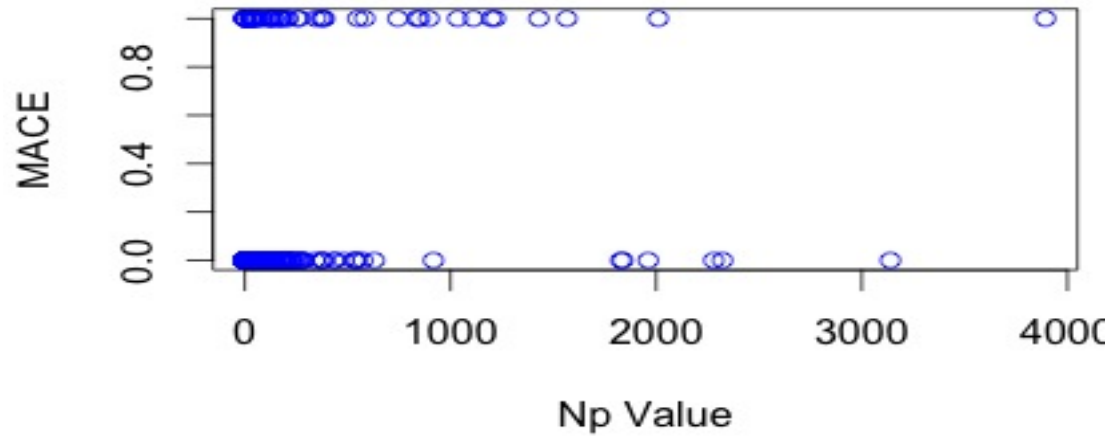


Figure 4.2.9: Actual Relationship between NTproBNP Concentration Levels and MACE

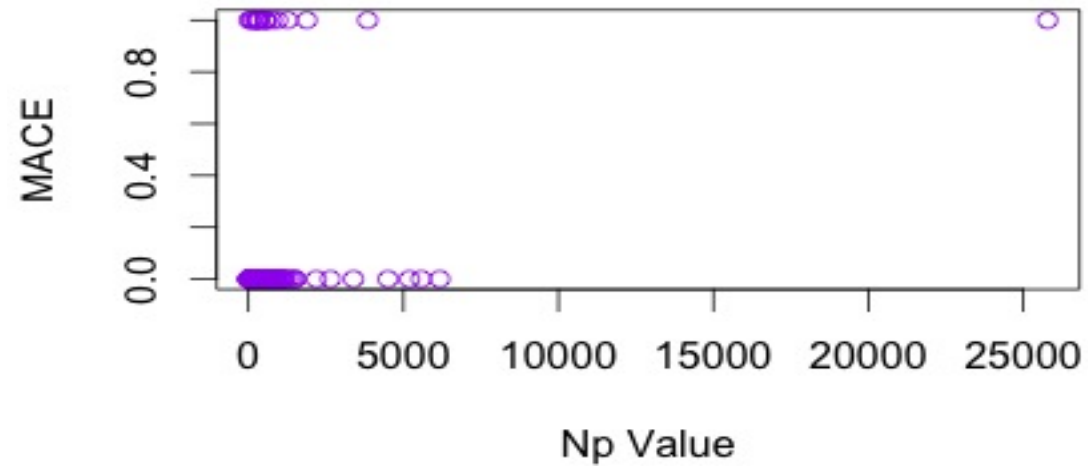


Figure 4.3.1: Potential BNP Threshold Values and Corresponding Chi-Square Statistics

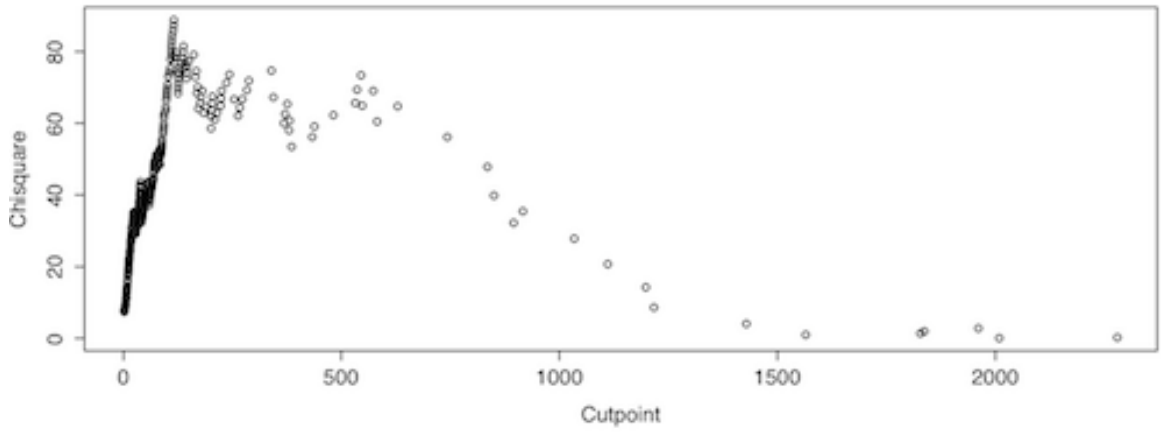


Figure 4.3.2: Potential BNP Threshold Values and Corresponding P-Values

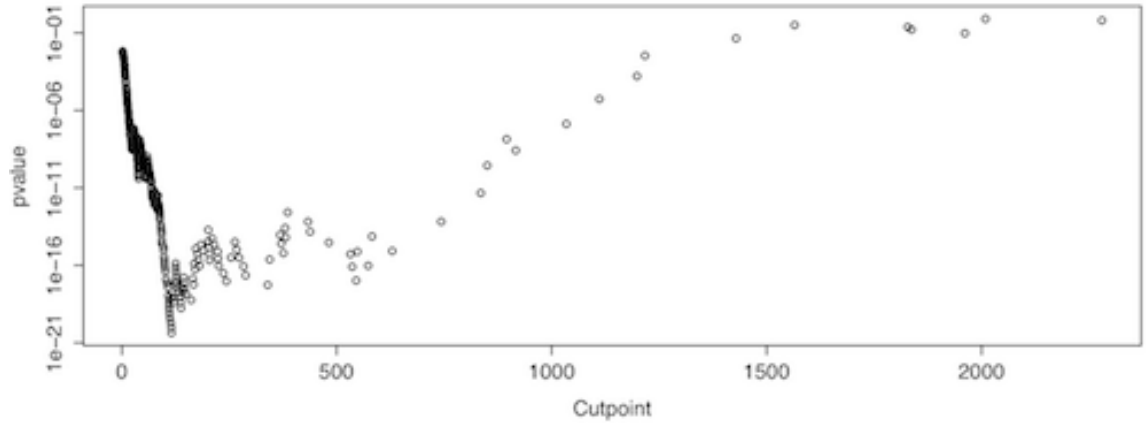


Figure 4.3.3: Potential BNP Threshold Values and Corresponding Relative Risks

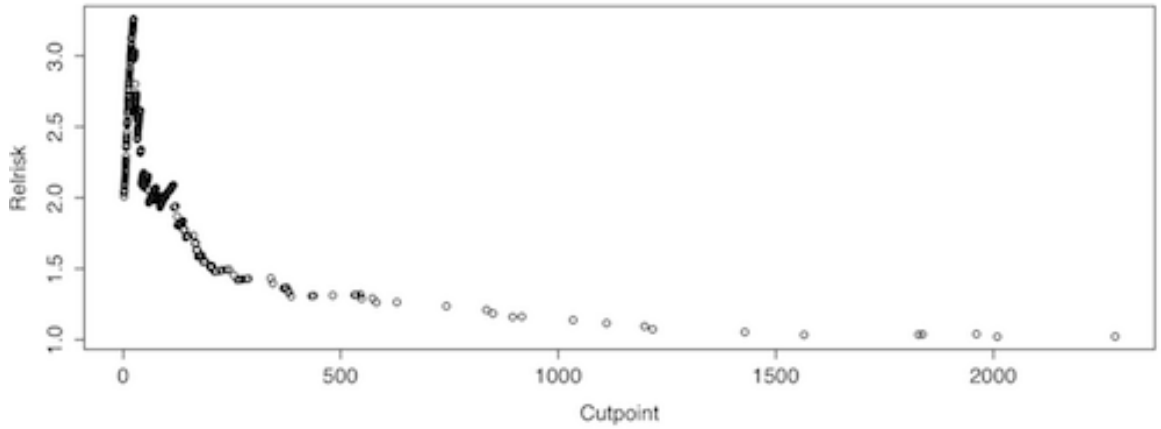


Figure 4.3.4: Potential NTproBNP Threshold Values and Corresponding Chi-Square Statistics

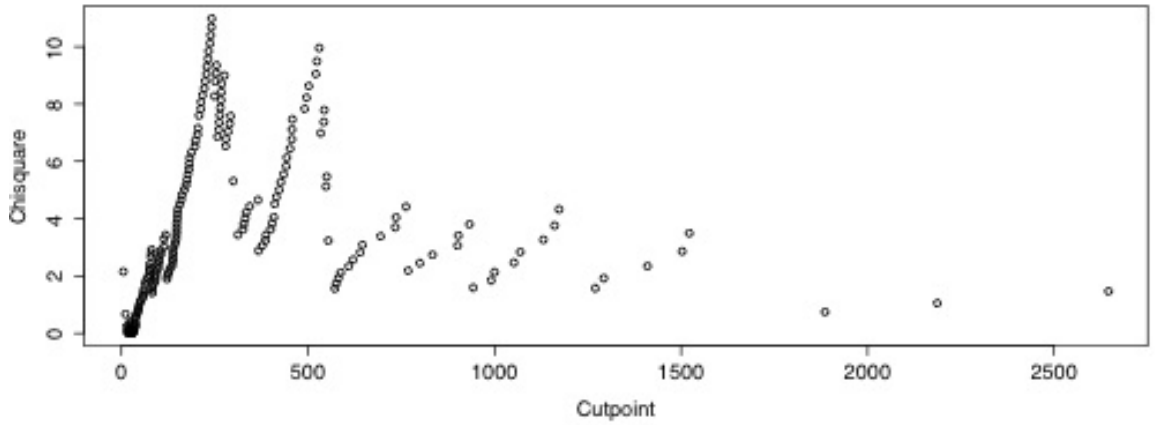


Figure 4.3.5: Potential NTproBNP Threshold Values and Corresponding P-Values

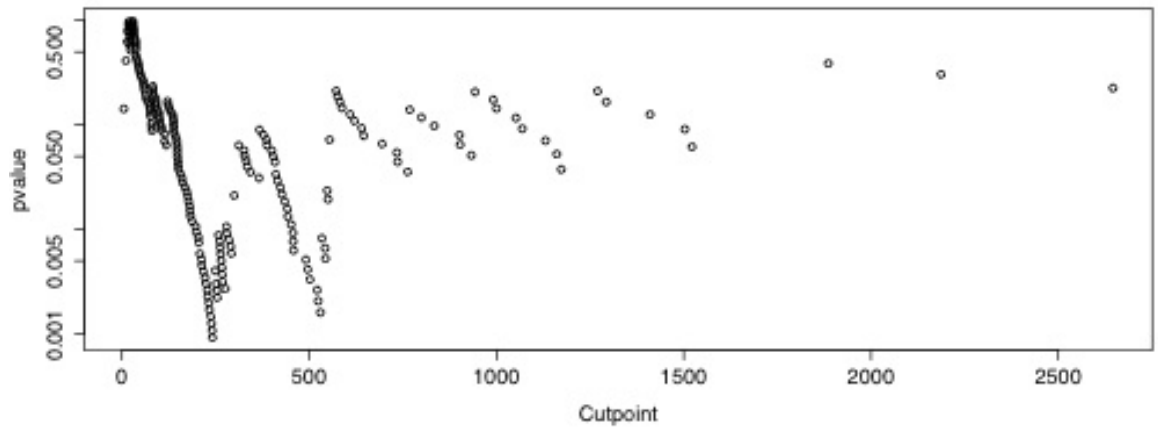


Figure 4.3.6: Potential NTproBNP Threshold Values and Corresponding Relative Risks

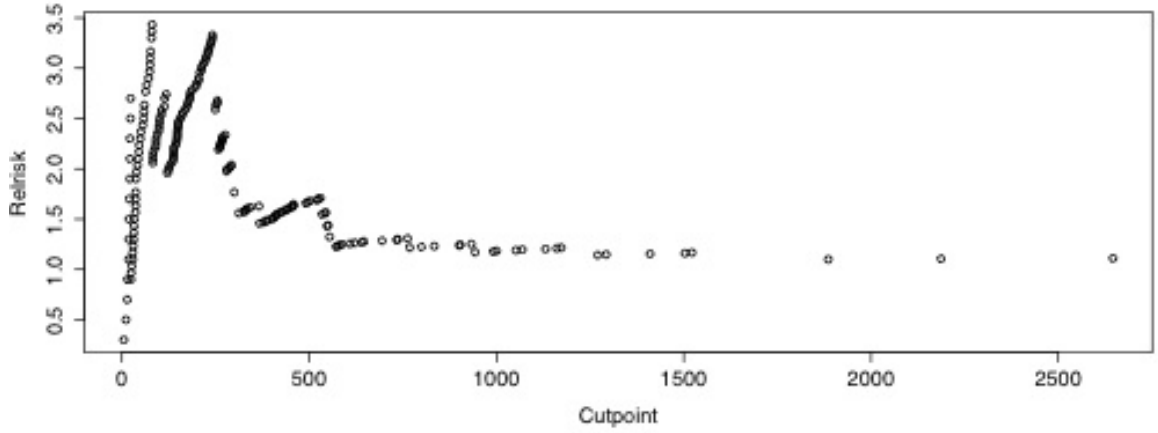


Table 4.3.7: Results of the Minimum P-Value Method

COVARIATE	BNP	NTproBNP
Threshold Value	115.57pg/mL	241.7 pg/mL
Chi-Squared Statistic	88.79	10.98
Relative Risk	2.09	3.33
Minimum p-value (P_{min})	4. 39E-21	9.20E-4
P_{ms}	6.85E-19	0.030
P_{alt510}, at 5%	1.05E-18	0.030
P_{alt510}, at 10%	7.81E-19	0.023
P_{modbon}	<0.0001	0.0001

P_{min}=minimum p-value of the evaluated potential thresholds; P_{ms}, P_{alt510}, P_{modbon}= p-value adjustment formulas for inflation of the Type I Error Rate; BNP=B-type Natriuretic Peptide; NTproBNP=N-Terminal-pro-BNP;

Appendix E

Logistic Regression Analysis and Determining Risk Estimates for a MACE Outcome

Table 4.5.1: Breakdown of Studies and Outcomes

Study	Included Biomarker	Reference	No. of Patients	With MACE n(%)	With All-Cause Mortality n(%)	With Cardiac Death n(%)	With Non-Fatal MI n(%)
1	BNP	(Bolliger et al., 2009)[16]	133	4(3)	4(3)	2(2)	2(2)
2	BNP	(Biccard et al., 2011)[17]	297	26(9)	10(3)	5(2)	21(7)
3	BNP	(Cuthbertson et al., 2007) [18]	70	2(3)	1(1)	1(1)	1(1)
4	BNP	(Gibson et al., 2004) [19]	129	22(17)	12(9)	9(7)	13(10)
5	BNP	(Leibowitz et al., 2008) [20]	3	2(67)	2(67)	1(33)	1(33)
6	NTproBNP	(Mahla et al.), 2004 [21]	218	19(9)	1(0)	1(0)	18(8)

*BNP=Pre-operative concentration levels of B-type naturietic peptide; NTproBNP= Pre-operative concentration levels of N-terminal pro-B-type natriuretic peptide; MACE=major adverse cardiac event; MI=myocardial infarction

**The percentages calculated for each outcome with MACE, all-cause mortality, cardiac death, and non-fatal MI are based on the total number of individuals in each study.

Table 4.6.1: Breakdown of Models

Outcome	Type of Analysis	Model	Method	Variables
MACE	Primary	A1	SLR	ROC_thrshld, SURGERY_TYPE, DIABETES
		A2	SLR	MINP_thrshld, SURGERY_TYPE, DIABETES
		A3	SLR	MINP_thrshld, SURGERY_TYPE, DIABETES, AGE_thrshld
		A4	SLR	MINP_thrshld, SURGERY_TYPE, DIABETES, HIGH_CREAT
		A5	SLR	MINP_thrshld, SURGERY_TYPE, DIABETES, Hx_IHD
	Sensitivity	B	MELR	MINP_thrshld, SURGERY_TYPE, DIABETES, STUDY
		C	GEE	MINP_thrshld, SURGERY_TYPE, DIABETES, STUDY
	Validation	F	SLR	MINP_thrshld, SURGERY_TYPE DIABETES
All-Cause Mortality	Secondary	G	MELR	MINP_thrshld, SURGERY_TYPE STUDY
Cardiac Death		H	MELR	MINP_thrshld, SURGERY_TYPE STUDY
Non-fatal MI		I	MELR	MINP_thrshld, SURGERY_TYPE STUDY

*ROC_thrshld=indicator variable of NP thresholds determined by the ROC curve method [1]; MINP_thrshld=indicator variable of NP thresholds determined by minimum p-value method; SURGERY_TYPE=type of surgery; DIABETES=whether or not the patient has diabetes mellitus; AGE_thrshld=if an individual is over 65 years old; HIGH_CREAT= history of renal failure; Hx_IHD= history of coronary artery disease; MINP_thrshld** =an indicator variable of the NP thresholds determined from the average results of the bootstrap samples; SLR= simple logistic regression; MELR=mixed effects logistic regression; GEE=generalized estimating equations

Table 4.6.2: Determining a Prediction Model for MACE

Model	Effect	Estimate	OR	95% OR CI	P-Value	AUC	AIC
A1	Intercept	-3.578	-	-	<.0001	0.768	431.63
	ROC_thrshld	2.126	8.4	(4.98,14.11)	<.0001		
	SURGERY_TYPE	0.882	2.4	(1.33, 4.37)	0.0036		
	DIABETES	0.677	2	(1.10, 3.54)	0.0234		
A2	Intercept	-3.668	-		<.0001	0.777	430.16
	MINP_thrshld	2.142	8.5	(5.03, 14.41)	<.0001		
	SURGERY_TYPE	0.930	2.5	(1.40, 4.60)	0.0022		
	DIABETES	0.726	2.1	(1.15, 3.71)	0.0151		
A3	Intercept	-3.799	-	-	<.0001	0.785	431.50
	MINP_thrshld	2.072	7.9	(4.59, 13.76)	<.0001		
	SURGERY_TYPE	0.902	2.5	(1.35, 4.49)	0.0033		
	DIABETES	0.751	2.1	(1.18, 3.82)	0.0124		
	AGE_thrshld	0.245	1.3	(0.70, 2.32)	0.4217		
A4	Intercept	-3.659	-	-	<.0001	0.777	431.67
	MINP_thrshld	2.119	8.3	(4.90, 14.14)	<.0001		
	SURGERY_TYPE	0.921	2.5	(1.38, 4.57)	0.0025		
	DIABETES	0.670	2	(1.06, 3.60)	0.0315		
	HIGH_CREAT	0.389	1.5	(0.50, 4.32)	0.4774		
A5	Intercept	-3.7886	-	-	<.0001	0.786	430.14
	MINP_thrshld	2.0922	8.1	(4.76, 13.78)	<.0001		
	SURGERY_TYPE	0.8887	2.4	(1.33, 4.43)	0.0037		
	DIABETES	0.6492	1.9	(1.05, 3.48)	0.0329		
	Hx_IHD	0.3781	1.5	(0.87, 2.46)	0.1551		

*ROC_thrshld=indicator variable of NP thresholds determined by the ROC curve method [1]; MINP_thrshld=indicator variable of NP thresholds determined by minimum p-value method; SURGERY_TYPE=type of surgery; DIABETES=whether or not patient has diabetes mellitus; AGE_thrshld=if an individual is over 65 years old; HIGH_CREAT= history of renal failure; Hx_IHD= history of coronary artery disease;
**OR= Odds Ratio; CI=confidence interval; AUC=Area under the ROC curve; AIC=Akaike Information Criteria; P.Chi=Pearson Chi Square; df=degrees of freedom
***Simple logistic regression was performed to create these models.

Table 4.6.3: Logistic Regression Analysis for MACE

Type of Analysis			Final Risk Factors	Details		
				OR	95% CI	P-Value
Primary Analysis	**MODEL A2**	**SLR**	MINP_thrshld	8.5	(5.03,14.41)	<0.0001
			SURGERY_TYPE	2.5	(1.40, 4.60)	0.0022
			DIABETES	2.1	(1.15, 3.71)	0.0151
Internal Validation Analysis	**MODEL F**	**SLR** (On bootstrap samples)	MINP_thrshld	8.6	(4.95, 14.65)	<0.0001
			SURGERY_TYPE	2.6	(1.37, 4.68)	0.004
			DIABETES	2.1	(1.11, 3.84)	0.023
Sensitivity Analysis	**MODEL B**	**MELR**	MINP_thrshld	10.0	(5.59, 18.06)	<0.0001
			SURGERY_TYPE	2.8	(1.51, 5.31)	0.0012
			DIABETES	1.6	(0.83, 2.96)	0.17
	MODEL C	**GEE**	MINP_thrshld	9.4	(3.81, 23.33)	<.0001
			SURGERY_TYPE	2.7	(1.27, 5.73)	0.0102
			DIABETES	1.8	(0.95, 3.46)	0.0721

Figure 4.6.4: Forest Plot of MACE outcome

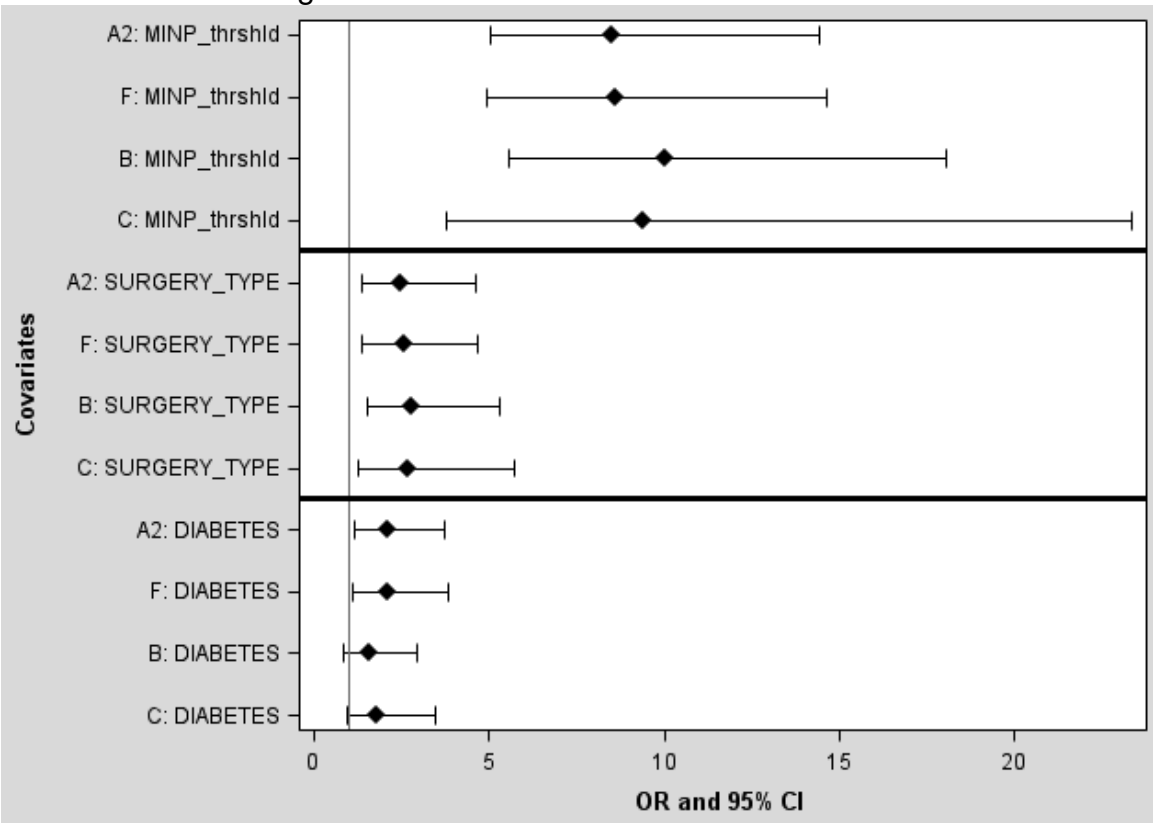


Table 4.7.1: Area Under the ROC Curve for MACE

Type of Analysis	Model	Type of Regression	AUC
Primary Analysis	**A2**	**SLR**	0.777
Validation Analysis	**F**	**SLR** (On bootstrap samples)	0.793
Sensitivity Analysis	**B**	**MELR**	0.776
	C	**GEE**	N/A

*SLR=simple logistic regression; MELR=mixed effects logistic regression; GEE= generalized estimating equations; ROC= receiver operating curve; AUC=area under the curve; MACE= major adverse cardiac events

Table 4.9.1: Regression Coefficients of Final Model (A2)

Risk Factor	Regression Coefficient (β_i)
Intercept	-3.6682
MINP_thrshld	2.1415
SURGERY_TYPE	0.9303
DIABETES	0.7262

Table 4.9.2: Developing a Point System

Risk factor	Categories	β_i	W_{ii}	$\beta_i(W_{ij}-W_{iREF})$	$Points_{ij} = \frac{\beta_i(W_{ij}-W_{iREF})}{B}$
MINP_thrshld	**< NP thresholds**	2.1415	**0=(W_{1REF})**	0	0
	≥ NP thresholds		1	2.1415	3
SURGERY_TYPE	**Infrainguinal**	0.9303	**0=(W_{2REF})**	0	0
	Aortoiliac		1	0.9303	1
DIABETES	**No**	0.7262	**0=(W_{3REF})**	0	0
	Yes		1	0.7262	1

*NP thresholds are: BNP= 115.57pg/mL, NTproBNP=241.7pg/mL; SURGERY_TYPE=type of noncardiac vascular surgery; W_{ij} = Reference value for i^{th} risk factor and j^{th} category;
**The bolded categories are the base categories for each risk factor; β_i is the regression coefficient corresponding to the risk factor; B=0.7262

Table 4.9.3: Point Total and Respective Estimate of Risk

Point Total	Estimate of Risk
0	0.0249
1	0.0501
2	0.0983
3	0.184
4	0.318
5	0.491

Appendix F

Logistic Regression Analysis of Secondary Outcomes

Table 4.10.1: Analysis of All-Cause Mortality, Cardiac Death, and Non-Fatal MI

MODEL	Outcome	Final Risk Factors	Development Model			
			OR	95% CI	P-Value	AUC
G	**All-Cause Mortality** MELR	MINP_thrshld	6.7	(2.76, 16.10)	<0.0001	0.714
		SURGERY_TYPE	2.8	(1.16, 6.79)	0.0218	
H	**Cardiac Death** MELR	MINP_thrshld	9.2	(3.10, 27.37)	<0.0001	0.750
		SURGERY_TYPE	2.6	(0.91, 7.64)	0.0752	
I	**Nonfatal MI** MELR	MINP_thrshld	8.7	(4.60, 16.33)	<0.0001	0.787
		SURGERY_TYPE	2.1	(1.06, 4.17)	0.0338	

Figure 4.10.1: Forest Plot of All-Cause Mortality, Cardiac Death, And Non-Fatal MI

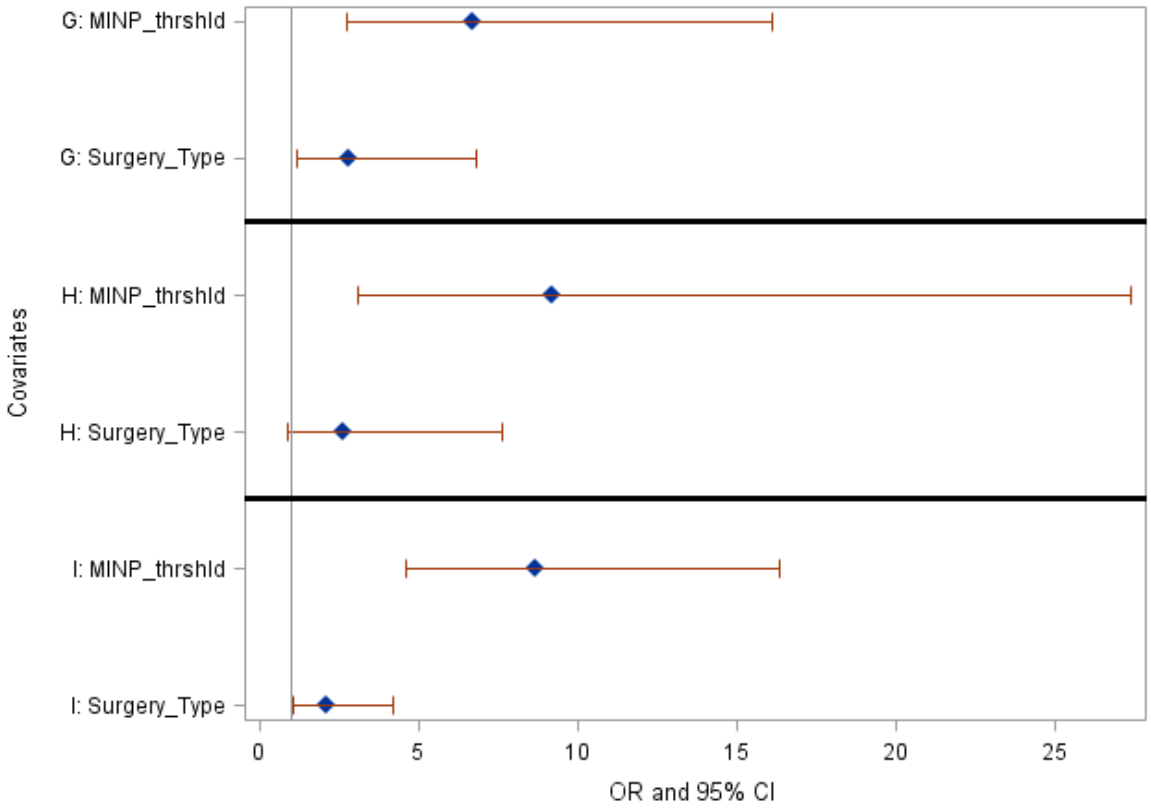


Table 4.10.2: Summary of All-Cause Mortality, Cardiac Death and Non-Fatal MI

		MINP_thrshld		
		0	**1**	**Total**
All-Cause Mortality	**0**	642	173	**815**
	1	14	14	**28**
	Total	**656**	**187**	**843**
Cardiac Death	**0**	649	176	**825**
	1	7	11	**18**
	Total	**656**	**187**	**843**
Nonfatal MI	**0**	636	152	**788**
	1	20	35	**55**
	Total	**656**	**187**	**843**

Table 4.10.3: Sensitivity, Specificity, and Accuracy of MINP_thrshld for Secondary Outcomes

	All-Cause Mortality	**Cardiac Death**	**Nonfatal MI**
Sensitivity	0.50	0.61	0.64
Specificity	0.79	0.79	0.81
Accuracy	0.78	0.78	0.80

Appendix G

RStudio Coding for the

Minimum P-Value Method

```
setwd("/Users/User1/Thabane projects/Thesis Data analysis/datasets")
library("ggplot2")
library(gdata)
library(MASS)
data<-read.xls("Preop BNP data set FINAL.xls")
bnp<-read.xls("BNP data 22FEb2012.xls")  ##subset of bnp data
nt<-read.xls("NTproBNP data 22FEb2012.xls")   ##subset of ntprobnp data
colnames(data)
count(as.numeric(data$BNP_NTproBNP)) #Frequency of BNP and NTproBNP
nrow(data) ##check number of observations in whole dataset
nrow(bnp) ##check number of observations in BNP dataset
nrow(nt) ##check number of observations in NTproBNP dataset

#############################################################################
#PART 1: EXPLORATORY ANALYSIS of pre-op BNP and NTproBNP concentration
levels
#############################################################################
#PRELIMINARY PLOTS
attach(nt);
nt2<-nt[order(NP_Value),] #order data by NP_Values
detach(nt)

attach(bnp);
```

```
bnp2<-bnp[order(NP_Value),] #order data by NP_Values
detach(bnp)

#BNP
par(mfrow=c(1,2))
box_bnp<-boxplot(bnp$NP_Value,xlab="BNP",main="BNP With Outliers", ylab="BNP
Concentration levels (pg/mL");
box_bnp #with outliers- look at output $out
box_bnp$out <- NULL
box_bnp$group <- NULL
bxp(box_bnp,xlab="BNP", main="Outliers Removed", ylab="BNP Concentration levels
(pg/mL")  #plot without outliers
#-->68 outliers, values >=161.25

#NTPROBNP
box_nt<-boxplot(nt$NP_Value,xlab="NTproBNP",main="NTproBNP With Outliers",
ylab="NTproBNP Concentration levels (pg/mL"); box_nt
box_nt$out <- NULL
box_nt$group <- NULL
bxp(box_nt,xlab="NTproBNP",main="Outliers Removed", ylab="NTproBNP
Concentration levels (pg/mL")
#-->20 outliers, values >=1522 are outliers
```

```
par(mfrow=c(3,1))
plot(bnp2$NP_Value,col="blue", ylab="BNP Levels (pg/ml)",
ylim=c(0,4200),main="Observed BNP Levels")
plot(nt2$NP_Value,col="purple", ylab= "NTproBNP Levels (pg/ml)", main="Observed
NTproBNP Levels") #with outlier
plot(nt2$NP_Value,col="purple", ylab= "NTproBNP Levels (pg/ml)",ylim=c(0,7000),
main="Observed NTproBNP Levels, Outlier removed") #without outlier

#Looking for any patterns between (MACE vs bnp_NP_Value) & (MACE vs
nt_NP_Value)
par(mfrow=c(1,1))
with(bnp, plot(NP_Value, MACE, main="Np Values of BNP vs. MACE",xlab="Np
Value", col="blue"))
with(nt, plot(NP_Value, MACE,main="Np values of NTproBNP vs. MACE", xlab="Np
Value", col="purple"))


#########################################################################
#PART 2: MINIMUM P-VALUE METHOD
#########################################################################

#MINP
```

```
MINP_int <- function(x0, x, ybin) {

  if (all(x<=x0) || all(x>x0)) {
    return(c(x0,NA,NA,NA))
  }
  tmp <- suppressWarnings(chisq.test(as.numeric(x<=x0), ybin)) ##p-value is used as a
criterion for this
              ##analysis (corresponds to the maximum chi-square), it will not be used as a
probability of the Type I Error so the warnings can be ignored, thus SuppressWarnings
will get rid of them.
  tab1<-table (as.numeric(x>x0), ybin)   #organizes into table with x>x0 vs ybin;  x0 is
the cutpoint being tested; its value is read from the list given as the 1st argument to
sapply
  tabc <- tab1 + 0.5   #One-way tabulation with automatic bar chart
  rr <- (tabc[1,1]/sum(tabc[,1]))/(tabc[1,2]/sum(tabc[,2]))
  cbind(x0, tmp$statistic, tmp$p.value, rr) #tmp$p.value changes everything else to an
exponential value unneccesarily
  }

MINP<-function (x, ybin, xcutint) {
  ##sapply is a looping function that applies the function given as its
  ##2nd argument repeatedly to each element in the list given as its 1st argument
  tmp1<-sapply (sort(unique(xcutint)), MINP_int, x, ybin)
```

```
  ####sapply is a looping function to repeat MINP_int for each uniqe(repeats removed)
potential cutpoint:
  ###ie. MINP_int(unique xcutint, x,ybin) = (unique xcutint, NP_Value, MACE)
  tmp1<-data.frame(t(tmp1))
  ##transpose to get a column instead of a row matrix
  names (tmp1)<-c("Cutpoint", "Chisquare", "pvalue", "Relrisk")
  tmp1
  }



##############################################################################
#PART 3: ADJUSTMENT FORMULAS OF MINIMUM P-VALUES with 3 functions:
PADJMS, PALT510, PMODBONF.
##############################################################################
#Function Performed
#Computes the adjusted minimum p-value formulae derived by
#Miller and Siegmund, Altman, and Lausen and Schumaker (Section 2.2).

##Description of Input variables
#Cut.point: "(scalar) the Cutpoint associated with the minimum pvalue;"
#pmin: "(scalar) the minimum pvalue;"
#pvalue:   "output vector from MINP; "
```

```
#epsi.high:"proportion of observed values of factor x that are at or below the highest
cutpoint value tested;"
#epsi.low: "proportion of observed values of factor x that are below the lowest cutpoint
value tested; "
#x:        "vector of observed values of continuous prognostic factor."
#pms.palt5, palt10, pmodbon = "the adjusted minimum p-values."


#PADJMS(Cutpoint, pvalue, epsi.high, epsi.low)
PADJMS<-function(Cutpoint, pvalue, epsi.high, epsi.low)
{
    pmin<-min(pvalue)
    Cut.point<-Cutpoint[pvalue == min (pvalue)]
    z<- -qnorm(pmin/2)
    f.z. <- (dnorm(z) )
    pacor1<- (z-1/z) * log(((epsi.high * (1-epsi.low))/(( 1-epsi.high) * epsi.low))) + (4 *
f.z.)/z
    pacor<-(f.z.)*pacor1
    pval<-c(Cut.point, pmin, ## round(pmin, 6),
          epsi.high, epsi.low, pacor) ## round (pacor, 6))
    names(pval)<-c("Cut.point", "p-min", "epsi.high", "epsi.low", "pms")
    pval
}
```

```
#PALT510(Cutpoint, pvalue )
PALT510<-function(Cutpoint, pvalue)
{
    pmin<-min(pvalue)
    Cut.point<-Cutpoint [pvalue == min(pvalue)]
    pcor10<- -1.63 * pmin * (1+2.35*log(pmin))
    pcor5<- -3.13 * pmin *(1+1.65 * log(pmin))
    pval<-c(Cut.point, pmin, pcor5, pcor10)
    names(pval)<-c("Cut.point", "p-min" , "palt5", "palt10")
    pval
}

#PMODBONF(x, Cutpoint, pvalue)
PMODBONF<-function(x, Cutpoint, pvalue)
{
    pmin<-min(pvalue)
    Cut.point<-Cutpoint[pvalue == min(pvalue)]
    z<- -qnorm(pmin/2)
    f.z.<- dnorm(z)
    n<-length(x)
    dsum<-0
```

```
    for(i in 1:(length(Cutpoint)-1)) {
        eps0<-mean(x<=Cutpoint[i])  ##proportion of bnp values less than or equal to
ith cutpoint
        eps1<-mean(x<=Cutpoint[i+1])  ##proportion
        a<-sqrt(1-(eps0*(1-eps1))/((1-eps0)*eps1))
        d<-(exp(-z^2/2)/pi)*f.z.*(a-(z^2/4-1)*a^3/6)
        dsum<-dsum + d
    }
    pmodbonf<-(pmin + dsum)
    pval<-c(Cut.point, pmin,round(pmodbonf,45))
    names(pval)<-c("Cut.point", "p-min" , "pmodbonf")
    round(pval,15)
}

#############################################################################
PART 4a: MINIMUM P-VALUE METHOD for Pre-Op BNP
#############################################################################
cutpts<-with(bnp,NP_Value[NP_Value>=2 & NP_Value<376])
pvalues<-with(bnp,MINP(NP_Value,MACE,cutpts))
pvalues[pvalues$pvalue==min(pvalues$pvalue),]
```

```
#GRAPH OF POTENTIAL CUTPOINTS AND PVALUES
par(mfrow=c(3,1))
with(pvalues,plot(pvalue~Cutpoint,xlim=c(0,3000),main="BNP: Potential cutpoints and corresponding p-values"))
with(pvalues,plot(pvalue~Cutpoint,log="y",xlim=c(0,3000),main="BNP: closer Y-axis scale"))
with(pvalues,plot(pvalue~Cutpoint,log="y",xlim=c(0,600),main="BNP: Zoom on min p-value"))

#COMPARING GRAPHS WITH CHISQUARE STATISTICS (look for MAX) and P-VALUES (look for MIN)
par(mfrow=c(3,1))
with(pvalues,plot(Chisquare~Cutpoint,main="Chisquare Statistic vs Potential Threshold Values"))
with(pvalues,plot(pvalue~Cutpoint,log="y",main="BNP p-values vs. Potential Threshold values"))
with(pvalues,plot(Relrisk~Cutpoint,main="Relative Risk vs. Potential Threshold values"))

#########################################################################
PART 4b: ADJUSTMENT FORMULAS for Pro-op BNP
#########################################################################
#PAJMS
```

```
epsi.high=mean(bnp$NP_Value<=max(cutpts))   #same as writing proportion 630/632
(below max cutpoint)
epsi.low=mean(bnp$NP_Value<min(cutpts))     #same as writing proportion 288/632
(below min cutpoint)
with(pvalues,PADJMS(Cutpoint, pvalue, epsi.high, epsi.low))

#PALT510
with(pvalues,PALT510(Cutpoint,pvalue))

#PMODBONF
PMODBONF(bnp$NP_Value,pvalues$Cutpoint,pvalues$pvalue)

##########################################################################
PART 5a: MINIMUM P-VALUE METHOD for Pre-Op NTproBNP
##########################################################################
cutpts<-with(nt,NP_Value[NP_Value>21.5 & NP_Value<1572]) #creating column of
potential cutpoints to be analyzed
pvalues<-MINP(nt$NP_Value,nt$MACE,cutpts) #p-value is used as a criterion for this
analysis (corresponds to the maximum chi-square), it will not be used a probability of the
Type I Error so the warnings can be ignored.
pvalues[pvalues$pvalue==min(pvalues$pvalue),]
pvalues[pvalues$Relrisk==max(pvalues$Relrisk),]
pvalues[pvalues$Chisquare==max(pvalues$Chisquare),]
```

```
##############################################################################
PART 5b: ADJUSTMENT FORMULAS for Pre-op NTproBNP
##############################################################################
#PADJMS(Cutpoint, pvalue, epsi.high, epsi.low)
epsi.high=mean(nt$NP_Value<=max(cutpts))
epsi.low=mean(nt$NP_Value<min(cutpts))
with(pvalues, PADJMS(Cutpoint, pvalue, epsi.high, epsi.low))

#PALT510(Cutpoint, pvalue )
PALT510(pvalues$Cutpoint,pvalues$pvalue)

#PMODBONF(x, Cutpoint, pvalue)
PMODBONF(nt$NP_Value,pvalues$Cutpoint,pvalues$pvalue)
```

Appendix H

SAS Coding for Logistic Regression and Bootstrap Analysis

```
Proc import out= data datafile= "C:\Documents and Settings\vanniyt\Desktop\Preop BNP
data set FINAL.xls" dbms=xls replace;getnames=yes; run;

*REMOVE STUDY FIVE AND SURGERY_TYPE=2;
data data; set data; if study=5 then delete; run;
data data; set data; if Surgery_type=2 then delete; run;

*CREATE THRESHOLD VARIABLES;
data data;
set data;
        if BNP_NTproBNP=1 and np_value ge 241.7 then MINP_thrshld=1 ;
        else if BNP_NTproBNP=1 and np_value lt 241.7 then MINP_thrshld=0 ;
        else if BNP_NTproBNP=0 and np_value ge 115.57 then MINP_thrshld=1 ;
        else if BNP_NTproBNP=0 and np_value lt 115.57 then MINP_thrshld=0 ;

        if BNP_NTproBNP=1 and np_value ge 277.5 then ROC_thrshld=1 ;
        else if BNP_NTproBNP=1 and np_value lt 277.5 then ROC_thrshld=0 ;
        else if BNP_NTproBNP=0 and np_value ge 116 then ROC_thrshld=1 ;
        else if BNP_NTproBNP=0 and np_value lt 116 then ROC_thrshld=0 ;

        if age ge 65 then age_thrshld=1; else age_thrshld=0;
run;
```

```
/************DETERMINING FINAL MODEL FOR MACE************/
*PRIMARY ANALYSIS;
*MODEL A1: Rodseth's original ;
proc logistic data=data descend;
        model MACE(EVENT='1')=roc_thrshld Surgery_type diabetes/
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;

*MODEL A2;
proc logistic data=data descend;
        model MACE(EVENT='1')=MINP_thrshld Surgery_type diabetes/
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;

*MODEL A3;
Proc logistic data=data descend;
        model MACE(EVENT='1')=MINP_thrshld Surgery_type diabetes age_thrshld/
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;
```

```
*MODEL A4;
proc logistic data=data descend;
        model MACE(EVENT='1')=MINP_thrshld Surgery_type diabetes high_creat/
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;

*MODEL A5;
proc logistic data=data descend;
        model MACE(EVENT='1')=MINP_thrshld Surgery_type diabetes hx_ihd/
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;

/************ VALIDATION ANALYSIS************/
data b1;
        do replicate=1 to 1000;
        do i=1 to nobs;
                x=round(ranuni(023423)*nobs);
                set data
                        nobs = nobs
                        point = x;
                output;
```

```
        end;
        end;
        stop;
run;
proc sort data=b1; by replicate; run;

proc logistic data=b1 descend;
        model MACE(EVENT='1')=MINP_thrshld Surgery_type diabetes /rsq lackfit
        outroc=roc_rocthrshld_lr;
        by replicate;
        ods output ParameterEstimates =estimates oddsratios=or;
        run;

*LOOK AT ESTIMATES;
proc sort data=estimates;by variable; run;
proc means data=estimates noprint;
        var estimate ProbChiSq;
        by variable;
        output out=estimates2; run;

*LOOK AT ORs;
data or_2;
        set or;
```

```
        count + 1;
        by replicate;
        if first.replicate then count=1; run;

proc sort data=or_2; by count; run;
proc means data=or_2 noprint;
        by count;
        output out=or_3; run;

*LOOK AT p-VALUES and 95%CIs ;
*################ MINP_thrshld ###############;
*generate p-values;
data MINP_thrshld;
set estimates;
if variable="MINP_thrshld";
b1=abs(estimate-2.1415);
b2=2.1415;
if b1>b2 then indicator=1;
else indicator=0;
run;
proc means data=MINP_thrshld; var indicator; run; *check the mean of this for p-value;
```

```
*generate st. error and 95%ci;
proc means data=MINP_thrshld; var estimate; run; *take the mean of bootstrap estimates;
data MINP_thrshld;
set MINP_thrshld;
SS=(estimate-2.1535343)*(estimate-2.1535343);
run;
proc means data=MINP_thrshld; var ss; run; *take the mean, we want the sum;
data MINP_thrshld;
set MINP_thrshld;
SE=sqrt((0.0765965*1000)/(1000-1));
lower_ci= 2.1415-1.96*SE;
upper_ci= 2.1415+1.96*SE;
run;

*################ SURGERY_TYPE ###############;
*generate p-values;
data surgery_type;
set estimates;
if variable="Surgery_type";
b1=abs(estimate-0.9303);
b2=0.9303;
if b1>b2 then indicator=1;
else indicator=0;
```

```
run;

proc means data=surgery_type; var indicator; run;

*generate st. error and 95%ci;

proc means data=surgery_type; var estimate; run; *take the mean of bootstrap estimates;

data surgery_type;

set surgery_type;

SS=(estimate-0.9400341)*(estimate-0.9400341);

run;


proc means data=surgery_type; var ss; run; *take the mean, we want the sum;

data surgery_type;

set surgery_type;

SE=sqrt((0.0980561*1000)/(1000-1));

lower_ci= 0.9303-1.96*SE;

upper_ci= 0.9303+1.96*SE;

run;


*################# DIABETES ################;

*generate p-values;

data diabetes;

set estimates;

if variable="Diabetes";

b1=abs(estimate-0.7262);
```

```
b2=0.7262;
if b1>b2 then indicator=1;
else indicator=0;
run;
proc means data=diabetes; var indicator; run;

*generate st. error and 95%ci;
proc means data=diabetes; var estimate; run; *take the mean of bootstrap estimates;
data diabetes;
set diabetes;
SS=(estimate-0.7347928)*(estimate-0.7347928);
run;

proc means data=diabetes; var ss; run; *take the mean, we want the sum;
data diabetes;
set diabetes;
SE=sqrt((0.0997915*1000)/(1000-1));
lower_ci= 0.7262-1.96*SE;
upper_ci= 0.7262+1.96*SE;
run;

/*************** SENSITIVITY ANALYSIS**********/
```

```
*MODEL B - MELR  ;
proc glimmix data=data method=laplace;
        class  study;
        model MACE(event="1")=MINP_thrshld Surgery_type diabetes/
        dist=bin link=logit oddsratio s solution;
        random int/ subject=study ;
        output out=ROC_out_MINP pred=xbeta pred(ilink)=MINP_predprob;
        run;
proc logistic data=ROC_out_MINP plots(only)=roc;
        model MACE(event='1') = MINP_predprob;
        ods select roccurve; run;

*MODEL C - GEE ;
proc genmod data=data descend;
        class study MINP_thrshld surgery_type diabetes/param=ref descending ;
        model MACE=MINP_thrshld Surgery_type diabetes  /
         dist=bin link=logit waldci ;
        repeated subject=study / TYPE=EXCH PRINTMLE;
        *TYPE=EXCH option specifies an exchangeable correlation structure ;
        run;

/*************** SECONDARY ANALYSIS ***********/
*MODEL G;
```

```
proc logistic data=data descend;
        model mortality_allcause(EVENT='1')=MINP_thrshld Surgery_type diabetes /
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;


*MODEL H;
proc logistic data=data descend;
        model mortality_cardiac(EVENT='1')=MINP_thrshld Surgery_type diabetes /
        rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;
*MODEL I;
proc logistic data=data descend;
        model non_fatal_cardiac_event(EVENT='1')=MINP_thrshld Surgery_type
        diabetes/rsq lackfit outroc=roc_rocthrshld_lr;
        ods output ParameterEstimates = model_roc_thrshld;
        run;
```