This supplement contains the following items:

1. Summary of changes.
2. Original version of the trial protocol, including the statistical analysis plan.
3. Final version of the trial protocol, including the statistical analysis plan.

1. Summary of changes.

Protocol version 3.0 is the first version of the protocol approved by the Medical Ethical Committee VUmc (before recruitment of the first patient).

Protocol version 4 describes the addition of a new participating center (Slotervaart hospital). Shortly after approval of protocol version 4 this hospital went bankrupt and did not participate.

Protocol version 5 describes the addition of a new participating center (Medical Center Alkmaar, later renamed as Noordwest Ziekenhuisgroep)

Protocol version 6 describes additional investigations in a predefined subgroup with intensive monitoring. We added diaphragm function measurements (using electromyography and ultrasonography) to the subgroup with more intensive monitoring (8.1.4). The results are not reported in the present manuscript.

Protocol version 7 describes the addition of a new participating center (Sint Franciscus Gasthuis & Vlietland).

Protocol version 8 through 10 describe additional procedures for patients who did not recover sufficiently during their hospital stay to understand the information provided with respect to informed consent for the use of data. We added a paragraph to the deferred consent section (11.2.1)

Protocol version 10.0 is the final approved protocol.

2. Original version of the trial protocol, including the statistical analysis plan.

The effects of hyperoxia on organ dysfunction and outcome in critically ill patients with SIRS

PROTOCOL TITLE: The effects of hyperoxia on organ dysfunction and outcome in critically ill patients with SIRS

Protocol ID	
Short title	**Hyperoxia and SIRS**
Version	**3.0**
Date	**December 15, 2014**
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PROTOCOL SIGNATURE SHEET

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LIST OF ABBREVIATIONS AND RELEVANT DEFINITIONS

ABR	**ABR form, General Assessment and Registration form, is the application form that is required for submission to the accredited Ethics Committee (In Dutch, ABR = Algemene Beoordeling en Registratie)**
AE	**Adverse Event**
AR	**Adverse Reaction**
CA	**Competent Authority**
CABG	**Coronary Bypass Graft Operation**
CCMO	**Central Committee on Research Involving Human Subjects; in Dutch: Centrale Commissie Mensgebonden Onderzoek**
CO/CI	**Cardiac output/cardiac index**
CPB	**Cardiopulmonary Bypass**
CV	**Curriculum Vitae**
DSMB	**Data Safety Monitoring Board**
EU	**European Union**
EudraCT	**European drug regulatory affairs Clinical Trials**
FiO2	**Fraction of inspired oxygen**
GCP	**Good Clinical Practice**
IB	**Investigator's Brochure**
IC	**Informed Consent**
IMP	**Investigational Medicinal Product**
IMPD	**Investigational Medicinal Product Dossier**
METC	**Medical research ethics committee (MREC); in Dutch: medisch ethische toetsing commissie (METC)**
PaO2	**Partial Pressure of Oxygen in Arterial Blood**
ROS	**Reactive oxygen species**
(S)AE	**(Serious) Adverse Event**
SDF	**Sidestream Dark Field Imaging**
SPC	**Summary of Product Characteristics (in Dutch: officiële productinfomatie IB1-tekst)**
Sponsor	**The sponsor is the party that commissions the organisation or performance of the research, for example a pharmaceutical company, academic hospital, scientific organisation or investigator. A party that provides funding for a study but does not commission it is not regarded as the sponsor, but referred to as a subsidising party.**
SUSAR	**Suspected Unexpected Serious Adverse Reaction**
SVR(I)	**Systemic vascular resistance (index)**
Wbp	**Personal Data Protection Act (in Dutch: Wet Bescherming Persoonsgevens)**

WMO	**Medical Research Involving Human Subjects Act (in Dutch: Wet Medisch-wetenschappelijk Onderzoek met Mensen**

1. SUMMARY

Rationale:

Contrary to hypoxia, many physicians do not consider hyperoxia harmful for their patients. To prevent hypoxia, superfluous administration of oxygen is common practice, and hyperoxia is seen in many patients, especially on Intensive Care units. However, an increasing number of studies not only confirm the known negative pulmonary effects of chronic oxygen oversupply, but also important and more acute circulatory effects, characterised by decreased cardiac output (CO), increased systemic vascular resistance (SVR), and impaired microvascular perfusion. These phenomena can impair perfusion of organs, which may outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and perturbed organ function. This may for example be responsible for hyperoxia-associated increased infarct size and increased mortality after myocardial infarction and cardiac arrest. The underlying mechanisms are not clarified yet, but probably involve increased oxidative stress with systemic vasoconstriction.

On the other hand, hyperoxia can also induce several favourable effects. The majority of ICU-patients have a systemic inflammatory response syndrome (SIRS) with concomitant vasoplegia due to trauma, sepsis or ischemia/reperfusion injury. Vasoconstriction could benefit these patients with severe SIRS, reducing the need for intravenous volume resuscitation and vasopressor requirements. Furthermore, hyperoxia may exert a preconditioning effect in patients with ischemia/reperfusion injury and prevent new infections due to its antimicrobacterial properties.

Hypothesis: Hyperoxia during SIRS ultimately has unfavourable effects on organ function, especially on a longer term.

Objectives:

1. To study the short- and long-term effect of two different PaO_2 targets on circulatory status, organ dysfunction and outcome.
2. To study underlying mechanisms of hyperoxia by determining differences in oxidative stress response between the hyperoxic and the normoxic patients.

Study design:

Randomized, prospective multicentre clinical trial

Study population:

Patients admitted to the Intensive Care unit with ≥ 2 positive SIRS-criteria and an expected ICU stay of more than 48 hours

Intervention:
We will investigate 2 groups with PaO2 targets both within the range of current practice
Group 1: target PaO_2 120 (105 – 135) mmHg (hyperoxemic)
Group 2: PaO_2 75 (60 – 90) mmHg (normoxemic)

Primary endpoints:
The primary endpoint will be cumulative daily delta SOFA score (CDDS) from day 1 to day 14.

Secondary endpoints:
Total maximum SOFA score, total maximum SOFA score - SOFA score on admission, SOFA rate of decline
Hypoxic events (PaO2 <55 mmHg)
Vasopressor / Inotrope requirements
Renal function, fluid balance
Oxidative stress (F2-isoprostanes)
Clinical endpoints: duration of mechanical ventilation, ventilator-free days, length of stay (in ICU, in hospital) and mortality.
Subgroup: SVRI, CI, EVLW (PiCCO), microcirculatory flow index, perfused vessel density, fluid status by bio-impedance

Feasability endpoint: Time spent in the assigned PaO2 range

Nature and extent of the burden and risks associated with participation, benefit and group relatedness:
The risk and burden for study subjects are small. Placement of central venous catheters and arterial cannulas are part of standard ICU care. Blood sampling is combined with sampling for normal care of patients and will be taken from either the arterial cannula or the central venous catheter. SDF and bio-impedance measurements are non-invasive causing slight discomfort but no pain or risk for patients. The titrated oxygen levels administered to the patients are based on the PaO_2 measured in blood and pulse oximetry, and the oxygen levels are within the range of current practice, therefore we do not expose the patients to additional risk.

2. INTRODUCTION AND RATIONALE

Hyperoxia has been encountered in 44% of the patients requiring ventilatory support in the Intensive Care [1]. However, contrary to hypoxia, many physicians do not consider hyperoxia harmful for their patients. To stay away from hypoxia, superfluous administration of oxygen is common practice. Since the pulse oximeter never indicates more than 100%, physicians are often not aware of the unphysiological high PaO_2 level. Hyperoxic arterial blood gas values do not commonly cause concern, as physicians lower the FiO_2 in only 25% of the observed cases [1].

However, an increasing number of studies not only confirm the well-known negative pulmonary effects of chronic hyperoxia, but also point to more acute circulatory and perfusion effects[2, 3]. In patients with myocardial or cerebral infarction, for example, hyperoxia increases infarct size and mortality [4, 5]. After cardiac arrest, hyperoxia is associated with worse functional outcome and increased mortality.[6]

The underlying mechanisms of hyperoxia's detrimental effects are not clarified. Increased production of reactive oxygen species (ROS), causing oxidative stress, may play a pivotal role[7], although not all study results are unequivocal [8, 9]. Both animal and human studies suggest that oxidative stress induces systemic vasoconstriction, especially in the microcirculation with a loss of functional capillary density and diminished microvascular flow [10]. This in turn augments systemic vascular resistance and impairs cardiac output [11-14]. Impaired effective circulating volume and microvascular tissue perfusion will outweigh marginally higher arterial oxygen content (dissolved oxygen hardly contributes to blood oxygen content). Hence, a loss of organ perfusion and oxygen delivery may occur.

However, hyperoxia can also induce several favourable effects, illustrating the need for more clinical and preclinical studies. In patients with severe systemic inflammatory response syndrome (SIRS) with concomitant vasoplegia hyperoxia-induced vasoconstriction may stabilize hemodynamics and reduce the need for intravenous volume resuscitation and vasopressor treatment. Common causes of SIRS in the ICU are trauma, sepsis and ischemia/reperfusion after cardiac arrest or cardiopulmonary bypass. [15] Pretreatment with hyperoxia induces a low-graded systemic oxidative stress which may exert a preconditioning effect on the ischemia/reperfusion injury. This may, in contrast to hyperoxia during or after the ischemia/reperfusion insult, decrease myocardial damage and other organ injury.[16] The patients with sepsis can also benefit from the potential antimicrobacterial properties of hyperoxia, which may also prevent new infections.[17-19] Furthermore, in patients with haemorrhage, systemic vasoconstriction due to hyperoxia may cause redistribution of blood flow to the vital organs [20] with amelioration of haemorrhagic shock-induced acute kidney injury. [21]

In critically ill patients, a recent retrospective observational study suggested an independent association between both low and high PaO_2 with in-hospital mortality, with the nadir of mortality between the 70 and 160 mmHg[22]. However, such studies are subject to many forms of bias, and another retrospective study did not confirm these results.[23] Clearly, prospective trials are needed to search for the optimal pO2 range. Hence, it is not just the uncertainty of hyperoxia's untoward effects, but also the possibility of some favourable effects that generates the need for prospective studies.

To the best of our knowledge, no prospective clinical studies have shown benefits of supranormal oxygen levels in any subgroup of critically ill patients.
In this study, we will investigate two different oxygenation levels both near to the nadir of mortality as estimated in an earlier retrospective trial, but one being within the natural range and the other in the supranatural range.[22] In critically ill patients with SIRS, we will assess the effect on organ dysfunction and circulatory parameters. We will separately analyze the predefined subgroups sepsis, trauma/hemorrhage and post-resuscitation.

3. OBJECTIVES

3.1 Primary Objective

The primary aim is to determine whether the normoxic target leads to a lower cumulative daily delta SOFA score (CDDS) from day 1 to day 14 as compared to the hyperoxic group. For detailed calculations and justification of the CDDS endpoint, see section 10.2

3.2 Secondary Objective(s)

The secondary aims are to compare total maximum SOFA score (=total of the most deranged score of each organ system during ICU stay), total maximum SOFA score - SOFA score on admission, SOFA rate of decline, hypoxic episodes (PaO_2 <55 mmHg), vasopressor / inotrope requirements (max dose every 24 hours), need for renal replacement therapy and fluid balances (every 24 hours) between the normoxic and the hyperoxic groups.
Furthermore, we will determine whether oxidative stress parameters F_2-isoprostanes (on day 1,2, and 4) and as clinical endpoints: duration of mechanical ventilation, lung injury score, ventilator-free days, length of stay (in ICU, in hospital) and mortality (ICU and hospital) are different between both groups. Interim analyses will take place after inclusion of 100 patients and 250 to detect possible differences in mortality.
The amount, modality and duration of oxygen administered prior to ICU admission will be recorded. This will be treated as a potential effect-modifying variable and reported in the baseline characteristics.

Feasability endpoint:

Time spent in the assigned PaO_2 range

Subgroup

To further investigate the circulatory changes due to differences in oxygen suppletion, we will study additional parameters in a subgroup of 40 patients, which are too time-consuming to be performed in the whole group. We will estimate hemodynamics by PICCO (C.I., SVRI, extravascular lung water), microcirculation by sublingual Sidestream Dark Field imaging, and body fluid status by bio-impedance on day 1, 2 and 4.

4. STUDY DESIGN

Design:	randomized, clinical multicenter trial
Duration:	24 -30 months
Setting:	Intensive Care Units of VU University Medical Center, Academic Medical Center and Tergooiziekenhuizen

5. STUDY POPULATION

5.1 Population (base)

The study population consists of patients admitted to the Intensive Care Units of the VU University Medical Center, Academic Medical Center and Tergooiziekenhuizen

5.2 Inclusion criteria

In order to be eligible to participate in this study, a subject must meet all of the following criteria:

-Age ≥18 years

-≥2 positive SIRS-criteria:

Temperature >38^{o}C or hypothermia <36^{o}C

Heart rate >90 bpm

Respiratory rate >20 /min or pCO2 <32 mmHg (4.3 kPa)

Number of leucocytes >12 x 109/l of <4 x 109/l of >10% bands

-Within 12 hours of admittance to the ICU

-Expected stay of more than 48 hours as estimated by the attending physician

5.3 Exclusion criteria

A potential subject who meets any of the following criteria will be excluded from participation in this study:

-Elective surgery

-Carbon monoxide poisoning

-Cyanide intoxication

-Methemoglobinemia

-Sickle cell anemia

-Severe pulmonary arterial hypertension (WHO class III or IV)

-Known severe ARDS (PaO2/FiO2 ≤100 mmHg and PEEP ≥ 5 H2O)[24]

-Known cardiac right to left shunting

-Pregnancy

-Severe COPD (Gold class III or IV) or other severe chronic pulmonary disease

5.4 Sample size calculation

Power calculations are based on mean delta SOFA scores, which are invariably reflected in CDDS.

The trial is designed to detected a difference of 0.33 standard deviation (SD) on the primary endpoint with 90% power and a 2-sided alpha of 0.05, with 2 interim analyses.

The trial will be stopped if there is evidence for outcome differences in either direction. Alpha spending for the interim analyses is approximated with a Lan-DeMets O'Brien-Fleming spending function. The total sample size thus needed is 385 patients.

6. TREATMENT OF SUBJECTS

6.1 Investigational product/treatment

In this study we will investigate 2 groups with PaO2 targets both within the range of current practice:

Group 1: target PaO_2 120 (105 – 135) mmHg (hyperoxic)

Group 2: target PaO_2 75 (60 – 90) mmHg (normoxic)

The oxygenation goals are the long-term PaO_2 targets for the participant's entire stay in the ICU.

We will not use excessive measures to achieve the PaO2 targets in group 1. For patients with mechanical ventilation excessive measures include FiO2 settings of >0.60 or prone position. For patients who are not intubated excessive measures include non-invasive mechanical ventilation or intubation. For patients who will not be able to achieve the PaO2 targets of their group, PaO2 targets will be determined by the treating physicians feasible with conventional measures not including those mentioned above. Temporary measures to improve oxygenation for planned procedures involving upper airways such as tracheostomy, bronchoscopy etc will follow standard practices of the participating centers. These aberrations from study targets will be limited to the shortest duration possible.

6.2 Use of co-intervention (if applicable)

Not applicable

7. INVESTIGATIONAL PRODUCT

7.1 Name and description of investigational product(s)

Oxygen

7.2 Summary of findings from clinical and non-clinical studies with known and potential risks and benefits

Animal and human studies indicate that hyperoxia (mostly severe hyperoxia with PaO2 > 300 mmHg after a period of ventilation with an FiO2 of 100%) can lead to important circulatory effects. The hypothesis is that hyperoxia causes vasoconstriction in the microcirculation with a loss of functional capillary density and disturbed microvascular flow. [10] Vasoconstriction causes an increase in systemic vascular resistance and a reduction of the cardiac output. [11-14] This loss of perfusion has been suggested to outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and an increase of ischemia/reperfusion injury.

In patients with myocardial or cerebral infarction, hyperoxia increases infarct size and mortality, and in patients after cardiac arrest hyperoxia is associated with worse functional outcome and increased mortality. [4, 5 6]

Up until now, no prospective, randomized controlled studies investigating the effect of hyperoxia in ICU patients have been performed. A recent large retrospective study showed an independent association between both low and high PaO_2 with in-hospital mortality [22]. However, another retrospective study [23] did not find this association.

7.3 Description and justification of route of administration and dosage

Fraction of inspired oxygen (FiO_2) which is applied to the patient by the mechanical ventilator or oxygen suppletion by nasal cannula or oxygen mask.

7.4 Dosages, dosage modifications and method of administration

In this study we will investigate 2 groups with PaO2 targets both within the range of current practice:

Group 1: target PaO_2 at ICU 120 (105 – 135) mm Hg
Group 2: target PaO_2 at ICU 75 (60 – 90) mmHg
The oxygenation goals are the long-term PaO2 target for the participant's entire stay in the ICU.

We will not use excessive measures to achieve the PaO2 targets in group 1. For patients with mechanical ventilation excessive measures include FiO2 settings of >0.60 or prone

position. For patients who are not intubated excessive measures include non-invasive mechanical ventilation or intubation. For patients who will not be able to achieve the PaO2 targets of their group, PaO2 targets will be determined by the treating physicians feasible with conventional measures not including those mentioned above. Temporary measures to improve oxygenation for planned procedures involving upper airways such as tracheostomy, bronchoscopy etc will follow standard practices of the participating centers. These aberrations from study targets will be limited to the shortest duration possible.

7.5 Preparation and labelling of Investigational Medicinal Product

Not applicable

7.6 Drug accountability

Not applicable

8. METHODS

8.1 Study parameters/endpoints

8.1.1 Main study parameter/endpoint

The primary endpoint will be cumulative daily delta SOFA score (CDDS) from day 1 to day 14. For detailed calculations and justification of the CDDS endpoint, see section 10.2

8.1.2 Secondary study parameters/endpoints

Secondary parameters will include total maximum SOFA score(= total of the most deranged score of each organ system during ICU stay), total maximum SOFA score - SOFA score on admission, SOFA rate of decline, hypoxic episodes (PaO_2 <55 mmHg), vasopressor / inotrope requirements (max dose every 24 hours), need for renal replacement therapy and fluid balances (every 24 hours). Furthermore, oxidative stress parameters F_2-isoprostanes will be determined (on day 1, 2 and 4) and as clinical endpoints: duration of mechanical ventilation, ventilator-free days, length of stay (in ICU, in hospital) and mortality (ICU and hospital). Interim analyses will take place after inclusion of 100 patients to detect possible differences in mortality.
The amount, modality and duration of oxygen administered prior to ICU admission will be recorded. This will be treated as a potential effect-modifying variable and reported in the baseline characteristics.

8.1.3 Feasibility endpoint

Time spent in the assigned PaO_2 range

8.1.4 Subgroup

To further investigate the circulatory changes due to differences in oxygen suppletion, we will study additional parameters in a subgroup of patients, which are too time-consuming to be performed in the whole group. We will estimate hemodynamics by PICCO (C.I., SVRI, extravascular lung water), microcirculation by sublingual Sidestream Dark Field imaging, and body fluid status by bio-impedance.

a) Arterial blood gas analysis with PaO_2 and lactate measurement will be determined initially at least every 4 hours, and at least every 6 hours after stabilisation. Data will be collected during the entire stay on the ICU. Area under the curve will be calculated for PaO_2, O_2 saturation, FiO_2 and lactate.

b) Hemodynamics
Whenever patients for more intensive hemodynamic monitoring are provided with continuous cardiac output measurement by Swan Ganz catheter or PiCCO, SVRI, CO and CI and EVLW will be measured every 4 hours. In all patients with a central line in the jugular vein, SvO2 will be determined daily.

c) Sublingual mucosal microcirculation measurements will be performed on day 1, 2, and 4 using sidestream dark field (SDF) imaging and quantified as the level of perfused small vessel density and microvascular flow index (vessel diameter < 20 µm).

d) Bio-impedance measured resistance/reactance/impedance at day 1, 2, and 4.

8.2 Randomisation, blinding and treatment allocation

Randomisation will be performed with the use of randomisation list generated by a web-based computer program

8.3 Study procedures

1) Deferred consent by patient representative
2) Routine clinical data (demographic characteristics, reason of admission, comorbidity, APACHE II)
3) Titration of FiO_2, based on measured PaO_2
   group 1: target PaO_2 at ICU of 120 (105 – 135) mm Hg
   group 2: : target PaO_2 at ICU of 75 (60 – 90) mmHg
4) Blood sample collection at baseline and on day 2, and 4 for determination of parameters of oxidative stress and tissue/organ perfusion *(in total 40 ml extra for study*). Remaining blood material after analysis will be stored for additional analyses in the future.

Substudy:

In a subgroup of 40 patients, hemodynamics will be more intensively monitored by SDF, PiCCO and bio-impedance. These additional measurements will only be done in a subgroup of patients because they are very time consuming and it is logistically impossible to perform them in all the patients.

8.4 Withdrawal of individual subjects

Subjects can leave the study at any time for any reason if they wish to do so without any consequences. The investigator can decide to withdraw a subject from the study for urgent medical reasons.

9. SAFETY REPORTING

9.1 Section 10 WMO event

In accordance to section 10, subsection 1, of the WMO, the investigator will inform the subjects and the reviewing accredited METC if anything occurs, on the basis of which it appears that the disadvantages of participation may be significantly greater than was foreseen in the research proposal. The study will be suspended pending further review by the accredited METC, except insofar as suspension would jeopardise the subjects' health. The investigator will take care that all subjects are kept informed.

9.2 SAEs and SUSARs

This study compares two treatment targets that are used in standard care. All included patients are admitted to the Intensive Care with a life-threatening disease. Any major adverse development can be expected as part of the underlying disease. Therefore, it is not achievable to report all these developments individually as SAEs. Every three months we will send a line-listing of all SAEs to the METc. Furthermore, all deaths will be reported via ToetsingOnline to the METc/CCMO within one week of the event.

9.3 Data Safety Monitoring Board (DSMB) / Safety Committee

Since the titration of FiO_2 based on the measured pO_2 is within the range of standard care, we do not expose the patients to additional risk. Therefore a data safety monitoring board will not be instituted.

10. STATISTICAL ANALYSIS

10.1 Descriptive statistics

Statistical analysis will be performed using the SPSS statistical software package (SPSS Inc.®, Chicago USA). All included patients will be incorporated in the statistical analysis. Descriptive statistics of these quantitative data will include mean, median, standard deviation and interquartile range.

10.2 Primary and secondary study parameter(s)

Calculations of CDDS primary endpoint

The primary endpoint will be *cumulative daily delta SOFA score* (CDDS) from day 1 to day 14, calculated as:

$$CDDS = \sum_{i=day\,2\,to\,14} (SOFA_{DAYi} - SOFA_{ADMISSION})$$

or alternatively,

$$CDDS = SOFA_{DAY\,2} + SOFA_{DAY\,3} + SOFA_{DAY\,4} + ...etc + SOFA_{DAY\,14} - (13 \cdot SOFA_{ADMISSION})$$

Daily SOFA score is calculated as the total of maximum scores for each organ system excluding respiratory system (because of possible PaO_2/FiO_2 distortion). For patients discharged from the ICU, SOFA score will be registered as 0 from the day of discharge to day 14. Death in the ICU will be registered as a score of 20 (maximum) from the day of death to day 14.

CDDS reflects a balanced weighting of the following clinically relevant preconditions:

1. Low (or negative) delta SOFA score is better than high (or positive) delta SOFA score.
2. Shorter duration of ICU stay is better than longer stay.
3. Discharge from the ICU is better than death in the ICU.
4. Eventual discharge after a high admission SOFA score is better than discharge after a low admission SOFA score.
5. Eventual death after a low admission SOFA score is worse than discharge after a high admission SOFA score.
6. Early SOFA score reduction is better than late SOFA score reduction.

Other possible endpoints (such as *delta SOFA score* or *SOFA rate of decline*) fail to satisfy most of these criteria.

The differences of primary and secondary parameters between the 2 groups will be calculated by intention-to-treat analysis. The primary endpoint (CDDS) is designed and

tested as a tool for ranking outcomes. The between-group comparison on the primary endpoint will therefore be made using the Wilcoxon rank-sum test.
Secondary outcomes will be compared using ANOVA for repeated measurements, Chi-Squared, Mann-Whitney U or Fisher's Exact test where appropriate. Mortality will be assessed with the Kaplan Meier and log rank test.

10.3 Interim analysis (if applicable)
Interim analyses (blinded for the intervention arm) will take place after inclusion of 150 and 275 patients to detect possible differences in mortality.

The interim analyses are planned as a symmetric two-sided group sequential design with 90% power and 5% two-sided Type I Error. Spending computations assume the trial stops if a bound is crossed.

Analysis	N	Boundary two-sided P	Spend
1	150	0.0006	0.0006
2	275	0.0156	0.0152
3	385	0.0452	0.0342
total			0.050

++ alpha spending:
Lan-DeMets O'brien-Fleming approximation spending function

For example, the trial will be stopped after 275 patients if there is a sufficient difference in primary endpoint between the two groups, such that P<0.0156.

In addition to the trial stopping boundaries for superior outcomes in one of the trial arms (outlined above), we have defined a futility stopping boundary at interim-analysis 2 (after N=275 patients): The trial will be stopped if there is almost no difference in the primary endpoint between the treatment arms, such that the conditional power is less than 20% at interim-analysis 2. Practically, if P>0.459 after 275 patients, there is a less than 20% probability of obtaining a significant result after 385 patients.

The METc/CRB will be consulted when, at the interim analysis, between-group differences in mortality are large enough so that P<0.05 but not large enough to reach one of the stopping criteria outlined above.

11. ETHICAL CONSIDERATIONS

11.1 Regulation statement

The study will be conducted according to the principles of the Declaration of Helsinki (59th WMA General Assembly, Seoul 2008) and in accordance with the Medical Research Involving Human Act (WMO) and the statements of the CCMO as presented in the publication "Uitgestelde toestemming voor inclusie van beslissingsonbekwame patiënten in studies van spoedeisende geneeskunde" by E.J.O.Kompanje.[25]

11.2 Recruitment and consent

11.2.1. Deferred consent

For this study we ask for deferred consent and we appeal to the emergency procedure for consent in medical research as stated in article 6, paragraph 4 of the WMO because of the following reasons.

Oxygen suppletion is applied to almost all ICU-patients. To alleviate respiratory distress oxygen therapy is initiated and cannot be postponed. Patients admitted to an Intensive Care Unit (ICU) are mostly incompetent to give informed consent. Obtaining informed consent from a legal representative takes time (on average up to 12 hours), even by an experienced research team.[26]

We propose to randomize each patient who meets the inclusion criteria ultimately within 2 hours after ICU admission. Oxygenation targets according to randomization are applied immediately thenceforth. Informed consent from the legal representative will be requested as soon as possible thereafter, but never later than 24 hours after randomization. If informed consent is not obtained within those 24 hours, or if a legal representative denies participation within this time frame, the patient is excluded and data will no longer be used. Thenceforth the patient is oxygenated according to the policy of the attending physician.

11.2.2 Effects of hyperoxia

Hyperoxia can induce more chronic, harmful effects to the lungs, but also more acute circulatory effects, characterised by decreased cardiac output, increased systemic vascular resistance, and impaired microvascular perfusion. These phenomena can impair perfusion of organs, which may outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and perturbed organ function. This may be responsible for hyperoxia-associated increased infarct size and increased mortality after myocardial infarction and cardiac arrest. [2, 3, 6] On the other hand, hyperoxia can also induce several favourable effects. The majority of ICU-patients have a systemic inflammatory response syndrome (SIRS) with concomitant

vasoplegia due to trauma, sepsis or ischemia/reperfusion injury. Vasoconstriction could benefit these patients with severe SIRS, reducing the need for intravenous volume resuscitation and vasopressor requirements. Furthermore, hyperoxia may exert a preconditioning effect in patients with ischemia/reperfusion injury and prevent new infections due to its antimicrobacterial properties.

If a patient, in the proposed trial, is already hyperoxic for several hours, effects of this oxygenation target could already be in place, largely reducing validity of the trial outcomes. From experimental animal studies we know that hyperoxia can cause circulatory changes within an hour.[27, 28] These findings are in line with results from clinical studies, showing effects on cardiac output and systemic vascular resistance even within 10 minutes.[29]

11.2.3. Time between asking for and obtaining informed consent

Most critically ill patients cannot give informed consent for a study at ICU admission. They are in severe respiratory distress, sedated or in coma. Time to obtain informed consent after recognition of study eligibility by a legal representative could take as much as 12 hours, even when a legal representative could be contacted fast. This is in line with a recent observational study performed in the Academic Medical Center, Amsterdam, the Netherlands. Time to obtain informed consent after recognition of study eligibility from a legal representative was as high as 14 hours (study ID NL34294.018.10). For relatives of a severely ill patient who has been acutely admitted to the ICU it is difficult to make a quick, but balanced decision. [26, 30, 31]

Most ICU patients are positive about their enrollment under deferred consent. A contentment questionnaire of the large NICE–SUGAR trial [32], a trial studying different targets in blood glucose control [33], showed that the large majority (96%) would have granted consent if they would have been asked. 93% of the patients were content with the decision made by the representative at the moment they were incapable of giving informed consent. [33]

11.2.4. Ethical aspects

We can underpin the idea of 'clinical equipoise'.[34] The study participant can benefit from the intervention, but up to now there is a state of honest, professional disagreement in the community of expert practitioners as to the optimal oxygenation targets for ICU-patients. Currently, an implementation trial (Oxytar, de Jonge LUMC, NTR 3424) is running, aiming for target PaO_2 values of 55 – 80 mmHg in ICU patients. However, this target is not based on prospective, let alone randomised clinical evidence. Some arguments act in favour of this target oxygen range [35], but others do not. For example, the nadir for unadjusted hospital mortality as observed in a Dutch retrospective study was substantially higher than this target, i.e. just below 150 mmHg [22]. International guidelines for oxygen suppletion in medical

emergencies developed by the British Thoracic Society and endorsed by several other societies, recommend a target peripheral O_2 saturation range measured by pulse oximetry of 94 – 98%. [36] It is difficult to translate this O_2 saturation range to target pO_2 range, since their relation fluctuates dependent on other factors like temperature, perfusion, and pH. However, saturations as recommended will commonly result in PaO_2 in a higher range than 55 – 80 mmHg. More importantly, however, these recommendations are also unsupported by randomized controlled trials. Furthermore, since hyperoxia can induce both negative (pulmonary and circulatory) and positive (antimicrobial, preconditioning) also from a pathophysiological view the optimal target for ICU-patients is unclear. Both oxygenation targets in our trial are within the range used in common practice in ICUs worldwide and are considered safe.

11.2.5. Similarities with previous studies using deferred consent

The proposed trial has many similarities with recently published interventional trials in ICU patients using deferred consent. [32, 37-40] The investigated therapies were: intravenous fluid resuscitation with colloids or crystalloids [37], fluid resuscitation with 4% albumin infusion or normal saline infusion [38], RENAL replacement therapy with a lower or higher intensity [39] and ventilation with lower or higher tidal volumes (PReVent). In these trials, as in our trial, patients were incapable at the moment therapy had to be started, the effects of the interventions were for a substantial part induced on its start, and the strategies under study were both used in daily practice.

11.2.6. Conclusions

Critically ill patients are mostly incapable at the moment of ICU admission. Obtaining informed consent from a legal representative takes mostly half a day. Start of oxygen therapy in ICU-patients cannot wait. Both oxygenation targets are within the range used in common practice in ICUs worldwide and are considered safe.

11.3 Benefits and risks assessment, group relatedness

The risk and burden for study subjects are small. Blood sampling is combined with sampling for normal care of patients. SDF and bio-impedance measurements are non-invasive causing slight discomfort but no pain or risk for patients. Since the titration of FiO_2 based on the measured PaO_2 is within the range of standard care, we do not expose the patients to additional risk.

11.4 Compensation for injury

The sponsor/investigator has a liability insurance which is in accordance with article 7, subsection 6 of the WMO.

The sponsor (also) has an insurance which is in accordance with the legal requirements in the Netherlands (Article 7 WMO and the Measure regarding Compulsory Insurance for Clinical Research in Humans of 23th June 2003). This insurance provides cover for damage to research subjects through injury or death caused by the study.

1. € 450.000,-- (i.e. four hundred and fifty thousand Euro) for death or injury for each subject who participates in the Research;
2. € 3.500.000,-- (i.e. three million five hundred thousand Euro) for death or injury for all subjects who participate in the Research;
3. € 5.000.000,-- (i.e. five million Euro) for the total damage incurred by the organisation for all damage disclosed by scientific research for the Sponsor as 'verrichter' in the meaning of said Act in each year of insurance coverage.

The insurance applies to the damage that becomes apparent during the study or within 4 years after the end of the study.

11.5 Incentives (if applicable)

None

11.6 Monitoring Clinical Research Bureau

An independent monitor (quality officer) will monitor the study data according to the regulations described under Good Clinical Practice (GCP). In a selection of study subjects the Informed Consents are controlled. Additionally, during onsite monitoring the officer will perform a Source Data verification of data described in the Case Report Forms to investigation the agreement between source data and study reports. The intensity of this verification is related to the study risk assessment. In particular, inclusion and exclusion criteria and the primary endpoints of the investigation are subject to monitoring. The monitor will evaluate whether SAE's en SUSAR's are adequately reported within the time frame as directed by the Dutch law.

12. ADMINISTRATIVE ASPECTS, MONITORING AND PUBLICATION

12.1 Handling and storage of data and documents

Patient data will be stored anonymously. Data will not be directly traceable to the individual patients, as all patients are coded. The key to the code will be separately safeguarded by the primary investigator. Data will be stored for 15 years.

12.2 Amendments

Amendments are changes made to the research after a favourable opinion by the accredited METC has been given. All amendments will be notified to the METC that gave a favourable opinion.

A 'substantial amendment' is defined as an amendment to the terms of the METC application, or to the protocol or any other supporting documentation, that is likely to affect to a significant degree:

- the safety or physical or mental integrity of the subjects of the trial;
- the scientific value of the trial;
- the conduct or management of the trial; or
- the quality or safety of any intervention used in the trial.

All substantial amendments will be notified to the METC and to the competent authority. Non-substantial amendments will not be notified to the accredited METC and the competent authority, but will be recorded and filed by the sponsor.

12.3 Annual progress report

The sponsor/investigator will submit a summary of the progress of the trial to the accredited METC once a year. Information will be provided on the date of inclusion of the first subject, numbers of subjects included and numbers of subjects that have completed the trial, serious adverse events/ serious adverse reactions, other problems, and amendments.

12.4 End of study report

The investigator will notify the accredited METC of the end of the study within a period of 8 weeks. The end of the study is defined as the last patient's last visit.
In case the study is ended prematurely, the investigator will notify the accredited METC within 15 days, including the reasons for the premature termination.
Within one year after the end of the study, the investigator/sponsor will submit a final study report with the results of the study, including any publications/abstracts of the study, to the accredited METC.

12.5 Public disclosure and publication policy

We are free to make a publication and have no restrictions made by a sponsor. Patient data will be published anonymously.

13. REFERENCES

Reference List

(1) de Graaff AE, Dongelmans DA, Binnekade JM, de JE. Clinicians' response to hyperoxia in ventilated patients in a Dutch ICU depends on the level of FiO2. *Intensive Care Med* 2011 January;37(1):46-51.

(2) Cornet AD, Kooter AJ, Peters MJ, Smulders YM. Supplemental oxygen therapy in medical emergencies: more harm than benefit? *Arch Intern Med* 2012 February 13;172(3):289-90.

(3) Cornet AD, Kooter AJ, Peters MJ, Smulders YM. The potential harm of oxygen therapy in medical emergencies. *Crit Care* 2013 April 18;17(2):313.

(4) Rawles JM, Kenmure AC. Controlled trial of oxygen in uncomplicated myocardial infarction. *Br Med J* 1976 May 8;1(6018):1121-3.

(5) Ronning OM, Guldvog B. Should stroke victims routinely receive supplemental oxygen? A quasi-randomized controlled trial. *Stroke* 1999 October;30(10):2033-7.

(6) Kilgannon JH, Jones AE, Shapiro NI et al. Association between arterial hyperoxia following resuscitation from cardiac arrest and in-hospital mortality. *JAMA* 2010 June 2;303(21):2165-71.

(7) Jamieson D, Chance B, Cadenas E, Boveris A. The relation of free radical production to hyperoxia. *Annu Rev Physiol* 1986;48:703-19.

(8) Hauser B, Barth E, Bassi G et al. Hemodynamic, metabolic, and organ function effects of pure oxygen ventilation during established fecal peritonitis-induced septic shock. *Crit Care Med* 2009 August;37(8):2465-9.

(9) Barth E, Bassi G, Maybauer DM et al. Effects of ventilation with 100% oxygen during early hyperdynamic porcine fecal peritonitis. *Crit Care Med* 2008 February;36(2):495-503.

(10) Kamler M, Wendt D, Pizanis N, Milekhin V, Schade U, Jakob H. Deleterious effects of oxygen during extracorporeal circulation for the microcirculation in vivo. *Eur J Cardiothorac Surg* 2004 September;26(3):564-70.

(11) Bak Z, Sjoberg F, Rousseau A, Steinvall I, Janerot-Sjoberg B. Human cardiovascular dose-response to supplemental oxygen. *Acta Physiol (Oxf)* 2007 September;191(1):15-24.

(12) Ihnken K, Winkler A, Schlensak C et al. Normoxic cardiopulmonary bypass reduces oxidative myocardial damage and nitric oxide during cardiac operations in the adult. *J Thorac Cardiovasc Surg* 1998 August;116(2):327-34.

(13) Harten JM, Anderson KJ, Kinsella J, Higgins MJ. Normobaric hyperoxia reduces cardiac index in patients after coronary artery bypass surgery. *J Cardiothorac Vasc Anesth* 2005 April;19(2):173-5.

(14) Anderson KJ, Harten JM, Booth MG, Kinsella J. The cardiovascular effects of inspired oxygen fraction in anaesthetized patients. *Eur J Anaesthesiol* 2005 June;22(6):420-5.

(15) Horeczko T, Green JP, Panacek EA. Epidemiology of the Systemic Inflammatory Response Syndrome (SIRS) in the Emergency Department. *West J Emerg Med* 2014 May;15(3):329-36.

(16) Tahep lP, Valen G, Starkopf J, Kairane C, Zilmer M, Vaage J. Pretreating rats with hyperoxia attenuates ischemia-reperfusion injury of the heart. *Life Sci* 2001 February 23;68(14):1629-40.

(17) Calzia E, Asfar P, Hauser B et al. Hyperoxia may be beneficial. *Crit Care Med* 2010 October;38(10 Suppl):S559-S568.

(18) Greif R, Akca O, Horn EP, Kurz A, Sessler DI. Supplemental perioperative oxygen to reduce the incidence of surgical-wound infection. *N Engl J Med* 2000 January 20;342(3):161-7.

(19) Belda FJ, Aguilera L, Garcia de la AJ et al. Supplemental perioperative oxygen and the risk of surgical wound infection: a randomized controlled trial. *JAMA* 2005 October 26;294(16):2035-42.

(20) Bitterman H, Brod V, Weisz G, Kushnir D, Bitterman N. Effects of oxygen on regional hemodynamics in hemorrhagic shock. *Am J Physiol* 1996 July;271(1 Pt 2):H203-H211.

(21) Efrati S, Berman S, Ben AG, Siman-Tov Y, Averbukh Z, Weissgarten J. Application of normobaric hyperoxia therapy for amelioration of haemorrhagic shock-induced acute renal failure. *Nephrol Dial Transplant* 2008 July;23(7):2213-22.

(22) de JE, Peelen L, Keijzers PJ et al. Association between administered oxygen, arterial partial oxygen pressure and mortality in mechanically ventilated intensive care unit patients. *Crit Care* 2008;12(6):R156.

(23) Eastwood G, Bellomo R, Bailey M et al. Arterial oxygen tension and mortality in mechanically ventilated patients. *Intensive Care Med* 2012 January;38(1):91-8.

(24) Steinberg KP, Hudson LD, Goodman RB et al. Efficacy and safety of corticosteroids for persistent acute respiratory distress syndrome. *N Engl J Med* 2006 April 20;354(16):1671-84.

(25) Kompanje EJO, Jansen TC, Le Noble JLML, de Geus HR, Bakker J. [Deferred consent for inclusion of patients unable to give their consent in studies in the field of emergency medicine]. *Ned Tijdschr Geneeskd* 2008 September 20;152(38):2057-61.

(26) Burns KE, Zubrinich C, Tan W et al. Research recruitment practices and critically ill patients. A multicenter, cross-sectional study (the Consent Study). *Am J Respir Crit Care Med* 2013 June 1;187(11):1212-8.

(27) Tsai AG, Cabrales P, Winslow RM, Intaglietta M. Microvascular oxygen distribution in awake hamster window chamber model during hyperoxia. *Am J Physiol Heart Circ Physiol* 2003 October;285(4):H1537-H1545.

(28) Cabrales P, Tsai AG, Intaglietta M. Nitric oxide regulation of microvascular oxygen exchange during hypoxia and hyperoxia. *J Appl Physiol (1985)* 2006 April;100(4):1181-7.

(29) Park JH, Balmain S, Berry C, Morton JJ, McMurray JJ. Potentially detrimental cardiovascular effects of oxygen in patients with chronic left ventricular systolic dysfunction. *Heart* 2010 April;96(7):533-8.

(30) Kompanje EJ, Jansen TC, Le Noble JL, de Geus HR, Bakker J. [Deferred consent for inclusion of patients unable to give their consent in studies in the field of emergency medicine]. *Ned Tijdschr Geneeskd* 2008 September 20;152(38):2057-61.

(31) Jansen TC, Kompanje EJ, Druml C, Menon DK, Wiedermann CJ, Bakker J. Deferred consent in emergency intensive care research: what if the patient dies early? Use the data or not? *Intensive Care Med* 2007 May;33(5):894-900.

(32) Finfer S, Chittock DR, Su SY et al. Intensive versus conventional glucose control in critically ill patients. *N Engl J Med* 2009 March 26;360(13):1283-97.

(33) Potter JE, McKinley S, Delaney A. Research participants' opinions of delayed consent for a randomised controlled trial of glucose control in intensive care. *Intensive Care Med* 2013 March;39(3):472-80.

(34) Freedman B. Equipoise and the ethics of clinical research. *N Engl J Med* 1987 July 16;317(3):141-5.

(35) Ventilation with lower tidal volumes as compared with traditional tidal volumes for acute lung injury and the acute respiratory distress syndrome. The Acute Respiratory Distress Syndrome Network. *N Engl J Med* 2000 May 4;342(18):1301-8.

(36) O'Driscoll BR, Howard LS, Davison AG. BTS guideline for emergency oxygen use in adult patients. *Thorax* 2008 October;63 Suppl 6:vi1-68.

(37) Annane D, Siami S, Jaber S et al. Effects of fluid resuscitation with colloids vs crystalloids on mortality in critically ill patients presenting with hypovolemic shock: the CRISTAL randomized trial. *JAMA* 2013 November 6;310(17):1809-17.

(38) Finfer S, Bellomo R, Boyce N, French J, Myburgh J, Norton R. A comparison of albumin and saline for fluid resuscitation in the intensive care unit. *N Engl J Med* 2004 May 27;350(22):2247-56.

(39) Bellomo R, Cass A, Cole L et al. Intensity of continuous renal-replacement therapy in critically ill patients. *N Engl J Med* 2009 October 22;361(17):1627-38.

(40) Jansen TC, van BJ, Schoonderbeek FJ et al. Early lactate-guided therapy in intensive care unit patients: a multicenter, open-label, randomized controlled trial. *Am J Respir Crit Care Med* 2010 September 15;182(6):752-61.

3. Final version of the trial protocol, including the statistical analysis plan.

The effects of hyperoxia on organ dysfunction and outcome in critically ill patients with SIRS

Implementation title:

O_2-ICU

PROTOCOL TITLE: The effects of hyperoxia on organ dysfunction and outcome in critically ill patients with SIRS

Protocol ID	**NL 50040.029.14**
Short title	**Hyperoxia and SIRS / O_2-ICU**
Version	**10.0**
Date	**August 6, 2018**
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Independent physician	**Prof. dr. N. van Royen (cardiology)**
Laboratory sites	**Clinical Chemical Laboratory, VUmc** **De Boelelaan 1117, 1081 HV Amsterdam** **Laboratory for Physiology** **Van der Boechorststraat 7, 1081 BT Amsterdam**

PROTOCOL SIGNATURE SHEET

Name	Signature	Date
Project leaders Prof. dr. Y.M. Smulders Prof. dr. H. Oudemans – van Straaten **Coordinating investigator** Dr. A.M.E. Spoelstra – de Man		
Principal Investigator: H.J.S. de Grooth		
Head of the Department of Intensive Care Prof. Dr. A.R.J. Girbes		

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LIST OF ABBREVIATIONS AND RELEVANT DEFINITIONS

ABR	**ABR form, General Assessment and Registration form, is the application form that is required for submission to the accredited Ethics Committee (In Dutch, ABR = Algemene Beoordeling en Registratie)**
AE	**Adverse Event**
AR	**Adverse Reaction**
CA	**Competent Authority**
CABG	**Coronary Bypass Graft Operation**
CCMO	**Central Committee on Research Involving Human Subjects; in Dutch: Centrale Commissie Mensgebonden Onderzoek**
CO/CI	**Cardiac output/cardiac index**
CPB	**Cardiopulmonary Bypass**
CV	**Curriculum Vitae**
DSMB	**Data Safety Monitoring Board**
EU	**European Union**
EudraCT	**European drug regulatory affairs Clinical Trials**
FiO2	**Fraction of inspired oxygen**
GCP	**Good Clinical Practice**
IB	**Investigator's Brochure**
IC	**Informed Consent**
IMP	**Investigational Medicinal Product**
IMPD	**Investigational Medicinal Product Dossier**
METC	**Medical research ethics committee (MREC); in Dutch: medisch ethische toetsing commissie (METC)**
PaO2	**Partial Pressure of Oxygen in Arterial Blood**
ROS	**Reactive oxygen species**
(S)AE	**(Serious) Adverse Event**
SDF	**Sidestream Dark Field Imaging**
SPC	**Summary of Product Characteristics (in Dutch: officiële productinfomatie IB1-tekst)**
Sponsor	**The sponsor is the party that commissions the organisation or performance of the research, for example a pharmaceutical company, academic hospital, scientific organisation or investigator. A party that provides funding for a study but does not commission it is not regarded as the sponsor, but referred to as a subsidising party.**
SUSAR	**Suspected Unexpected Serious Adverse Reaction**
SVR(I)	**Systemic vascular resistance (index)**
Wbp	**Personal Data Protection Act (in Dutch: Wet Bescherming Persoonsgevens)**

WMO	**Medical Research Involving Human Subjects Act (in Dutch: Wet Medisch-wetenschappelijk Onderzoek met Mensen**

1. SUMMARY

Rationale

Contrary to hypoxia, many physicians do not consider hyperoxia harmful for their patients. To prevent hypoxia, superfluous administration of oxygen is common practice, and hyperoxia is seen in many patients, especially on Intensive Care units. However, an increasing number of studies not only confirm the known negative pulmonary effects of chronic oxygen oversupply, but also important and more acute circulatory effects, characterised by decreased cardiac output (CO), increased systemic vascular resistance (SVR), and impaired microvascular perfusion. These phenomena can impair perfusion of organs, which may outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and perturbed organ function. This may for example be responsible for hyperoxia-associated increased infarct size and increased mortality after myocardial infarction and cardiac arrest. The underlying mechanisms are not clarified yet, but probably involve increased oxidative stress with systemic vasoconstriction.

On the other hand, hyperoxia can also induce several favourable effects. The majority of ICU-patients have a systemic inflammatory response syndrome (SIRS) with concomitant vasoplegia due to trauma, sepsis or ischemia/reperfusion injury. Vasoconstriction could benefit these patients with severe SIRS, reducing the need for intravenous volume resuscitation and vasopressor requirements. Furthermore, hyperoxia may exert a preconditioning effect in patients with ischemia/reperfusion injury and prevent new infections due to its antimicrobacterial properties.

Hypothesis: Hyperoxia during SIRS ultimately has unfavourable effects on organ function, especially on a longer term.

Objectives

3. To study the short- and long-term effect of two different PaO_2 targets on circulatory status, organ dysfunction and outcome.
4. To study underlying mechanisms of hyperoxia by determining differences in oxidative stress response between the hyperoxic and the normoxic patients.

Study design

Randomized, prospective multicentre clinical trial

Study population

Patients admitted to the Intensive Care unit with ≥ 2 positive SIRS-criteria and an expected ICU stay of more than 48 hours

Intervention

We will investigate 2 groups with PaO2 targets both within the range of current practice
Group 1: target PaO_2 120 (105 – 135) mmHg (hyperoxemic)
Group 2: PaO_2 75 (60 – 90) mmHg (normoxemic)

Primary endpoints

The primary endpoint will be cumulative daily delta SOFA score (CDDS) from day 1 to day 14.

Secondary endpoints

Total maximum SOFA score, total maximum SOFA score - SOFA score on admission, SOFA rate of decline
Hypoxic events (PaO2 <55 mmHg)
Vasopressor / Inotrope requirements
Renal function, fluid balance
Oxidative stress (F2-isoprostanes)
Clinical endpoints: duration of mechanical ventilation, ventilator-free days, length of stay (in ICU, in hospital) and mortality.
Subgroup: SVRI, CI, EVLW (PiCCO), microcirculatory flow index, perfused vessel density, fluid status by bio-impedance

Feasability endpoint: Time spent in the assigned PaO2 range

Nature and extent of the burden and risks associated with participation, benefit and group relatedness:

The risk and burden for study subjects are small. Placement of central venous catheters and arterial cannulas are part of standard ICU care. Blood sampling is combined with sampling for normal care of patients and will be taken from either the arterial cannula or the central venous catheter. SDF and bio-impedance measurements are non-invasive causing slight discomfort but no pain or risk for patients. The titrated oxygen levels administered to the patients are based on the PaO_2 measured in blood and pulse oximetry, and the oxygen levels are within the range of current practice, therefore we do not expose the patients to additional risk.

2. INTRODUCTION AND RATIONALE

Hyperoxia has been encountered in 44% of the patients requiring ventilatory support in the Intensive Care [1]. However, contrary to hypoxia, many physicians do not consider hyperoxia harmful for their patients. To stay away from hypoxia, superfluous administration of oxygen is common practice. Since the pulse oximeter never indicates more than 100%, physicians are often not aware of the unphysiological high PaO_2 level. Hyperoxic arterial blood gas values do not commonly cause concern, as physicians lower the FiO_2 in only 25% of the observed cases [1].

However, an increasing number of studies not only confirm the well-known negative pulmonary effects of chronic hyperoxia, but also point to more acute circulatory and perfusion effects[2, 3]. In patients with myocardial or cerebral infarction, for example, hyperoxia increases infarct size and mortality [4, 5]. After cardiac arrest, hyperoxia is associated with worse functional outcome and increased mortality.[6]

The underlying mechanisms of hyperoxia's detrimental effects are not clarified. Increased production of reactive oxygen species (ROS), causing oxidative stress, may play a pivotal role[7], although not all study results are unequivocal [8, 9]. Both animal and human studies suggest that oxidative stress induces systemic vasoconstriction, especially in the microcirculation with a loss of functional capillary density and diminished microvascular flow [10]. This in turn augments systemic vascular resistance and impairs cardiac output [11-14]. Impaired effective circulating volume and microvascular tissue perfusion will outweigh marginally higher arterial oxygen content (dissolved oxygen hardly contributes to blood oxygen content). Hence, a loss of organ perfusion and oxygen delivery may occur.

However, hyperoxia can also induce several favourable effects, illustrating the need for more clinical and preclinical studies. In patients with severe systemic inflammatory response syndrome (SIRS) with concomitant vasoplegia hyperoxia-induced vasoconstriction may stabilize hemodynamics and reduce the need for intravenous volume resuscitation and vasopressor treatment. Common causes of SIRS in the ICU are trauma, sepsis and ischemia/reperfusion after cardiac arrest or cardiopulmonary bypass. [15] Pretreatment with hyperoxia induces a low-graded systemic oxidative stress which may exert a preconditioning effect on the ischemia/reperfusion injury. This may, in contrast to hyperoxia during or after the ischemia/reperfusion insult, decrease myocardial damage and other organ injury.[16] The patients with sepsis can also benefit from the potential antimicrobacterial properties of hyperoxia, which may also prevent new infections.[17-19] Furthermore, in patients with haemorrhage, systemic vasoconstriction due to hyperoxia may cause redistribution of blood

flow to the vital organs [20] with amelioration of haemorrhagic shock-induced acute kidney injury. [21]

In critically ill patients, a recent retrospective observational study suggested an independent association between both low and high PaO_2 with in-hospital mortality, with the nadir of mortality between the 70 and 160 mmHg[22]. However, such studies are subject to many forms of bias, and another retrospective study did not confirm these results.[23] Clearly, prospective trials are needed to search for the optimal pO2 range. Hence, it is not just the uncertainty of hyperoxia's untoward effects, but also the possibility of some favourable effects that generates the need for prospective studies.

To the best of our knowledge, no prospective clinical studies have shown benefits of supranormal oxygen levels in any subgroup of critically ill patients.
In this study, we will investigate two different oxygenation levels both near to the nadir of mortality as estimated in an earlier retrospective trial, but one being within the natural range and the other in the supranatural range.[22] In critically ill patients with SIRS, we will assess the effect on organ dysfunction and circulatory parameters. We will separately analyze the predefined subgroups sepsis, trauma/hemorrhage and post-resuscitation.

Amendment July 26, 2016: Rationale for diaphragm function measurements
Diaphragm dysfunction or ICU-acquired respiratory muscle weakness is a common and challenging problem in mechanically ventilated patients. It is associated with increased duration of mechanical ventilation and worse outcomes. Hyperoxia is known to be a possible risk factor for diaphragm weakness. The measurement of diaphragm(dys)function in patients randomized to hyperoxia vs. normoxia could elucidate the clinical relevance of hyperoxia as a causal mechanism of ICU-acquired respiratory muscle weakness.

References to rationale for diaphragm function measurements:

- Heunks, Doorduin, van der Hoeven. Monitoring and preventing diaphragm injury. Curr Opin Crit Care 2015.
- Andrade, dos Santos, Silva, et al. Influence of Hyperoxia and Mechanical Ventilation in Lung Inflammation and Diaphragm Function in Aged Versus Adult Rats. Inflammation 2014.
- Anzueto, Brassard, Andrade, et al. Effects of hyperoxia on rat diaphragm function. J Appl Physiol 1985.

3. OBJECTIVES

3.1 Primary Objective

The primary aim is to determine whether the normoxic target leads to a lower cumulative daily delta SOFA score (CDDS) from day 1 to day 14 as compared to the hyperoxic group. For detailed calculations and justification of the CDDS endpoint, see section 10.2

3.2 Secondary Objective(s)

The secondary aims are to compare total maximum SOFA score (=total of the most deranged score of each organ system during ICU stay), total maximum SOFA score - SOFA score on admission, SOFA rate of decline, hypoxic episodes (PaO_2 <55 mmHg), vasopressor / inotrope requirements (max dose every 24 hours), need for renal replacement therapy and fluid balances (every 24 hours) between the normoxic and the hyperoxic groups. Furthermore, we will determine whether oxidative stress parameters F_2-isoprostanes (on day 1,2, and 4) and as clinical endpoints: duration of mechanical ventilation, lung injury score, ventilator-free days, length of stay (in ICU, in hospital) and mortality (ICU and hospital) are different between both groups. Interim analyses will take place after inclusion of 100 patients and 250 to detect possible differences in mortality.

Feasability endpoint:

Time spent in the assigned PaO_2 range

Subgroup

To further investigate the circulatory changes due to differences in oxygen suppletion, we will study additional parameters in a subgroup of 40 patients, which are too time-consuming to be performed in the whole group. We will estimate hemodynamics by PICCO (C.I., SVRI, extravascular lung water), microcirculation by sublingual Sidestream Dark Field imaging, and body fluid status by bio-impedance on day 1, 2 and 4.

4. STUDY DESIGN

Design:	**randomized, clinical multicenter trial**
Duration:	24 -30 months
Setting:	Intensive Care Units of VU University Medical Center, Academic Medical Center, Tergooiziekenhuizen, Slotervaartziekenhuis, Medisch Centrum Alkmaar and Sint Franciscus Gasthuis.

5. STUDY POPULATION

5.1 Population (base)

The study population consists of patients admitted to the Intensive Care Units of the VU University Medical Center, Academic Medical Center, Tergooiziekenhuizen, Slotervaartziekenhuis, Medisch Centrum Alkmaar and Sint Franciscus Gasthuis.

5.2 Inclusion criteria

In order to be eligible to participate in this study, a subject must meet all of the following criteria:

- Age ≥18 years
- ≥2 positive SIRS-criteria:
  - Temperature >38°C or hypothermia <36°C
  - Heart rate >90 bpm
  - Respiratory rate >20 /min or pCO2 <32 mmHg (4.3 kPa)
  - Number of leucocytes >12 x 109/l of <4 x 109/l of >10% bands
- Within 12 hours of admittance to the ICU
- Expected stay of more than 48 hours as estimated by the attending physician

5.3 Exclusion criteria

A potential subject who meets any of the following criteria will be excluded from participation in this study:

- Elective surgery
- Carbon monoxide poisoning
- Cyanide intoxication
- Methemoglobinemia
- Sickle cell anemia
- Severe pulmonary arterial hypertension (WHO class III or IV)
- Known severe ARDS (PaO2/FiO2 ≤100 mmHg and PEEP ≥ 5 H2O)[24]
- Known cardiac right to left shunting
- Pregnancy
- Severe COPD (Gold class III or IV) or other severe chronic pulmonary disease

5.4 Sample size calculation

Power calculations are based on mean delta SOFA scores, which are invariably reflected in CDDS.

The trial is designed to detected a difference of 0.33 standard deviation (SD) on the primary endpoint with 90% power and a 2-sided alpha of 0.05, with 2 interim analyses.

The trial will be stopped if there is evidence for outcome differences in either direction. Alpha spending for the interim analyses is approximated with a Lan-DeMets O'Brien-Fleming spending function. The total sample size thus needed is 385 patients.

6. TREATMENT OF SUBJECTS

6.1 Investigational product/treatment

In this study we will investigate 2 groups with PaO2 targets both within the range of current practice:

Group 1: target PaO_2 120 (105 – 135) mmHg (hyperoxic)

Group 2: target PaO_2 75 (60 – 90) mmHg (normoxic)

The oxygenation goals are the long-term PaO_2 targets for the participant's entire stay in the ICU.

We will not use excessive measures to achieve the PaO2 targets in group 1. For patients with mechanical ventilation excessive measures include FiO2 settings of >0.60 or prone position. For patients who are not intubated excessive measures include non-invasive mechanical ventilation or intubation. For patients who will not be able to achieve the PaO2 targets of their group, PaO2 targets will be determined by the treating physicians feasible with conventional measures not including those mentioned above. Temporary measures to improve oxygenation for planned procedures involving upper airways such as tracheostomy, bronchoscopy etc will follow standard practices of the participating centers. These aberrations from study targets will be limited to the shortest duration possible.

6.2 Use of co-intervention (if applicable)

Not applicable

7. INVESTIGATIONAL PRODUCT

7.1 Name and description of investigational product(s)

Oxygen

7.2 Summary of findings from clinical and non-clinical studies with known and potential risks and benefits

Animal and human studies indicate that hyperoxia (mostly severe hyperoxia with PaO2 > 300 mmHg after a period of ventilation with an FiO2 of 100%) can lead to important circulatory effects. The hypothesis is that hyperoxia causes vasoconstriction in the microcirculation with a loss of functional capillary density and disturbed microvascular flow. [10] Vasoconstriction causes an increase in systemic vascular resistance and a reduction of the cardiac output. [11-14] This loss of perfusion has been suggested to outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and an increase of ischemia/reperfusion injury.

In patients with myocardial or cerebral infarction, hyperoxia increases infarct size and mortality, and in patients after cardiac arrest hyperoxia is associated with worse functional outcome and increased mortality. [4, 5 6]

Up until now, no prospective, randomized controlled studies investigating the effect of hyperoxia in ICU patients have been performed. A recent large retrospective study showed an independent association between both low and high PaO_2 with in-hospital mortality [22]. However, another retrospective study [23] did not find this association.

7.3 Description and justification of route of administration and dosage

Fraction of inspired oxygen (FiO_2) which is applied to the patient by the mechanical ventilator or oxygen suppletion by nasal cannula or oxygen mask.

7.4 Dosages, dosage modifications and method of administration

In this study we will investigate 2 groups with PaO2 targets both within the range of current practice:

Group 1: target PaO_2 at ICU 120 (105 – 135) mm Hg
Group 2: target PaO_2 at ICU 75 (60 – 90) mmHg
The oxygenation goals are the long-term PaO2 target for the participant's entire stay in the ICU.

We will not use excessive measures to achieve the PaO2 targets in group 1. For patients with mechanical ventilation excessive measures include FiO2 settings of >0.60 or prone

position. For patients who are not intubated excessive measures include non-invasive mechanical ventilation or intubation. For patients who will not be able to achieve the PaO2 targets of their group, PaO2 targets will be determined by the treating physicians feasible with conventional measures not including those mentioned above. Temporary measures to improve oxygenation for planned procedures involving upper airways such as tracheostomy, bronchoscopy etc will follow standard practices of the participating centers. These aberrations from study targets will be limited to the shortest duration possible.

7.5 Preparation and labelling of Investigational Medicinal Product

Not applicable

7.6 Drug accountability

Not applicable

8. METHODS

8.1 Study parameters/endpoints

8.1.1 Main study parameter/endpoint

The primary endpoint will be cumulative daily delta SOFA score (CDDS) from day 1 to day 14. For detailed calculations and justification of the CDDS endpoint, see section 10.2

8.1.2 Secondary study parameters/endpoints

Secondary parameters will include total maximum SOFA score(= total of the most deranged score of each organ system during ICU stay), total maximum SOFA score - SOFA score on admission, SOFA rate of decline, hypoxic episodes (PaO_2 <55 mmHg), vasopressor / inotrope requirements (max dose every 24 hours), need for renal replacement therapy and fluid balances (every 24 hours). Furthermore, oxidative stress parameters F_2-isoprostanes will be determined (on days 1, 2 and 4) and as clinical endpoints: duration of mechanical ventilation, ventilator-free days, length of stay (in ICU, in hospital) and mortality (ICU and hospital). Interim analyses will take place after inclusion of 100 patients to detect possible differences in mortality.
The amount, modality and duration of oxygen administered prior to ICU admission will be recorded. This will be treated as a potential effect-modifying variable and reported in the baseline characteristics.

8.1.3 Feasibility endpoint

Time spent in the assigned PaO_2 range

8.1.4 Subgroup

To further investigate the circulatory changes due to differences in oxygen suppletion, we will study additional parameters in a subgroup of patients, which are too time-consuming to be performed in the whole group. We will estimate hemodynamics by PICCO (C.I., SVRI, extravascular lung water), microcirculation by sublingual Sidestream Dark Field imaging, and body fluid status by bio-impedance.

a) Arterial blood gas analysis with PaO_2 and lactate measurement will be determined initially at least every 4 hours, and at least every 6 hours after stabilisation. Data will be collected during the entire stay on the ICU. Area under the curve will be calculated for PaO_2, O_2 saturation, FiO_2 and lactate.

b) Hemodynamics
Whenever patients for more intensive hemodynamic monitoring are provided with continuous cardiac output measurement by Swan Ganz catheter or PiCCO, SVRI, CO and CI and EVLW will be measured every 4 hours. In all patients with a central line in the jugular vein, SvO2 will be determined daily.

e) Sublingual mucosal microcirculation measurements will be performed on day 1, 2, and 4 using sidestream dark field (SDF) imaging and quantified as the level of perfused small vessel density and microvascular flow index (vessel diameter < 20 µm).

f) Bio-impedance measured resistance/reactance/impedance at day 1, 2, and 4.
g) Diaphragm dysfunction will be quantified (and compared between groups) using electromyography (Edi) and ultrasonography (diaphragm thickness, thickening fraction) at days 1, 2 and 4.

8.2 Randomisation, blinding and treatment allocation

Randomisation will be performed with the use of randomisation list generated by a web-based computer program

8.3 Study procedures

5) Deferred consent by patient representative
6) Routine clinical data (demographic characteristics, reason of admission, comorbidity, APACHE II)
7) Titration of FiO_2, based on measured PaO_2
   group 1: target PaO_2 at ICU of 120 (105 – 135) mm Hg
   group 2: : target PaO_2 at ICU of 75 (60 – 90) mmHg
8) Blood sample collection at baseline and on day 2, and 4 for determination of parameters of oxidative stress and tissue/organ perfusion *(in total 40 ml extra for study*). Remaining blood material after analysis will be stored for additional analyses in the future.

Substudy

In a subgroup of 100 patients, hemodynamics will be more intensively monitored by SDF, PiCCO and bio-impedance. Diaphragm function will be measured using electromyography (Maquet NAVA® feeding tube or similar) and ultrasonography at days 1, 2 and 4. Ultrasonography of heart and lungs (and diaphragm) is part of routine ICU treatment. Patients who do not need a feeding tube will not be eligible for inclusion in the substudy. These additional measurements will only be done in a subgroup of patients because they are very time consuming and it is logistically impossible to perform them in all the patients.

8.4 Withdrawal of individual subjects

Subjects can leave the study at any time for any reason if they wish to do so without any consequences. The investigator can decide to withdraw a subject from the study for urgent medical reasons.

9. SAFETY REPORTING

9.1 Section 10 WMO event

In accordance to section 10, subsection 1, of the WMO, the investigator will inform the subjects and the reviewing accredited METC if anything occurs, on the basis of which it appears that the disadvantages of participation may be significantly greater than was foreseen in the research proposal. The study will be suspended pending further review by the accredited METC, except insofar as suspension would jeopardise the subjects' health. The investigator will take care that all subjects are kept informed.

9.2 SAEs and SUSARs

This study compares two treatment targets that are used in standard care. All included patients are admitted to the Intensive Care with a life-threatening disease. Any major adverse development can be expected as part of the underlying disease. Therefore, it is not achievable to report all these developments individually as SAEs. Every three months we will send a line-listing of all SAEs to the METc. Furthermore, all deaths will be reported via ToetsingOnline to the METc/CCMO within one week of the event.

9.3 Data Safety Monitoring Board (DSMB) / Safety Committee

Since the titration of FiO_2 based on the measured pO_2 is within the range of standard care, we do not expose the patients to additional risk. Therefore a data safety monitoring board will not be instituted.

10. STATISTICAL ANALYSIS

10.1 Descriptive statistics

Statistical analysis will be performed using the SPSS statistical software package (SPSS Inc.®, Chicago USA). All included patients will be incorporated in the statistical analysis. Descriptive statistics of these quantitative data will include mean, median, standard deviation and interquartile range.

10.2 Primary and secondary study parameter(s)

Calculations of CDDS primary endpoint

The primary endpoint will be *cumulative daily delta SOFA score* (CDDS) from day 1 to day 14, calculated as:

$$CDDS = \sum_{i=day\,2\,to\,14} (SOFA_{DAYi} - SOFA_{ADMISSION})$$

or alternatively,

$$CDDS = SOFA_{DAY\,2} + SOFA_{DAY\,3} + SOFA_{DAY\,4} + ...etc + SOFA_{DAY\,14} - (13 \cdot SOFA_{ADMISSION})$$

Daily SOFA score is calculated as the total of maximum scores for each organ system excluding respiratory system (because of possible PaO_2/FiO_2 distortion). For patients discharged from the ICU, SOFA score will be registered as 0 from the day of discharge to day 14. Death in the ICU will be registered as a score of 20 (maximum) from the day of death to day 14.

CDDS reflects a balanced weighting of the following clinically relevant preconditions:

7. Low (or negative) delta SOFA score is better than high (or positive) delta SOFA score.
8. Shorter duration of ICU stay is better than longer stay.
9. Discharge from the ICU is better than death in the ICU.
10. Eventual discharge after a high admission SOFA score is better than discharge after a low admission SOFA score.
11. Eventual death after a low admission SOFA score is worse than discharge after a high admission SOFA score.
12. Early SOFA score reduction is better than late SOFA score reduction.

Other possible endpoints (such as *delta SOFA score* or *SOFA rate of decline*) fail to satisfy most of these criteria.

The differences of primary and secondary parameters between the 2 groups will be calculated by intention-to-treat analysis. The primary endpoint (CDDS) is designed and

tested as a tool for ranking outcomes. The between-group comparison on the primary endpoint will therefore be made using the Wilcoxon rank-sum test.
Secondary outcomes will be compared using ANOVA for repeated measurements, Chi-Squared, Mann-Whitney U or Fisher's Exact test where appropriate. Mortality will be assessed with the Kaplan Meier and log rank test.

10.3 Interim analysis (if applicable)
Interim analyses (blinded for the intervention arm) will take place after inclusion of 150 and 275 patients to detect possible differences in mortality.

The interim analyses are planned as a symmetric two-sided group sequential design with 90% power and 5% two-sided Type I Error. Spending computations assume the trial stops if a bound is crossed.

Analysis	N	Boundary two-sided P	Spend
1	150	0.0006	0.0006
2	275	0.0156	0.0152
3	385	0.0452	0.0342
total			0.050

++ alpha spending:
Lan-DeMets O'brien-Fleming approximation spending function

For example, the trial will be stopped after 275 patients if there is a sufficient difference in primary endpoint between the two groups, such that P<0.0156.

In addition to the trial stopping boundaries for superior outcomes in one of the trial arms (outlined above), we have defined a futility stopping boundary at interim-analysis 2 (after N=275 patients): The trial will be stopped if there is almost no difference in the primary endpoint between the treatment arms, such that the conditional power is less than 20% at interim-analysis 2. Practically, if P>0.459 after 275 patients, there is a less than 20% probability of obtaining a significant result after 385 patients.

The METc/CRB will be consulted when, at the interim analysis, between-group differences in mortality are large enough so that P<0.05 but not large enough to reach one of the stopping criteria outlined above.

11. ETHICAL CONSIDERATIONS

11.1 Regulation statement

The study will be conducted according to the principles of the Declaration of Helsinki (59th WMA General Assembly, Seoul 2008) and in accordance with the Medical Research Involving Human Act (WMO) and the statements of the CCMO as presented in the publication "Uitgestelde toestemming voor inclusie van beslissingsonbekwame patiënten in studies van spoedeisende geneeskunde" by E.J.O.Kompanje.[25]

11.2 Recruitment and consent

11.2.1. Deferred consent

For this study we ask for deferred consent and we appeal to the emergency procedure for consent in medical research as stated in article 6, paragraph 4 of the WMO because of the following reasons.

Oxygen suppletion is applied to almost all ICU-patients. To alleviate respiratory distress oxygen therapy is initiated and cannot be postponed. Patients admitted to an Intensive Care Unit (ICU) are mostly incompetent to give informed consent. Obtaining informed consent from a legal representative takes time (on average up to 12 hours), even by an experienced research team.[26]

We propose to randomize each patient who meets the inclusion criteria ultimately within 2 hours after ICU admission. Oxygenation targets according to randomization are applied immediately thenceforth. Informed consent from the legal representative will be requested as soon as possible thereafter, but never later than 24 hours after randomization. If informed consent is not obtained within those 24 hours, or if a legal representative denies participation within this time frame, the patient is excluded and data will no longer be used. Thenceforth the patient is oxygenated according to the policy of the attending physician.

During hospital admission, we will attempt to achieve informed consent from the patients themselves. If this was not possible (*for example* due to incomplete neurological or physical recovery, or due to early transfer to another hospital) we will send a letter to the patients to inform them that we would like to use the data and blood samples obtained during the study. If they don't want their data to be used, they can contact us by the telephone numbers or email addresses we have provided in the letter. We will not send this letter to the patients who cannot recover sufficiently to understand the letter and take a considered decision about the study.

11.2.2 Effects of hyperoxia

Hyperoxia can induce more chronic, harmful effects to the lungs, but also more acute circulatory effects, characterised by decreased cardiac output, increased systemic vascular resistance, and impaired microvascular perfusion. These phenomena can impair perfusion of organs, which may outweigh higher arterial oxygen content, resulting in a net loss of oxygen delivery and perturbed organ function. This may be responsible for hyperoxia-associated increased infarct size and increased mortality after myocardial infarction and cardiac arrest. [2, 3, 6] On the other hand, hyperoxia can also induce several favourable effects. The majority of ICU-patients have a systemic inflammatory response syndrome (SIRS) with concomitant vasoplegia due to trauma, sepsis or ischemia/reperfusion injury. Vasoconstriction could benefit these patients with severe SIRS, reducing the need for intravenous volume resuscitation and vasopressor requirements. Furthermore, hyperoxia may exert a preconditioning effect in patients with ischemia/reperfusion injury and prevent new infections due to its antimicrobacterial properties.

If a patient, in the proposed trial, is already hyperoxic for several hours, effects of this oxygenation target could already be in place, largely reducing validity of the trial outcomes. From experimental animal studies we know that hyperoxia can cause circulatory changes within an hour.[27, 28] These findings are in line with results from clinical studies, showing effects on cardiac output and systemic vascular resistance even within 10 minutes.[29]

11.2.3. Time between asking for and obtaining informed consent

Most critically ill patients cannot give informed consent for a study at ICU admission. They are in severe respiratory distress, sedated or in coma. Time to obtain informed consent after recognition of study eligibility by a legal representative could take as much as 12 hours, even when a legal representative could be contacted fast. This is in line with a recent observational study performed in the Academic Medical Center, Amsterdam, the Netherlands. Time to obtain informed consent after recognition of study eligibility from a legal representative was as high as 14 hours (study ID NL34294.018.10). For relatives of a severely ill patient who has been acutely admitted to the ICU it is difficult to make a quick, but balanced decision. [26, 30, 31]

Most ICU patients are positive about their enrollment under deferred consent. A contentment questionnaire of the large NICE–SUGAR trial [32], a trial studying different targets in blood glucose control [33], showed that the large majority (96%) would have granted consent if they would have been asked. 93% of the patients were content with the decision made by the representative at the moment they were incapable of giving informed consent. [33]

11.2.4. Ethical aspects

We can underpin the idea of 'clinical equipoise'.[34] The study participant can benefit from the intervention, but up to now there is a state of honest, professional disagreement in the community of expert practitioners as to the optimal oxygenation targets for ICU-patients. Currently, an implementation trial (Oxytar, de Jonge LUMC, NTR 3424) is running, aiming for target PaO_2 values of 55 – 80 mmHg in ICU patients. However, this target is not based on prospective, let alone randomised clinical evidence. Some arguments act in favour of this target oxygen range [35], but others do not. For example, the nadir for unadjusted hospital mortality as observed in a Dutch retrospective study was substantially higher than this target, i.e. just below 150 mmHg [22]. International guidelines for oxygen suppletion in medical emergencies developed by the British Thoracic Society and endorsed by several other societies, recommend a target peripheral O_2 saturation range measured by pulse oximetry of 94 – 98%. [36] It is difficult to translate this O_2 saturation range to target pO_2 range, since their relation fluctuates dependent on other factors like temperature, perfusion, and pH. However, saturations as recommended will commonly result in PaO_2 in a higher range than 55 – 80 mmHg. More importantly, however, these recommendations are also unsupported by randomized controlled trials. Furthermore, since hyperoxia can induce both negative (pulmonary and circulatory) and positive (antimicrobial, preconditioning) also from a pathophysiological view the optimal target for ICU-patients is unclear. Both oxygenation targets in our trial are within the range used in common practice in ICUs worldwide and are considered safe.

11.2.5. Similarities with previous studies using deferred consent

The proposed trial has many similarities with recently published interventional trials in ICU patients using deferred consent. [32, 37-40] The investigated therapies were: intravenous fluid resuscitation with colloids or crystalloids [37], fluid resuscitation with 4% albumin infusion or normal saline infusion [38], RENAL replacement therapy with a lower or higher intensity [39] and ventilation with lower or higher tidal volumes (PReVent). In these trials, as in our trial, patients were incapable at the moment therapy had to be started, the effects of the interventions were for a substantial part induced on its start, and the strategies under study were both used in daily practice.

11.2.6. Conclusions

Critically ill patients are mostly incapable at the moment of ICU admission. Obtaining informed consent from a legal representative takes mostly half a day. Start of oxygen therapy in ICU-patients cannot wait. Both oxygenation targets are within the range used in common practice in ICUs worldwide and are considered safe.

11.3 Benefits and risks assessment, group relatedness

The risk and burden for study subjects are small. Blood sampling is combined with sampling for normal care of patients. SDF and bio-impedance measurements are non-invasive causing slight discomfort but no pain or risk for patients. Since the titration of FiO_2 based on the measured PaO_2 is within the range of standard care, we do not expose the patients to additional risk.

11.4 Compensation for injury

The sponsor/investigator has a liability insurance which is in accordance with article 7, subsection 6 of the WMO.

The sponsor (also) has an insurance which is in accordance with the legal requirements in the Netherlands (Article 7 WMO and the Measure regarding Compulsory Insurance for Clinical Research in Humans of 23th June 2003). This insurance provides cover for damage to research subjects through injury or death caused by the study.

4. € 450.000,-- (i.e. four hundred and fifty thousand Euro) for death or injury for each subject who participates in the Research;
5. € 3.500.000,-- (i.e. three million five hundred thousand Euro) for death or injury for all subjects who participate in the Research;
6. € 5.000.000,-- (i.e. five million Euro) for the total damage incurred by the organisation for all damage disclosed by scientific research for the Sponsor as 'verrichter' in the meaning of said Act in each year of insurance coverage.

The insurance applies to the damage that becomes apparent during the study or within 4 years after the end of the study.

11.5 Incentives (if applicable)

None

11.6 Monitoring Clinical Research Bureau

An independent monitor (quality officer) will monitor the study data according to the regulations described under Good Clinical Practice (GCP). In a selection of study subjects the Informed Consents are controlled. Additionally, during onsite monitoring the officer will perform a Source Data verification of data described in the Case Report Forms to investigation the agreement between source data and study reports. The intensity of this verification is related to the study risk assessment. In particular, inclusion and exclusion criteria and the primary endpoints of the investigation are subject to monitoring. The monitor

will evaluate whether SAE's en SUSAR's are adequately reported within the time frame as directed by the Dutch law.

12. ADMINISTRATIVE ASPECTS, MONITORING AND PUBLICATION

12.1 Handling and storage of data and documents

Patient data will be stored anonymously. Data will not be directly traceable to the individual patients, as all patients are coded. The key to the code will be separately safeguarded by the primary investigator. Data will be stored for 15 years.

12.2 Amendments

Amendments are changes made to the research after a favourable opinion by the accredited METC has been given. All amendments will be notified to the METC that gave a favourable opinion.

A 'substantial amendment' is defined as an amendment to the terms of the METC application, or to the protocol or any other supporting documentation, that is likely to affect to a significant degree:

- the safety or physical or mental integrity of the subjects of the trial;
- the scientific value of the trial;
- the conduct or management of the trial; or
- the quality or safety of any intervention used in the trial.

All substantial amendments will be notified to the METC and to the competent authority. Non-substantial amendments will not be notified to the accredited METC and the competent authority, but will be recorded and filed by the sponsor.

12.3 Annual progress report

The sponsor/investigator will submit a summary of the progress of the trial to the accredited METC once a year. Information will be provided on the date of inclusion of the first subject, numbers of subjects included and numbers of subjects that have completed the trial, serious adverse events/ serious adverse reactions, other problems, and amendments.

12.4 End of study report

The investigator will notify the accredited METC of the end of the study within a period of 8 weeks. The end of the study is defined as the last patient's last visit.
In case the study is ended prematurely, the investigator will notify the accredited METC within 15 days, including the reasons for the premature termination.
Within one year after the end of the study, the investigator/sponsor will submit a final study report with the results of the study, including any publications/abstracts of the study, to the accredited METC.

12.5 Public disclosure and publication policy

We are free to make a publication and have no restrictions made by a sponsor. Patient data will be published anonymously.

13. REFERENCES

Reference List

(1) de Graaff AE, Dongelmans DA, Binnekade JM, de JE. Clinicians' response to hyperoxia in ventilated patients in a Dutch ICU depends on the level of FiO2. *Intensive Care Med* 2011 January;37(1):46-51.

(2) Cornet AD, Kooter AJ, Peters MJ, Smulders YM. Supplemental oxygen therapy in medical emergencies: more harm than benefit? *Arch Intern Med* 2012 February 13;172(3):289-90.

(3) Cornet AD, Kooter AJ, Peters MJ, Smulders YM. The potential harm of oxygen therapy in medical emergencies. *Crit Care* 2013 April 18;17(2):313.

(4) Rawles JM, Kenmure AC. Controlled trial of oxygen in uncomplicated myocardial infarction. *Br Med J* 1976 May 8;1(6018):1121-3.

(5) Ronning OM, Guldvog B. Should stroke victims routinely receive supplemental oxygen? A quasi-randomized controlled trial. *Stroke* 1999 October;30(10):2033-7.

(6) Kilgannon JH, Jones AE, Shapiro NI et al. Association between arterial hyperoxia following resuscitation from cardiac arrest and in-hospital mortality. *JAMA* 2010 June 2;303(21):2165-71.

(7) Jamieson D, Chance B, Cadenas E, Boveris A. The relation of free radical production to hyperoxia. *Annu Rev Physiol* 1986;48:703-19.

(8) Hauser B, Barth E, Bassi G et al. Hemodynamic, metabolic, and organ function effects of pure oxygen ventilation during established fecal peritonitis-induced septic shock. *Crit Care Med* 2009 August;37(8):2465-9.

(9) Barth E, Bassi G, Maybauer DM et al. Effects of ventilation with 100% oxygen during early hyperdynamic porcine fecal peritonitis. *Crit Care Med* 2008 February;36(2):495-503.

(10) Kamler M, Wendt D, Pizanis N, Milekhin V, Schade U, Jakob H. Deleterious effects of oxygen during extracorporeal circulation for the microcirculation in vivo. *Eur J Cardiothorac Surg* 2004 September;26(3):564-70.

(11) Bak Z, Sjoberg F, Rousseau A, Steinvall I, Janerot-Sjoberg B. Human cardiovascular dose-response to supplemental oxygen. *Acta Physiol (Oxf)* 2007 September;191(1):15-24.

(12) Ihnken K, Winkler A, Schlensak C et al. Normoxic cardiopulmonary bypass reduces oxidative myocardial damage and nitric oxide during cardiac operations in the adult. *J Thorac Cardiovasc Surg* 1998 August;116(2):327-34.

(13) Harten JM, Anderson KJ, Kinsella J, Higgins MJ. Normobaric hyperoxia reduces cardiac index in patients after coronary artery bypass surgery. *J Cardiothorac Vasc Anesth* 2005 April;19(2):173-5.

(14) Anderson KJ, Harten JM, Booth MG, Kinsella J. The cardiovascular effects of inspired oxygen fraction in anaesthetized patients. *Eur J Anaesthesiol* 2005 June;22(6):420-5.

(15) Horeczko T, Green JP, Panacek EA. Epidemiology of the Systemic Inflammatory Response Syndrome (SIRS) in the Emergency Department. *West J Emerg Med* 2014 May;15(3):329-36.

(16) Tahep lP, Valen G, Starkopf J, Kairane C, Zilmer M, Vaage J. Pretreating rats with hyperoxia attenuates ischemia-reperfusion injury of the heart. *Life Sci* 2001 February 23;68(14):1629-40.

(17) Calzia E, Asfar P, Hauser B et al. Hyperoxia may be beneficial. *Crit Care Med* 2010 October;38(10 Suppl):S559-S568.

(18) Greif R, Akca O, Horn EP, Kurz A, Sessler DI. Supplemental perioperative oxygen to reduce the incidence of surgical-wound infection. *N Engl J Med* 2000 January 20;342(3):161-7.

(19) Belda FJ, Aguilera L, Garcia de la AJ et al. Supplemental perioperative oxygen and the risk of surgical wound infection: a randomized controlled trial. *JAMA* 2005 October 26;294(16):2035-42.

(20) Bitterman H, Brod V, Weisz G, Kushnir D, Bitterman N. Effects of oxygen on regional hemodynamics in hemorrhagic shock. *Am J Physiol* 1996 July;271(1 Pt 2):H203-H211.

(21) Efrati S, Berman S, Ben AG, Siman-Tov Y, Averbukh Z, Weissgarten J. Application of normobaric hyperoxia therapy for amelioration of haemorrhagic shock-induced acute renal failure. *Nephrol Dial Transplant* 2008 July;23(7):2213-22.

(22) de JE, Peelen L, Keijzers PJ et al. Association between administered oxygen, arterial partial oxygen pressure and mortality in mechanically ventilated intensive care unit patients. *Crit Care* 2008;12(6):R156.

(23) Eastwood G, Bellomo R, Bailey M et al. Arterial oxygen tension and mortality in mechanically ventilated patients. *Intensive Care Med* 2012 January;38(1):91-8.

(24) Steinberg KP, Hudson LD, Goodman RB et al. Efficacy and safety of corticosteroids for persistent acute respiratory distress syndrome. *N Engl J Med* 2006 April 20;354(16):1671-84.

(25) Kompanje EJO, Jansen TC, Le Noble JLML, de Geus HR, Bakker J. [Deferred consent for inclusion of patients unable to give their consent in studies in the field of emergency medicine]. *Ned Tijdschr Geneeskd* 2008 September 20;152(38):2057-61.

(26) Burns KE, Zubrinich C, Tan W et al. Research recruitment practices and critically ill patients. A multicenter, cross-sectional study (the Consent Study). *Am J Respir Crit Care Med* 2013 June 1;187(11):1212-8.

(27) Tsai AG, Cabrales P, Winslow RM, Intaglietta M. Microvascular oxygen distribution in awake hamster window chamber model during hyperoxia. *Am J Physiol Heart Circ Physiol* 2003 October;285(4):H1537-H1545.

(28) Cabrales P, Tsai AG, Intaglietta M. Nitric oxide regulation of microvascular oxygen exchange during hypoxia and hyperoxia. *J Appl Physiol (1985)* 2006 April;100(4):1181-7.

(29) Park JH, Balmain S, Berry C, Morton JJ, McMurray JJ. Potentially detrimental cardiovascular effects of oxygen in patients with chronic left ventricular systolic dysfunction. *Heart* 2010 April;96(7):533-8.

(30) Kompanje EJ, Jansen TC, Le Noble JL, de Geus HR, Bakker J. [Deferred consent for inclusion of patients unable to give their consent in studies in the field of emergency medicine]. *Ned Tijdschr Geneeskd* 2008 September 20;152(38):2057-61.

(31) Jansen TC, Kompanje EJ, Druml C, Menon DK, Wiedermann CJ, Bakker J. Deferred consent in emergency intensive care research: what if the patient dies early? Use the data or not? *Intensive Care Med* 2007 May;33(5):894-900.

(32) Finfer S, Chittock DR, Su SY et al. Intensive versus conventional glucose control in critically ill patients. *N Engl J Med* 2009 March 26;360(13):1283-97.

(33) Potter JE, McKinley S, Delaney A. Research participants' opinions of delayed consent for a randomised controlled trial of glucose control in intensive care. *Intensive Care Med* 2013 March;39(3):472-80.

(34) Freedman B. Equipoise and the ethics of clinical research. *N Engl J Med* 1987 July 16;317(3):141-5.

(35) Ventilation with lower tidal volumes as compared with traditional tidal volumes for acute lung injury and the acute respiratory distress syndrome. The Acute Respiratory Distress Syndrome Network. *N Engl J Med* 2000 May 4;342(18):1301-8.

(36) O'Driscoll BR, Howard LS, Davison AG. BTS guideline for emergency oxygen use in adult patients. *Thorax* 2008 October;63 Suppl 6:vi1-68.

(37) Annane D, Siami S, Jaber S et al. Effects of fluid resuscitation with colloids vs crystalloids on mortality in critically ill patients presenting with hypovolemic shock: the CRISTAL randomized trial. *JAMA* 2013 November 6;310(17):1809-17.

(38) Finfer S, Bellomo R, Boyce N, French J, Myburgh J, Norton R. A comparison of albumin and saline for fluid resuscitation in the intensive care unit. *N Engl J Med* 2004 May 27;350(22):2247-56.

(39) Bellomo R, Cass A, Cole L et al. Intensity of continuous renal-replacement therapy in critically ill patients. *N Engl J Med* 2009 October 22;361(17):1627-38.

(40) Jansen TC, van BJ, Schoonderbeek FJ et al. Early lactate-guided therapy in intensive care unit patients: a multicenter, open-label, randomized controlled trial. *Am J Respir Crit Care Med* 2010 September 15;182(6):752-61.