Nordsjællands Hospital

Date: August 29th 2024

Dysglycemia and endotheliopathy in intensive care unit patients: statistical analysis plan

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Background

Dysglycemia, i.e. hypoglycemia, hyperglycemia, and high glycemic variability, during intensive care unit (ICU) admission has previously been associated with increased morbidity and mortality[1–4] and, likewise, endotheliopathy predicts poor outcome for critically ill patients[5–7]. Glycocalyx degradation and endotheliopathy has also shown to be present in patients with poorly managed diabetes and in healthy persons with acute hyperglycemia[8–12]. The purpose of this study is, therefore, to investigate if changes in biomarkers of the glycocalyx and endothelium are present in relation to dysglycemia in critically ill patients with and without diabetes, as a marker of endotheliopathy.

Hypothesis

We hypothesize that dysglycemia during in-hospital critical illness is associated with an increase in endothelial biomarkers as a sign of endotheliopathy. Further, we hypothesize that a time-wise relation exists depicting the association between dysglycemia and the subsequent damage to the endothelium.

Study population

Between November 2016 and June 2019, 577 ICU patients were included in a prospective cohort study, the Metabolomics cohort, at the general ICU at Copenhagen University Hospital – North Zealand, Denmark. This cohort is the basis for this study and is described in detail elsewhere[13,14].

Definitions

- Mean glucose is the mean of the glucose measurements for the individual patient.
- Glycemic variability (GV) will be defined as both the standard deviation (SD) of the glucose distribution and the coefficient of variation (CV). CV is the SD divided by the mean.
- Hypoglycemia will be defined as either level 1 (<3.9 mmol/L (<70mg/dL) and ≥3.0 mmol/L (54mg/dL) or level 2 (<3.0 mmol/L (54mg/dL) and number of hypoglycemic episodes for each

patient will be summed. We define an individual hypoglycemic event as being at least 30 minutes apart.

- Hyperglycemia will be defined as either level 1 hyperglycemia (>10.0 mmol/L (180mg/dL)) or level 2 hyperglycemia (>13.9 mmol/L (250mg/dL)). To quantify the degree of hyperglycemia, the percentage of glucose measurements for both level 1 and 2 hyperglycemia was estimated for each patient.
- Glycemic gap is defined as the difference between admission glucose and HbA1c-derived average glucose.
- Endothelial biomarkers used in this study are: Syndecan-1, Platelet Endothelial Cell Adhesion Molecule-1 (PECAM-1) and soluble Thrombomodulin (sTM).

Analysis 1

Aim: To determine the associations between glycemic parameters described in 'Definitions' and levels of endothelial markers during the first three days of ICU admission.

Outcome: Associations between predictors: mean glucose, glycemic variability (SD and CV), hypoglycemia, number of hypoglycemic episodes and percentage of time in level 1 and 2 hyperglycemia and the outcome endothelial biomarkers.

Statistical analysis: Linear mixed models will be used to investigate the association between glycemic parameters and endothelial biomarkers. Primary analysis will investigate the associations between mean glucose, glycemic variability (SD and CV), hypoglycemia (yes/no), number of hypoglycemic episodes and percentage of time in level 1 and 2 hyperglycemia and the endothelial biomarkers. Further analysis using other glycemic parameters may be conducted and considered exploratory. A timewise coupling will be made so that up to 24 hours of glycemia will be connected to the subsequent level of endotheliopathy for three consecutive days. Potential confounders will be identified using a Directed Acyclic Graph (DAG). We will model identified confounders as fixed effects and test for interactions. Prespecified fixed effects are haemoglobin A1c (HbA1c), inflammation (C-reactive protein), glucocorticoid administration ICU during admission, Simplified Acute Physiology Score (SAPS) and baseline level of endothelial marker. Subject will be modelled as a random effect. Assumptions will be checked visually by plotting of residuals and, if needed, modelled to fit presumably with logarithm transformation. Missing data will be imputed using multiple imputation[15]. Results of both the univariate and multivariate analysis will be reported.

Analysis 2

Aim: To determine the association between admission glucose levels, glycemic gap, HbA1c, diabetes status (yes/no), and baseline levels of endothelial markers

Outcome: Associations between baseline glucose level, glycemic gap, HbA1c, diabetes status, and baseline levels of endothelial markers.

Statistical analysis: Associations between baseline glucose level, glycemic variability and baseline levels of endothelial markers will be investigated using linear regression models. Confounders will be identified using DAG. Assumptions will be checked, modelled to fit, and we will test for interactions. Assumptions will likewise be checked and modelled to fit again presumably with logarithm transformation. Missing data will be imputed as described in Analysis 1. Results of both the univariate and multivariate analysis will be reported.

Level of statistical significance will be defined as two-sided P-value <0.05 or 95% confidence interval (CI). Analyses will be conducted using RStudio version 1.2.1335 or newer versions (Rstudio Inc.)[16].

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