# Statistical analysis plan, STEREO

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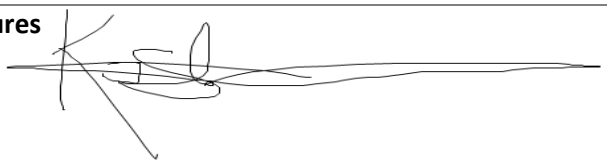
# Statistical analysis plan, STEREO

## Administrative information

| | |
|---|---|
| **Title and trial registration** | **Preoperative high dose steroids for liver resection- effect on complications in the immediate postoperative period- a randomised, double blind, controlled trial (STEREO)**<br>**NCT03403517, EudraCT 2017-002652-81** |
| **SAP version** | 1, 30.09.2020 |
| **Protocol version** | 9, 21.11.2019 |
| **Roles and responsibility** | Kristin J Steinthorsdottir (primary investigator, SAP creator)<br>Eske K Aasvang (Chief investigator/sponsor, final approval)<br>Theis Lange, biostatistician, consultation |
| **Date of approval** | 30-09-2020 |
| **Signatures** |  |

## Introduction

### Background and rationale

Preoperative use of glucocorticoids has been shown to reduce markers of systemic inflammation (IL-6, IL-10, TNF-α, CRP) after several procedures. In studies of preoperative glucocorticoid use in liver resections, markers of liver function (alanine amine transferase (ALT), prothrombin time/international normalized ratio (PT/INR) and total bilirubin) have also been positively affected, as a surrogate of decreased inflammatory response and liver dysfunction[1–4]. Few studies have also shown a clinical benefit, with reduced hospital length of stay[3,4] and the Enhanced Recovery After Surgery (ERAS) Society recommendations include preoperative glucocorticoids with moderate level of evidence to decrease liver injury and intraoperative stress[5]. However, meta-analyses of available RCTs have not shown an effect on morbidity or hospital length of stay[6,7]. Thus, the clinical impact during hospitalization is not clear, and whether high dose steroids influence clinically relevant complications altogether, and especially in the immediate postoperative phase has not been investigated.

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The most common postoperative complications in the immediate phase (in the post-anaesthesia care unit (PACU)) are pain, postoperative nausea and vomiting (PONV) and depending on extent of surgery and the surgical population; circulatory and respiratory complications and cognitive disturbances (emergence delirium and postoperative delirium)[8] After open liver resection surgery, the most common complications in the PACU are pain (including right shoulder pain *[reference theis studie]*), low oxygen saturation and hypotension, with around 40% of patients requiring treatment during PACU stay. All complications are caused by numerous factors, but in general controlling the inflammatory response and reducing the surgical stress response should theoretically reduce complication rates.

### Objectives- main study

The objective is to determine whether a high dose of glucocorticoids can reduce complications in the immediate postoperative phase after open liver resection, compared to a standard, low dose of glucocorticoid

### Objectives- sub study I (endothelial markers)

The objective of the endothelial sub study is to determine whether a single preoperative high dose of glucocorticoids reduce shedding of endothelial glycocalyx markers in the first 72 hours after liver resection, compared to a standard, low dose of glucocorticoids.

## Study methods

### Trial design

Double-blind, parallel group, 1:1 randomisation to either dexamethasone 8 mg or methylprednisolone 10 mg/kg

### Randomisation

Randomisation at the day of operation. Patients are stratified according to planned extent of operation (minor/major resection (<3/ ≥3 liver segments resected)). This stratification is only used for sub study I (endothelial markers). Only patients that are stratified to major resection, that are also operated accordingly, participate in sub study I.

### Sample size

174 patients, approximately 87 in each group. An estimated 20% are part of sub study I. The sample size assumes reducing the proportion of patients with complications requiring treatment in the PACU from 40% to 20%, with an estimated 10% drop-out rate, a statistical power (superiority trial) of 80% with an α level of 5%.

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## Framework

Superiority.

## Statistical interim analyses and stopping guidance

No planned.

## Timing of final analysis

Thirty days after the last patient is operated and after the final GCP monitoring visit, all analyses of the main and sub study I are performed, with allocation still blinded. The main findings are drafted in an abstract, and all authors approve of the analyses and the interpretation. Only thereafter the allocation is revealed. The main study and sub study I can be analysed at different times, but allocation can only be revealed when all authors have accepted drafts with allocation unknown.

## Timing of outcome assessments

Outcomes are assessed ongoing, up to 30 days after the operation.

# Statistical principles

## Confidence intervals and P values

Level of statistical significance: $P<0.05$. Confidence intervals to be reported: 95%CI.

## Adherence and protocol deviations, analysis populations

All randomized patients are analyzed as intention to treat. Patients that receive the intervention and undergo liver resection will be analyzed as per protocol.

Protocol deviations are recorded and presented if occurring in >5% of participants (deviations from the standard surgical, anesthetic and postoperative protocol, e.g. length of epidural anesthesia, deviations from postoperative analgesic regime).

# Trial population

## Eligibility

Patients aged ≥ 18 years scheduled for open liver resection surgery, including combined resection and ablation procedures, during the study period are eligible. Patients can be included if they speak Danish or

English (or if they have a translator during admission), if they can participate in answering questions about pain/nausea etc., and if they sign an informed consent form.

Exclusion criteria are insulin dependent diabetes mellitus, ongoing (within 10 days) treatment with systemic glucocorticoids or other immunosuppressive medication, pregnancy/breastfeeding, active hepatitis C virus, planned two-stage liver resection, planned combined liver resection and operation on other abdominal organs or herniotomy with implantation of a mesh.

## Recruitment

Information to be included in the CONSORT flow diagram: Screening data; includes all patients eligible to participate (e.g. adults planned for open liver resection in the study period). Reasons for non-inclusion listed. Included patients, all patients signing inclusion forms. Randomized patients; all patients included, that received the intervention. Reasons for non-randomization. Participants in each group. Intention to treat and per-protocol numbers.

## Baseline patient characteristics

List of baseline characteristics to be summarized. Categorical data will be described as numbers (percentage) and continuous data as mean (SD) or median (IQR) depending on distribution.

| | **A (n=86)** | **B (n=88)** | **Total** |
|---|---|---|---|
| **Age** | 64 ± 12 | 65 ± 11 | 65± 12 |
| **Sex (male)** | 56 (65%) | 61 (69%) | 117 (67) |
| **BMI** | 28 ± 6 | 27 ± 5 | 28 ± 5 |
| **Smoking status** | | | |
| Current (number of package years) | 16 (19%), xx (x,x) | 18 (20%), xx (x,x) | |
| Past (number of package years) | 46 (53%), xx (x,x) | 43 (49%), xx (x,x) | |
| Never | 24 (28%) | 27 (31%) | |
| **Alcohol intake** | | | |
| > 7(women)/14(men) units/week, no of units | Xx (x%), xx (x,x) | Xx (x%), xx (x,x) | |
| < 7(women)/14(men) units/week | Xx (x%) | Xx (x%) | |
| No | Xx (x%) | Xx (x%) | |
| **Disease/indication for surgery** | | | |
| - Colorectal liver metastasis | Xx (x%) | Xx (x%) | |
| - Non-colorectal metastasis | | | |
| - Hepatocellular carcinoma | Xx (x%), x (x%) | Xx (x%), x (x%) | |
| - Cholangiocellular carcinoma | Xx (x%) | Xx (x%) | |
| - Benign diseases | | | |
| **Comorbidities** | | | |
| - Hypertension | Xx (x%) | Xx (x%) | |
| - Chronic cardiac disease | Xx (x%) | Xx (x%) | |
| - Atrial fibrillation | Xx (x%) | Xx (x%) | |

| | | | |
|---|---|---|---|
| - Chronic pulmonary disease | Xx (x%) | Xx (x%) | |
| - Non-insulin dependent diabetes mellitus | Xx (x%) | Xx (x%) | |
| - Cirrhosis | Xx (x%) | Xx (x%) | |
| - Hypercholesterolemia | Xx (x%) | Xx (x%) | |
| - Depression | | | |
| - Other | | | |
| **ASA score** | | | |
| **I** | 1 (1) | 2 (2) | 3 (2) |
| **II** | 42 (49) | 41(47) | 83 (48) |
| **III** | 43 (50) | 45(51) | 88 (50) |
| **Prior abdominal operation** | Xx (x%) | Xx (x%) | |
| - Open, liver | Xx (x%) | Xx (x%) | |
| - Laparoscopic, liver | Xx (x%) | Xx (x%) | |
| - Open, other | Xx (x%) | Xx (x%) | |
| - Laparoscopic, other | Xx (x%) | Xx (x%) | |
| **Preoperative blood test results** | Xx (x%) | Xx (x%) | |
| - Haemoglobin (mmol/l) | Xx (x%) | Xx (x%) | |
| - Platelets (E9/l) | Xx (x%) | Xx (x%) | |
| - eGFR (ml/min) | Xx (x%) | Xx (x%) | |
| - Protrombin time (INR) | Xx (x%) | Xx (x%) | |
| - Alanine transaminase (IU/l) | | | |
| - Total bilirubin (mg/dl) | | | |
| **Preoperative stratification** | | | |
| - Minor | Xx (x%) | Xx (x%) | |
| - Major | Xx (x%) | Xx (x%) | |

## Operative characteristics

| | **A (n=86)** | **B (n=88)** |
|---|---|---|
| - | | |
| Liver-first (colorectal liver metastasis) | | |
| **Operation (primary)** | | |
| - Left hepatectomy | Xx (x%) | Xx (x%) |
| - Right hepatectomy | Xx (x%) | Xx (x%) |
| - Resection of two liver segments | Xx (x%) | Xx (x%) |
| - Resection of one liver segment | Xx (x%) | Xx (x%) |
| - Atypical resection | Xx (x%) | Xx (x%) |
| - Radio frequency ablation | Xx (x%) | Xx (x%) |
| **Other operation** | | |
| - Cholecystectomy | Xx (x%) | Xx (x%) |

| | | |
|---|---|---|
| - Radio frequency ablation | Xx (x%) | Xx (x%) |
| - Resection of one liver segment | Xx (x%) | Xx (x%) |
| - Colonic resection | Xx (x%) | Xx (x%) |
| - Hernia repair | Xx (x%) | Xx (x%) |
| - Resection, other | Xx (x%) | Xx (x%) |
| Operative time, min | Xx (xx) | Xx(xx) |
| Abdominal drain | Xx (x%) | Xx (x%) |
| Pringle manoeuvre | Xx (x%) | Xx (x%) |
| - Ischemia time (min) | Xx (x,x) | Xx (x,x) |
| Intraoperative bleeding (ml) | Xx (x,x) | Xx (x,x) |
| Intervention group, methylprednisolone dose | | XX (x,x) |

# Outcome definitions and analysis methods

## Primary outcome

Number of patients with a complication in the postoperative care unit (PACU). Complications are evaluated with the DASAIM discharge score (see appendix 1 in the protocol). Any score > 1 (except > 2 for saturation), on two consecutive measures 30 minutes apart OR relevant treatment (inotropic for hypotension, >2L oxygen supply or other respiratory treatment, frequency regulation/conversion for arrythmia, opioids for pain, antiemetics for nausea).

*Primary comparison will be a Fisher's exact test. Effects will be presented as number (percentage), with risk ratios and risk differences along with 95% CI based on Generalized Linear Model (GLM) with the appropriate link-functions.*

*Presence of individual complications of the composite primary outcome (described according to the DASAIM score, e.g. pain, PONV, respiratory complication, circulatory complication, sedation) will be analysed in the same way, and presented together with the primary outcome.*

## Secondary outcomes

1. Length of stay, PACU and Hospital (measured from time of operation until time of discharge). *Compared between groups using t-test and Mann-Whitney U test with the primary p-value being the one form the Mann-Whitney test.*
2. Mortality, all cause 30-day mortality. *Analysed as the primary outcome.*

3. Pain during movement. Pain scores (Numeric Rating Scale (NRS) 0-10), measured every 60$^{th}$ minute during PACU stay. Depending on length of stay in the PACU, there will be 1 to 8 measures per patient, that will be calculated into an average pain score. *Average pain scores for each patient will be compared between groups using t-test and Mann-Whitney U test with the primary p-value being the one form the Mann-Whitney test.*
4. Number of patients with a complication requiring treatment during the first 24 postoperative hours. Any complication requiring treatment (procedural, medication, alterations in standard care). Defined according to protocol. *Analysed as the primary outcome.*
5. Pain during admission. Reported once daily on a NRS scale (0-10), on postoperative day 0 to 4 (or day 3 if discharged). *Pain scores for each patient (worst and average pain) will be compared between groups using t-test and Mann-Whitney U test with the primary p-value being the one form the Mann-Whitney test.*
6. Analgesic medication, rescue opioids during admission (postoperative day 0 to 4 (or day 3 if discharged). Converted to oral morphine equivalents (OMEQ) according to a standardized opioid conversion chart. *Cumulated OMEQs will be compared and analysed as the primary outcome.*
7. Nausea during admission. Reported once daily on a 4-NRS scale (no, light, moderate or severe nausea), postoperative day 0 to 4 (or day 3 if discharged). *Compared between groups using t-test and Mann-Whitney U test with the primary p-value being the one form the Mann-Whitney test.*
8. Blood level of liver enzymes and function tests (ALT, INR, total bilirubin) at baseline (preoperative evaluation) and on postoperative days 1 (all patients), 2 and 3 (major resections). *Postoperative changes from baseline will be compared between groups using t-test and Mann-Whitney U test with the primary p-value being the one form the Mann-Whitney test.*
9. Morbidity, 30 days (liver failure, ascites, intraabdominal fluid collection, bleeding, cholascos, mechanical ileus, perforated visceral organ, fascial disruption, other causes for reoperation, pleural effusion, lung embolus, deep venous embolus, acute myocardial infarction, transitory cerebral ischemia, apoplexy, infections (pneumonia, UTI, sepsis, wound infection), other causes for prolonged hospitalisation). *Total morbidity and Individual complication rates will be analysed as the primary outcome.*

## Endothelial markers (sub study I)

Endothelial markers: syndecan-1, soluble thrombomodulin, SE-selectin, vascular endothelial growth factor and pecam.

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Blood samples are drawn preoperatively (baseline, T0), at the end of the operation (T1), 2 hours after the end of operation (T2), and once daily day 1-3 (T3,4,5). Analysis will be performed on T0, T2, T3 and T5.

Data will be evaluated for normal distribution, log transformed if required, before analysis. Postoperative changes from baseline will be compared with paired samples t-test. The overall development in endothelial markers will be assessed using a two-way mixed MANOVA with group as between-subjects factor and time as within-subjects factor.

## Missing data

For each analysis available data will be presented. No imputation of missing data will occur.

## Harms

Adverse events (AE) is any event occurring within 40 hours from administration of the trial drug, except events not requiring any treatment/actions (e.g. deviant laboratory results), moderate hypotension (MAP> 60 mmHg) and/or tachycardia (heartrate < 120 bpm) in the first 24 postoperative hours, pain from the operative area, urinary retention, intraoperative bleeding, affected motor function during epidural treatment. All AEs are evaluated by the primary investigator as to whether they are related, likely related, unlikely related or unrelated to the trial drug and graded from 1 to 4 (1 = mild, 2 = moderate, 3 = severe, 4 = life-threatening, disabling or deadly). All serious AEs (grade3/4) are reported and analysed for each group.

## Statistical software

R Studio for Windows, newest version available will be used for the statistical analysis. GraphPad Prism for Windows version 8.0.2 will be used for graphical presentation.


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