ORIGINAL ARTICLE

Inhaled nitric oxide dependency at the end of double-lung transplantation: a boosted propensity score cohort analysis

Julien Fessler[1,2], Mathieu Godement[3,4], Romain Pirracchio[5,6], Jean-Yves Marandon[1,2], Jacques Thes[1,2], Edouard Sage[2,7], Antoine Roux[2,8], François Parquin[2,7], Charles Cerf[9], Marc Fischler[1,2] & Morgan Le Guen[1,2]

1 Department of Anesthesiology, Hôpital Foch, Suresnes, France
2 Université Versailles-Saint-Quentin-en-Yvelines, Versailles, France
3 Department of Anesthesiology and Intensive Care Medicine, Hôpital Bichat, Paris, France
4 Université Paris Diderot, Paris, France
5 Department of Anesthesiology and Intensive Care Medicine, Hôpital Européen Georges Pompidou, Paris, France
6 Department of Biostatistics and of Medical Informatics, Inserm U1153, ECSTRA, Hôpital Saint Louis, Université Paris Diderot, Sorbonne Paris Cité, Paris, France
7 Department of Thoracic Surgery, Hôpital Foch, Suresnes, France
8 Department of Pneumology, Hôpital Foch, Suresnes, France
9 Department of Intensive Care Medicine, Hôpital Foch, Suresnes, France

Correspondence
Marc Fischler, MD, Department of Anesthesiology, Hospital Foch, 40 rue Worth, Suresnes, France and University Versailles Saint-Quentin en Yvelines, Versailles, France.
Tel: 0033 1 46 25 24 42;
fax: 0033 1 46 25 20 88;
e-mail: m.fischler@hopital-foch.org

This work is attributed to the Department of Anesthesiology, Hôpital Foch, 92150 Suresnes, France.

SUMMARY

Inhaled nitric oxide (iNO) is usually used during lung transplantation despite controversial postoperative benefits. Our group chose to administer iNO systematically during the procedure and stop at the end of surgery. This study aims to describe the features of patients who cannot be weaned from iNO, the reasons for this and its impact on postoperative outcomes.
This is a monocentric cohort study comprised all consecutive patients who underwent double-lung transplantation (DLT) between 1 January 2012 and 1 January 2016. The impact of iNO dependency on postoperative outcomes was estimated using a boosted inverse probability of treatment weighting estimator.
A total of 9.8% of the 173 patients included in the study could not be weaned from iNO at end-surgery stage. Body mass index (OR = 2.03, 95% CI = 1.14–3.29, $P = 0.02$) and intraoperative extracorporeal membrane oxygenation (OR = 1.80, 95% CI = 1.02–2.72, $P = 0.04$) were risk factors for iNO dependency In the weighted population, iNO dependency was associated with an increased prevalence of grade 3 primary graft dysfunction (adjusted RR = 4.20, 95% CI = 1.75–10.09, $P < 0.001$) and decreased postoperative survival during the first 1500 days of follow-up (adjusted HR = 5.0, 95% CI = 1.86–13.48, $P < 0.001$).
Inhaled nitric oxide dependency is an early marker of a poor prognosis following DLT.

Transplant International 2019; 32: 244–256

Key words
lung transplantation, nitric oxide

Received: 11 March 2018; Revision requested: 2 July 2018; Accepted: 14 November 2018;
Published online: 26 December 2018

doi:10.1111/tri.13381

Introduction

Inhaled nitric oxide (iNO) is a selective pulmonary vasodilator that can be useful during lung transplantation because it (i) decreases pulmonary arterial resistance and has a favorable effect on right ventricular function [1], (ii) enhances arterial oxygenation by optimizing the ventilation/perfusion ratio [2], (iii) prevents reperfusion edema by reducing pulmonary capillary pressure and inhibiting leukocyte-endothelial interactions [3,4], (iv) decreases ischemic-reperfusion lesions and early inflammatory insults to the graft [5,6] and (v) prevents apoptosis [7]. The potential beneficial effects of iNO may impact all phases of lung transplantation, especially during single-lung ventilation and pulmonary artery clamping or after reperfusion and re-ventilation of the grafted lungs. Hence, iNO is widely used in this indication despite a lack of randomized controlled trials supporting its benefits in terms of oxygenation, pulmonary ischemia-reperfusion injury or the prevention of primary graft dysfunction (PGD) [8].

However, because iNO is systematically delivered throughout a lung transplant procedure at our institution, we were able to identify a group of patients who could not be weaned from iNO at the end of the procedure ("iNO-dependent" patients) for hemodynamic and/or respiratory reasons. The goal of this study was to describe the features of these patients, the reasons for iNO dependency at the end of their surgery and its impact on postoperative outcome.

Methods

This study is a retrospective analysis of a prospectively maintained institutional Anesthesia Lung Transplant Database. Ethical approval for this study was provided by the Ethical Committee of the French Society of Anesthesia and Intensive Care (SFAR) (Chairperson, Prof. J. E. Bazin) on 11 November 2017 (Reference 00010254-2017-015). Patient consent was waived. The Foch Lung Transplant group attests that they performed all procedures in strict compliance with the International Society for Heart and Lung Transplantation ethics statement.

All consecutive patients who underwent double-lung transplantation (DLT) at Foch Hospital between 1 January 2012 and 1 January 2016 (a convenient sample population selected because of its stability in terms of medical and surgical practices) were eligible for inclusion in the registry. Patients transplanted twice during the study period or undergoing multiorgan transplantation, patients who required preoperative extracorporeal membrane oxygenation (ECMO) or intraoperative cardio-pulmonary bypass, and those in whom ECMO was started during the surgical procedure but could not be weaned before Intensive Care Unit (ICU) transfer were excluded from the analysis. All organs were obtained from brain-dead donors regardless of etiology.

Intraoperative transplant protocol

Surgical technique

DLT, consisting of two successive mono-pulmonary transplants [9], is performed via two antero-lateral thoracotomy incisions in almost all cases. Implantation of the first graft follows these steps: anastomose of the main bronchi, anastomose of the donor and recipient atrial cuffs and anastomose of the pulmonary arteries. The graft is re-inflated gently and then re-perfused gradually. Deairing of the graft is performed anteriorly, then the pulmonary artery clamp is replaced and the left atrial clamp is removed to allow back bleeding. Once good function of the new donor lung is confirmed, the second implantation follows the same sequence. Two drainage tubes are placed in each thoracic cavity at the end of the procedure.

Anesthetic management

The standardized perioperative anesthetic management protocol used at this institution has been published recently [10]. It combines total intra-venous anesthesia with propofol, remifentanil, atracurium and thoracic epidural analgesia through which an opioid and a local anesthetic is administered just before surgical incision.

Intraoperative hemodynamic monitoring includes systemic arterial pressure continuous monitoring, pulmonary artery catheterization (Swan-Ganz CCOmbo Pulmonary Artery Catheter; Edwards Lifesciences Corp, Irvine, CA, USA) and transesophageal echocardiography (TEE) (Vivid 7 and a multiplane probe 6.2/5.0 MHz, GE Healthcare, Fairfield, CT, USA). A standardized TEE examination is performed at each step of the procedure to examine especially left and right ventricular functions [11,12]. TEE is also used, in particular, to look for a patent foramen ovale, to help to position the venous cannula when using veno-arterial ECMO and to assess the vascular anastomoses [13]. Moreover, TEE shows, at any time during the procedure, the reason for acute hemodynamic modifications (decrease in blood

pressure, increase in pulmonary artery pressure, decrease in cardiac output and in mixed pulmonary venous oxygen saturation especially) or to diagnose the reason for failure of weaning ECMO or discontinuing iNO at the end of the surgical procedure.

Tranexamic acid is administered as a bolus dose of 30 mg/kg followed by a continuous infusion of 5 mg/kg/h. Cell salvage is used except in patients with cystic fibrosis.

A protective ventilation strategy is used after the first graft is implanted. The protocol is as follows: (i) a tidal volume of 5–6 ml/kg ideal body weight to reach a maximum plateau pressure of 30 cm H_2O; (ii) the respiratory rate adjusted to maintain arterial pH in a normal range (7.38–7.42) and (iii) positive end expiratory pressure (PEEP) maintained between 5 and 10 cm H_2O.

Inhaled nitric oxide protocol (iNO)

NO is delivered using OptiKINOX™, which was integrated into a mobile, ready-to-use treatment station consisting of two tanks containing a 225 ppm gas mixture NO (Air Liquide, 75321 Paris, France). OptiKINOX™allows NO to be sequentially administered during inspiration via the inspiratory limb of the ventilator to achieve a targeted dose of iNO.

In our series, iNO was administered in all cases at an initial concentration of 10 ppm after verification of the adequate positioning of the double lumen tube and beginning of ventilation of the native lungs and then through surgery. iNO was never stopped before the end of the procedure. Intravenous or inhaled prostaglandin is not used at our center.

ECMO management

Veno-arterial ECMO, mostly peripherally, was inserted during surgery in cases of a hemodynamic disturbance refractory to standard medical treatment such as after pulmonary artery clamping or respiratory failure refractory to ventilation optimization (i.e., major hypercarbia with significant respiratory acidosis or refractory hypoxemia).

End of the procedure

Patient care procedure before transfer to ICU is described in Figure 1. It comprised three successive trials: the first one to remove ECMO, the second to stop iNO and the third to extubate the patient in the operating theater.

Regarding ECMO, a positive weaning test was defined as (i) a stable mean arterial pressure above 60 mmHg with a minimal dose of vasopressors after reducing ECMO blood flow and (ii) a PaO2/FiO2 ratio >200 with FiO2 = 40% on ECMO and FiO2 < 50% on the ventilator. TEE was performed during this trial to look for a possibly treatable cause of failure: low filling state, right ventricular dysfunction (end-diastolic and end-systolic surfaces with a dilated right ventricle with a fractional area change <35%, interventricular septum curvature with the RV/LV ratio, reduced tricuspid annular plane systolic excursion, severe tricuspid regurgitation and backflow in the inferior vena cava and in the sus-hepathic veins, ...) or left ventricular dysfunction (global and segmental left ventricular systolic function). Moreover, a restricted or kinked or thrombotic vascular anastomosis is searched for whether it is a pulmonary artery anastomosis, which can lead to a right ventricular dysfunction, or a venous anastomosis, which can induce lung edema and hypoxemia. TEE could also find some unexpected causes such as heart compression from pleural fluid or pericardial fluid, a dynamic right ventricular outflow tract obstruction (the so-called "suicide right ventricle"). Finally, TEE allows also a qualitative evaluation of the right to left shunt due to a patent foramen ovale, for a better interpretation of a hypoxemic state. Patients were kept under ECMO if they did not pass the ECMO weaning trial (patients excluded from the analysis).

In all other cases, an iNO weaning trial was performed when the PaO2/FiO2 ratio was greater than 100. A failure was defined as a substantial decrease in PaO2 or as a hemodynamic worsening shown by Swan-Ganz parameters and on TEE which searched especially for an eventual right to left shunt due to a foramen ovale for a better interpretation of a hypoxemic state and a right ventricular dysfunction using the same criterion as during the ECMO cessation trial. In case of failure, iNO was reintroduced, and the patient was transferred to the ICU under mechanical ventilation.

If the iNO weaning test was successful and if the patient was deemed stable (i.e. hemodynamic stability, no active bleeding, etc.), the next step was to extubate the patient and attempt non-invasive ventilation (NIV) if the PaO_2/FiO_2 ratio was greater than 200. If extubation was well-tolerated, the patient was transferred to the ICU on oxygen therapy (via a high oxygen concentration face-mask). If an acute respiratory failure was observed after extubation and despite NIV, tracheal

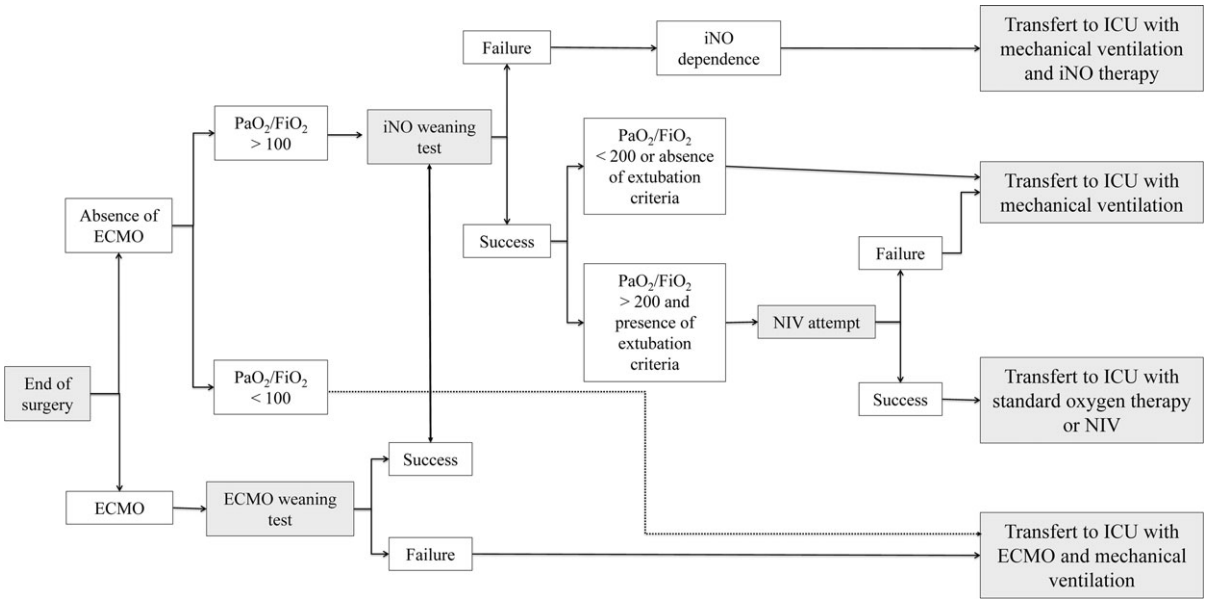

Figure 1 Protocol used for patient care. ECMO, extracorporeal membrane oxygenation; iNO, inhaled nitric oxide; NIV, non-invasive ventilation; ICU, intensive care unit.

intubation and mechanical ventilation were performed in the OR before transfer to the ICU.

ICU management

All extubated patients were placed on NIV for at least 6 h following ICU admission. Initial settings for inspiratory and expiratory airway pressures were 8 and 4 cmH_2O, respectively, with a FiO_2 of 1. NIV settings were then adjusted according to clinical judgment and arterial blood gases. During the following 24 h, NIV was performed for at least 1 h every 4 h and was later adjusted to the patient's respiratory status. Grade 3 PGD was defined as a PaO_2/FiO_2 ratio <200 at 72 h after transplantation in a patient showing no evidence of pulmonary infection or any other specific cause.

Data collection

Data related to patient and donor characteristics, durations of lung ischemia and surgery, hemodynamic parameters, arterial blood gases and transfusion of intraoperative ECMO were prospectively collected. Postoperative data were retrospectively collected from electronic medical records. The database was created as a FileMaker Pro file (FileMaker Company, Santa Clara, CA 95054). This file was encrypted and was accessible only on the local intranet with a password. All data were fully anonymized before they were extracted for statistical analysis.

Study goals and outcomes

The patients were separated into the following two groups: iNO non-dependent patients and iNO-dependent patients. The latter group included patients who could not be weaned from iNO at the end of the surgical procedure according to the criteria defined above.

The primary goal of the study was to identify the risk factors for iNO dependency. Secondary goals were to assess whether iNO dependency is associated with Grade 3 PGD, ICU and hospital length of stay and postoperative mortality.

As describe above, patients who required postoperative ECMO were excluded from the analysis. Nevertheless, we compare them to the study cohort. Data are shown in Supporting Information.

Statistical analysis

Categorical variables are described as counts (percentages) and were compared using the Fisher exact test. Continuous variables are described as medians (25th–75th percentiles) and were compared using the Mann–Whitney test.

The impact of iNO dependency on mortality was first examined by plotting actuarial survival curves using the Kaplan–Meier estimator. Patients who were still alive were right-censored at day 1500. No patient was lost to follow-up during this period.

To account for potential confounding-by-indication arising from the observational design, the impact of iNO dependency on postoperative outcomes was estimated using a propensity score approach. Specifically, we used the inverse probability weighting estimator to estimate the average treatment effect [14]. Propensity score was estimated using gradient boosting, a method that has previously proven useful for improving the consistency of propensity score estimators because it optimizes balance across baseline confounders between treatment groups [15]. The following variables were included in the propensity score model: age, body mass index, emergency surgery, time on waiting list, underlying lung disease, preoperative pulmonary hypertension, pulmonary reduction, donor score, lung ischemic time, amount of intraoperative fluid, estimated intraoperative blood loss, norepinephrine support at the end of the procedure and blood lactate at the end of the procedure. Propensity score was estimated using gradient boosting as suggested by McCaffrey *et al.* [15]. Balance across confounders was estimated using the standardized mean difference (SMD). In the weighted population, the impact of iNO dependency was estimated using a weighted log-rank test for mortality, and a weighted Cox Proportional Hazard model. A generalized linear model for complex survey designs was also used with a Poisson link for relative risks or a Gaussian link for differences in means. All confounders with a residual imbalance after inverse probability of treatment weighting (i.e. SMD > 10%) were adjusted for the multivariable models. Standard errors were estimated using a robust sandwich estimator accounting for weighting-induced correlation.

A lasso penalized logistic regression model was used to identify the risk factors for iNO dependency. The hyperparameter lambda was optimized using 10-fold cross-validation. Ninety-five per cent confidence intervals were computed using the method described by Taylor *et al.* [16]; this method is implemented in the selectiveInference R package [17].

All statistical analyses were performed in R (version 3.3.2 for Macintosh, licenses GNU GPL, The R foundation for statistical computing, Vienna, Austria) running on a MacOsX platform. A *P*-value <0.05 was considered to define statistical significance.

Results

Between 1 January 2012 and 1 January 2016, a total of 237 patients underwent DLT at our institution. After excluding the patients who did not meet the study criteria, 213 patients were assessed. Ninety-nine of these patients required an ECMO during the surgical procedure, and among them, 40 could not be weaned from ECMO at the end of the procedure (18.8% of all cases). Finally, 173 patients underwent an iNO weaning test at the end of the surgical procedure and 17 of them could not be weaned from iNO (9.8% of all cases) (Figure 2).

The most frequent diagnosis of failure to wean iNO was severe hypoxemia in seven cases; none were related to a significant right to left shunt due to a patent foramen ovale. Other reasons for iNO weaning failure were the association of hypoxemia and of pulmonary hypertension in six cases and right ventricular dysfunction in four cases (due to air embolism or increased grafted lungs arterial resistance since pulmonary artery anastomotic narrowing was never observed).

Patient characteristics

Patient characteristics are illustrated in Table 1. Pulmonary fibrosis was more frequent in iNO-dependent patients ($P = 0.007$, Table 1).

Intraoperative profiles

Intraoperative data are illustrated in Table 2. Ischemic times were similar between groups. iNO-dependent patients presented more frequently an hemodynamic or respiratory refractory failure having required intraoperative ECMO ($P = 0.005$) (Table 2).

Risk factors for iNO dependency

In univariate analysis, iNO dependency was more frequently observed in patients with pulmonary fibrosis ($P = 0.01$) (Table 1) and in those needing intraoperative ECMO ($P = 0.01$) (Table 2). In multivariable analysis (Table 3), the body mass index (OR = 2.03, 95% CI = 1.14–3.29, $P = 0.02$) and the need for intraoperative ECMO (OR = 1.80, 95% CI = 1.02–2.72, $P = 0.04$) were independent risk factors for iNO dependency.

Postoperative outcomes

Postoperative outcome is illustrated in Table 4 with results of the comparisons between iNO-non-dependent

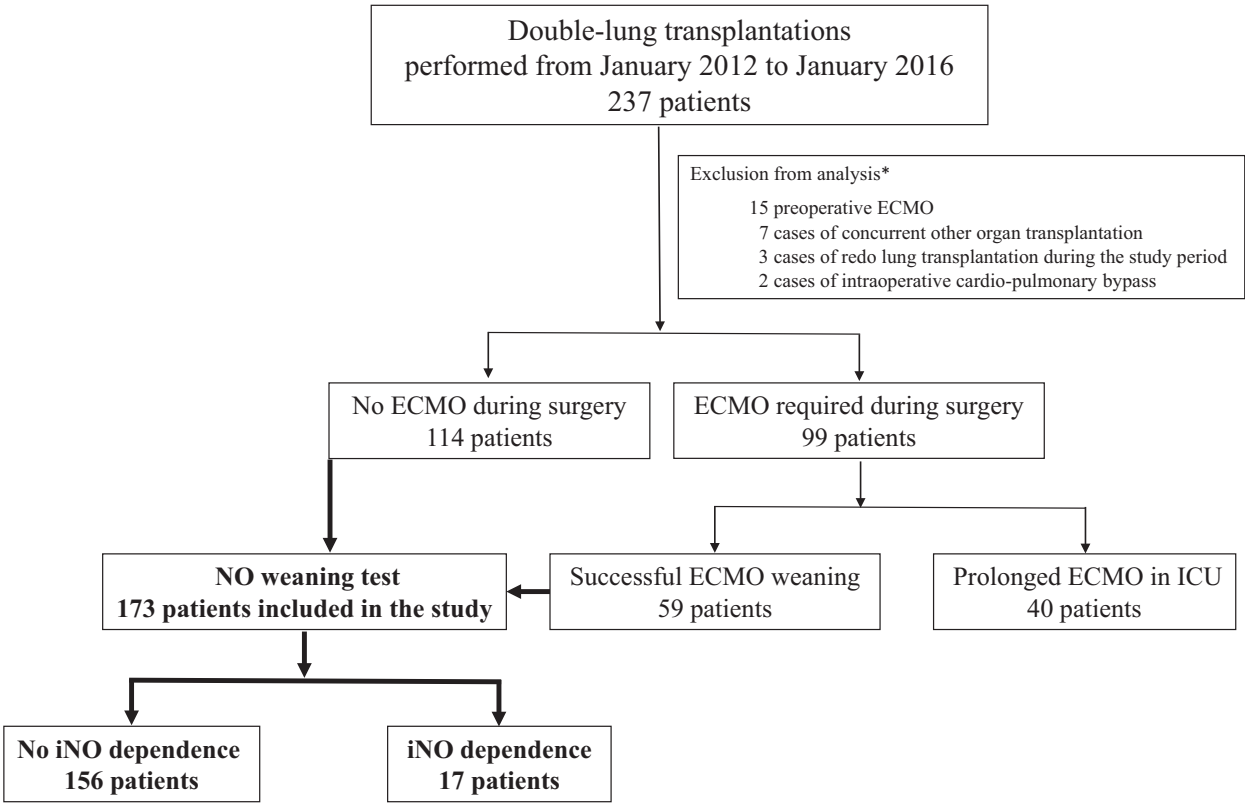

Figure 2 Flow chart. *Some patients were excluded for more than one reason.

patients and iNO-dependent patients using Fisher exact test or Mann–Whitney test as appropriate. In crude analysis, iNO dependency was associated with a significantly higher prevalence of Grade 3 PGD (crude Odds Ratio = 5.90, 95% CI = 1.54–21.15, $P = 0.004$) and higher mortality during the first 1500 days following transplantation (crude Hazard Ratio = 3.46, 95% CI = 1.37–8.73, $P = 0.008$). Covariate balance before and after inverse probability of treatment weighting is illustrated in the Appendix. In the weighted population, iNO dependency was associated with a higher prevalence of Grade 3 PGD (adjusted RR = 4.20, 95% CI = 1.75–10.09, $P < 0.001$). iNO dependency was also associated with a longer ICU length of stay (adjusted difference in means = 5.97 days, 95% CI = 2.36–9.58, $P < 0.001$). In the weighted population, iNO dependency was also associated with lower survival during the first 1500 days of follow-up (weighted log-rank test: $P = 0.006$; adjusted HR = 5.0, 95% CI = 1.86–13.48, $P < 0.001$) (Figure 3).

Eleven patients were iNO dependent after having had an intraoperative ECMO, three of them died during the 1-year follow-up. Forty patients required a postoperative ECMO, 10 of them died during the same period. The difference is not statistically significant ($P = 0.88$).

Discussion

The major findings of this study are that: (i) iNO dependency is observed in approximately 10% of lung-transplanted patients, (ii) body mass index and intraoperative ECMO seem to be risk factors for iNO dependency and (iii) iNO dependency is associated with grade 3 PGD and mortality.

Recently, Benedetto *et al.* [18] described the potential benefits of iNO in lung-transplanted patients and defined iNO as a "master reperfusion anesthetic". This refers to its multiple effects, including selective pulmonary vasodilation [19], better management of reperfusion [20], optimization of the ventilation-perfusion ratio, protection from inflammatory insults in the allograft [5,6] and the inhibition of ischemic reperfusion injury-induced apoptosis [7,21]. However, in a systematic review and meta-analysis on PGD risk factors after lung transplantation, Liu *et al.* [22] concluded in favor of a lack of association between intraoperative iNO and the development of PGD (OR 1.09, 95% CI 0.68–1.74, $P = 0.72$). In fact, systematic prophylactic use of iNO remains controversial [8,23] and there is no study evaluating the iNO dependency phenomena and its implication on patient outcome.

Table 1. Patient and donor characteristics

Variables	All patients (n = 173)	iNO-non-dependent patients (n = 156)	iNO-dependent patients (n = 17)	P
Demographic data and past medical history				
Age (years)	43 [28–55]	44 [29–55]	36 [23–51]	0.24
Male gender	88 (51)	78 (50)	10 (59)	0.49
BMI (kg/m^2)	18.9 [17.6–22.4]	18.9 [17.5–21.9]	19.0 [18.6–28.5]	0.13
Diabetes	54 (31)	50 (32)	4 (23)	0.47
Previous thoracic surgical procedure	37 (21)	35(22)	2 (12)	0.53
High emergency lung transplantation	12 (7)	11 (7)	1 (6)	1
Time on waiting list (days)	25 [11–60]	26 [11–60]	20 [13–56]	0.88
Transplant indication				
Cystic fibrosis	82 (47)	75 (48)	7 (41)	0.59
COPD	44 (25)	42 (27)	2 (12)	0.25
Pulmonary fibrosis	27 (16)	20 (13)	7 (41)	0.007
PPH	0	0	0	1
Redo transplantation	5 (3)	5 (3)	0	1
Other pathology	20 (11)	19 (12)	1 (6)	0.70
Preoperative status				
PAH	76 (44)	67 (43)	9 (53)	0.43
Hospitalized in ICU	16 (9)	14 (9)	2 (12)	0.66
On mechanical ventilation	4 (2)	3 (2)	1 (6)	0.34
Ex vivo procedure	25 (14)	23 (15)	2 (12)	1
Immediate preoperative plasmapheresis	52 (30)	48 (31)	4 (23)	0.54
Donor characteristics				
Men	93 (54)	82 (53)	11 (65)	0.34
Age (years)	51 [39–61]	49 [38–61]	55 [48–60]	0.39
Smoking history (pack-years)	0 [0–15]	0 [0–10]	15 [0–20]	0.005
PaO_2/FIO_2 ratio	364 [309–435]	369 [310–436]	332 [299–405]	0.26
Oto score	6 [4–8]	6 [4–8]	7 [5–8]	0.22

Data are presented as the median [interquartile range] or as a number (percentage).

iNO, inhaled nitric oxide; BMI, body mass index; COPD, chronic obstructive pulmonary disease; PPH, primary pulmonary hypertension; PAH, preoperative pulmonary hypertension; ICU, intensive care unit; PaO_2/FIO_2 ratio, arterial partial pressure of oxygen/inspired fraction of oxygen.

Table 2. Intraoperative variables

Variables	All patients (n = 173)	iNO - non - dependent patients (n = 156)	iNO - dependent patients (n = 17)	P
Intraoperative characteristics				
Ischemic time for the first graft (min)	279 [222–344]	280 [223–346]	255 [220–330]	0.59
Ischemic time for the second graft (min)	405 [347–479]	407 [347–490]	374 [354–456]	0.59
Absence of pulmonary resection	124 (72)	112 (72)	12 (71)	1
Volume loading (l)	3.0 [2.3–3.5]	3.0 [2.2–3.5]	3.0 [2.5–4.0]	0.57
Bleeding (l)	1.1 [0.8–1.7]	1.1 [0.8–1.6]	1.2 [0.8–1.9]	0.48
Packed red blood cells (units)	4 [3–6]	4 [3–6]	5 [4–5]	0.40
Fresh frozen plasma (units)	4 [3–6]	4 [3–6]	4 [4–7]	0.36
Platelets (units)	0 [0–0]	0 [0–0]	0 [0–0]	0.16
Intraoperative ECMO	59 (34)	48 (31)	11 (65)	0.005
Variables measured at the end of the surgical procedure				
Hemoglobin (gm/dl)	11.4 [10.7–12.0]	11.3 [10.7–12.0]	11.6 [10.9–12.6]	0.30
PaO_2/FIO_2 ratio	293 [189–387]	300 [210–394]	157 [126–193]	<0.001
Norepinephrine, μg/kg/min	8.3 [3.8–16.7]	6.7 [3.3–15.0]	13.3 [5.0–24.6]	0.10
Lactate level (mmol/l)	2.2 [1.6–3.1]	2.1 [1.6–3.0]	2.4 [2.1–4.0]	0.33

Data are presented as the median [interquartile range] or as a number (percentage).

iNO, inhaled nitric oxide; ECMO, extracorporeal membrane oxygenation; PaO_2/FIO_2 ratio, arterial partial pressure of oxygen/inspired fraction of oxygen.

Table 3. Multivariate analysis of risk factors for iNO dependency

Variables	Odds ratio for iNO dependency	95% Confidence interval	*P*
Age	0.49	0.70–3.30	0.13
Body mass index	2.03	1.14–3.29	0.02
Oto score	1.42	0.03–1.87	0.71
Intraoperative ECMO	1.80	1.02–2.72	0.04

ECMO, extracorporeal membrane oxygenation; iNO, inhaled nitric oxide.

Our group systematically administers iNO at an initial concentration of 10 ppm either at the beginning of the procedure in cases of pulmonary artery hypertension or before reperfusion of the first graft. At the end of the procedure, iNO weaning is systematically attempted. In the present cohort, nearly 10% of the patients could not be weaned from iNO at the end of the surgical procedure. Our results show that the risk of iNO dependency increases with body mass index and intraoperative use of ECMO.

Several studies have found an association between recipient increased body mass index and increased risk of PGD. Most of them have been analyzed in two successive systematic reviews and meta-analyses [22,24]; others have been described more recently [25–27]. Moreover, Shah *et al.* [28] developed prediction models for PGD and showed that patients with a normal BMI have a lower risk of PGD and Upala *et al.* [24] reported an increased risk of mortality after lung transplantation in recipients with underweight or obesity. By contrast, to the best of our knowledge, there is no study on the relation between body mass index and NO administration in the field of lung transplantation and we can only speculate that the increased risk of iNO dependency associated with the increase in body mass index could be due to the relation between permanent inflammatory state, due to obesity and leptin-mediated lung inflammation and the production of pro-inflammatory cytokines from adipose tissues and macrophages [26,29].

The relationship between intraoperative ECMO and iNO dependency has never been previously described but this seems logical since hypoxemia due to graft dysfunction is a frequent indication for intraoperative ECMO. The ECMO withdrawal trial at the end of the procedure was satisfactory in the patients analyzed in this study but some degree of graft dysfunction might persist explaining a significant effect of iNO and consequently iNO dependency. iNO dependency can thus be considered as an early sign of PGD, which may start during the surgical procedure. This is consistent with an experimental study that showed that the first phase of lung injury occurs within 30 min after reperfusion [30]. Accordingly, Pottecher *et al.* [31] recently demonstrated that extravascular lung water measured after unclamping of the second graft and elevated plasma concentrations of epithelial biomarkers may predict the occurrence of Grade 3 PGD.

In our series, iNO dependency was associated with higher prevalence of Grade 3 PGD and higher mortality. If we assume that iNO dependency is an early sign of PGD, it is not surprising that iNO dependency predicted bad outcome. Samano *et al.* [32] reported 177.7 mean survival days for patients with Grade 3 PGD and 1628.9 days for the other patients ($P < .001$). The meta-analysis performed by Liu *et al.* [22] showed a 3.95-fold (95% CI 2.80–5.57) increased risk of short-term mortality in patients with PGD. More recently, Sabashnikov *et al.* [33] identified the low PaO2/FiO2-ratio at 72 h after surgery as an independent predictor for 1-year mortality (95% CI: 0.988–0.999; $P = 0.024$).

Our study has some limitations, first, it is a single center retrospective study characterized by a particular intraoperative management: systematic iNO administration throughout the surgical procedure with an abrupt cessation which can expose to a rebound effect [34], exclusive intraoperative use of a veno-arterial ECMO, the indication of a veno-venous ECMO being discussed secondarily in case of isolated postoperative respiratory failure, ECMO and/or iNO weaning trials at the end of the procedure and immediate extubation in selected cases [10]. Second, iNO dependency was defined by the practitioner in charge of the patient. Third, we used TEE systematically and if necessary epicardic echocardiography [13] to search for a cause of ECMO and/or iNO negative weaning trials but it must be recognized that very often the analysis is more qualitative, i.e. visual estimation, than quantitative given the limited time for analysis, the sometimes average or mediocre quality of imaging, fluid fluctuations and rapid hemodynamic changes and occurrence of arrhythmias. However, most treatment decisions are based on major signs that qualitative analysis can reveal. Finally, another

Transplant International 2019; 32: 244–256

Table 4. Postoperative complications

Variables	All patients (n = 173)	iNO-non-dependent patients (n = 156)	iNO-dependent patients (n = 17)	P
Respiratory complications				
PGD 3 at 0 h	47 (27.2)	34 (21.8)	13 (76.5)	<0.001
PDG 3 at 24 h	30 (17.3)	23 (14.7)	7 (41.2)	0.01
PGD 3 at 48 h	32 (18.5)	24 (15.4)	8 (47.1)	0.004
PGD 3 at 72 h	19 (11)	13 (8.3)	6 (35.3)	0.005
Mechanical ventilation (days)	0 [0–3]	0 [0–2]	5 [2–11]	<0.001
Secondary intubation	22 (12.7)	19 (12.2)	3 (17.6)	0.46
Tracheostomy	26 (15)	20 (12.8)	6 (35.3)	0.03
Secondary ECMO	8 (4.6)	5 (3.2)	3 (17.6)	0.03
Reoperation for bleeding	10 (5.8)	6 (3.8)	4 (23.5)	0.009
Other complications				
Atrial fibrillation	38 (21.9)	33 (21.2)	5 (29.4)	0.54
Hemodialysis	4 (2.3)	3 (1.9)	1 (5.9)	0.34
Stroke	3 (1.7)	2 (1.3)	1 (5.9)	0.27
Thrombo-embolic complication	17 (9.8)	14 (9)	3 (17.6)	0.22
Lengths of stay				
In ICU (days)	5 [4–9]	5 [4–8]	11 [6–18]	<0.001
In Hospital (days)	29 [23–39]	29 [23–39]	30 [25–77]	0.21
Mortality				
Hospital mortality	5 (2.9)	4 (2.6)	1 (5.9)	0.41
At 30 days	2 (1.2)	1 (0.6)	1 (5.9)	0.19
At 90 days	5 (3.0)	4 (2.6)	1 (5.9)	0.41
At 1 year	11 (6.4)	8 (5.1)	3 (17.6)	0.08
At 3 years	24 (13.9%)	19 (12.2%)	5 (29.4%)	0.06

Data are presented as median [interquartile range] or as a number (percentage).

iNO, inhaled nitric oxide; ECMO, extracorporeal membrane oxygenation; ICU, Intensive care unit; PGD, pulmonary graft dysfunction.

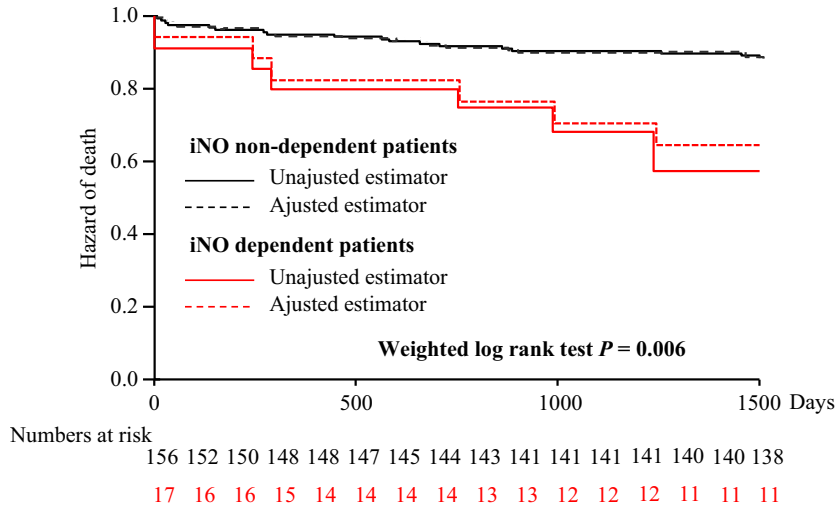

Figure 3 Weighted population survival curves (p values were generated from weighted log-rank tests).

limitation could have been our definition of PGD based on a PaO2/FiO2 ratio <200 at 72 h after transplantation and not on the ratio within 72 h of the intervention as usually used. However, our definition corresponds to the most recent definition of the PGD [35].

In conclusion, we observed in 9.8% of cases that iNO could not be interrupted at end-surgery stage after a rigorous weaning protocol. iNO dependency appears to be a very early marker of short-term outcome with an increased risk of PGD and of long-term poor outcome with an increased mortality at 1500 days. Although we observed such significant associations, we still cannot conclude on causation since the presence of unobserved confounders cannot be ruled out.

Acknowledgements

The authors acknowledged the Foch Lung Transplant Group: the surgical team: Pierre Bonnette, Alain Chapelier, Julien De Wolf, Philippe Puyo, Nicolas Salley, Delphine Mitilian and Matthieu Glorion; the intensive care unit team: Vincent Caille, Jérôme Devaquet, Anne Gaelle Si Larbi, Alexis Soummer and Grégoire Trebbia; the respiratory intermediate care unit team : Elise Cuquemelle, François Parquin and Benjamin Zuber; the anesthesiology team: Isabelle Aimé, Valentina Assenzo, Beatrice Angemont, Olivier Belze, Marie Binczak, Julie Bresson, Camille Cornet, Virginie Dumans-Nizard, Jean Louis Dumoulin, Sofian Faiz, Marie-Louise Felten, Sebastien Ghiglione, Sébastien Jacqmin, Laure Lamothe, Ewan Lepage, Léa Ley, Ngai Liu, Thibaut Mariaux de Serres, Mireille Michel-Cherqui;,Olivier Pruszkowski, Adrian Radu, Bertrand Rives, Mai Anh Céline Roussel, Barbara Szekely, Benoit Vandenbunder and Nicolas Verroust; and the pneumology team: Sandra De Miranda, Benoit Douvry, Dominique Grenet, Abdul Monem Hamid, Clement Picard and Marc Stern.

Authorship

JF, MG, MF and MLG: Research design, Data analysis, Writing and supervising the paper, Approval of article. RP: Research design, Data analysis, Statistics, Writing and supervising the paper, Approval of article. J-YM: Research design, Data collection, Data analysis, Writing the paper, Approval of article. JT, ES, AR, FP and CC: Research design, Data analysis, Writing the paper, Approval of article.

Funding

The Hôpital Foch supported the work submitted and had no role in study design, running and reporting of this study.

Conflict of interest

The authors of the manuscript have no conflicts of interest to disclose.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Survival analysis using Kaplan–Meier curve according to the three groups.

Table S1. Patient and donor characteristics.
Table S2. Intraoperative variables.
Table S3. Postoperative complications.

Appendix S1. Comparison of iNO non-dependent patients, iNO dependent patients and of patients requiring prolonged ECMO after surgery.

REFERENCES

1. Rossetti M, Guenard H, Gabinski C. Effects of nitric oxide inhalation on pulmonary serial vascular resistances in ARDS. *Am J Respir Crit Care Med* 1996; **154**: 1375.
2. Bhatraju P, Crawford J, Hall M, Lang JD Jr. Inhaled nitric oxide: current clinical concepts. *Nitric Oxide* 2015; **50**: 114.
3. Kemming GI, Merkel MJ, Schallerer A, *et al.* Inhaled nitric oxide (NO) for the treatment of early allograft failure after lung transplantation. Munich Lung Transplant Group. *Intensive Care Med* 1998; **24**: 1173.
4. Ardehali A, Laks H, Levine M, *et al.* A prospective trial of inhaled nitric oxide in clinical lung transplantation. *Transplantation* 2001; **72**: 112.
5. Shah RJ, Bellamy SL, Localio AR, *et al.* A panel of lung injury biomarkers enhances the definition of primary graft dysfunction (PGD) after lung transplantation. *J Heart Lung Transplant* 2012; **31**: 942.
6. Suzuki Y, Cantu E, Christie JD. Primary graft dysfunction. *Semin Respir Crit Care Med* 2013; **34**: 305.
7. Fischer S, Maclean AA, Liu M, *et al.* Dynamic changes in apoptotic and necrotic cell death correlate with severity of ischemia-reperfusion injury in lung transplantation. *Am J Respir Crit Care Med* 2000; **162**: 1932.
8. Ramadan ME, Shabsigh M, Awad H. Con: inhaled pulmonary vasodilators are not indicated in patients undergoing lung transplantation. *J Cardiothorac Vasc Anesth* 2017; **31**: 1127.
9. Pasque MK, Cooper JD, Kaiser LR, Haydock DA, Triantafillou A, Trulock EP. Improved technique for bilateral lung transplantation: rationale and initial clinical experience. *Ann Thorac Surg* 1990; **49**: 785.
10. Felten ML, Moyer JD, Dreyfus JF, *et al.* Immediate postoperative extubation in bilateral lung transplantation: predictive factors and outcomes. *Br J Anaesth* 2016; **116**: 847.
11. Lang RM, Bierig M, Devereux RB, *et al.* Recommendations for chamber quantification: a report from the American Society of Echocardiography's Guidelines and Standards Committee and the Chamber Quantification Writing Group, developed in conjunction with the European Association of Echocardiography, a branch of the European Society of Cardiology. *J Am Soc Echocardiogr* 2005; **18**: 1440.
12. Rudski LG, Lai WW, Afilalo J, *et al.* Guidelines for the echocardiographic assessment of the right heart in adults: a report from the American Society of Echocardiography endorsed by the European Association of Echocardiography, a registered branch of the European Society of Cardiology, and the Canadian Society of Echocardiography. *J Am Soc Echocardiogr* 2010; **23**: 685.
13. Felten ML, Michel-Cherqui M, Sage E, Fischler M. Transesophageal and contact ultrasound echographic assessments of pulmonary vessels in bilateral lung transplantation. *Ann Thorac Surg* 2012; **93**: 1094.
14. Mansournia MA, Altman DG. Inverse probability weighting. *BMJ* 2016; **352**: i189.
15. McCaffrey DF, Ridgeway G, Morral AR. Propensity score estimation with boosted regression for evaluating causal effects in observational studies. *Psychol Methods* 2004; **9**: 403.
16. Taylor J, Tibshirani R. Post-selection inference for l1-penalized likelihood models. *Can J Stat* 2018; **46**: 41.
17. Tools for Post-Selection Inference. R package version 1.2.4. https://CRAN.R-project.org/package=selectiveInference. Assessed on November 1, 2018
18. Benedetto M, Romano R, Baca G, *et al.* Inhaled nitric oxide in cardiac surgery: evidence or tradition? *Nitric Oxide* 2015; **49**: 67.
19. Frostell C, Fratacci MD, Wain JC, Jones R, Zapol WM. Inhaled nitric oxide. A selective pulmonary vasodilator reversing hypoxic pulmonary vasoconstriction. *Circulation* 1991; **83**: 2038.
20. Pasero D, Martin EL, Davi A, Mascia L, Rinaldi M, Ranieri VM. The effects of inhaled nitric oxide after lung transplantation. *Minerva Anestesiol* 2010; **76**: 353.
21. Meyer KC, Love RB, Zimmerman JJ. The therapeutic potential of nitric oxide in lung transplantation. *Chest* 1998; **113**: 1360.
22. Liu Y, Liu Y, Su L, Jiang SJ. Recipient-related clinical risk factors for primary graft dysfunction after lung transplantation: a systematic review and meta-analysis. *PLoS ONE* 2014; **9**: e92773.
23. Bhandary S, Stoicea N, Joseph N, Whitson B, Essandoh M. Pro: inhaled pulmonary vasodilators should be used routinely in the management of patients undergoing lung transplantation. *J Cardiothorac Vasc Anesth* 2017; **31**: 1123.
24. Upala S, Panichsillapakit T, Wijarnpreecha K, Jaruvongvanich V, Sanguankeo A. Underweight and obesity increase the risk of mortality after lung transplantation: a systematic review and meta-analysis. *Transpl Int* 2016; **29**: 285.
25. Cottini SR, Ehlers UE, Pagnamenta A, *et al.* Pretransplant dyslipidaemia influences primary graft dysfunction after lung transplantation. *Interact Cardiovasc Thorac Surg* 2016; **22**: 402.
26. Moon S, Park MS, Lee JG, *et al.* Risk factors and outcome of primary graft dysfunction after lung transplantation in Korea. *J Thorac Dis* 2016; **8**: 3275.
27. Fernandez R, Safaeinili N, Kurihara C, *et al.* Association of body mass index with lung transplantation survival in the United States following implementation of the lung allocation score. *J Thorac Cardiovasc Surg* 2018; **155**: 1871.
28. Shah RJ, Diamond JM, Cantu E, *et al.* Objective estimates improve risk stratification for primary graft dysfunction after lung transplantation. *Am J Transplant* 2015; **15**: 2188.
29. Lederer DJ, Kawut SM, Wickersham N, *et al.* Obesity and primary graft dysfunction after lung transplantation: the Lung Transplant Outcomes Group Obesity Study. *Am J Respir Crit Care Med* 2011; **184**: 1055.
30. Carter YM, Gelman AE, Kreisel D. Pathogenesis, management, and consequences of primary graft dysfunction. *Semin Thorac Cardiovasc Surg* 2008; **20**: 165.
31. Pottecher J, Roche AC, Degot T, *et al.* Increased extravascular lung water and plasma biomarkers of acute lung injury precede oxygenation impairment in primary graft dysfunction after lung transplantation. *Transplantation* 2017; **101**: 112.

32. Samano MN, Fernandes LM, Baranauskas JC, *et al.* Risk factors and survival impact of primary graft dysfunction after lung transplantation in a single institution. *Transplant Proc* 2012; **44**: 2462.
33. Sabashnikov A, Weymann A, Mohite PN, *et al.* Risk factors predictive of one-year mortality after lung transplantation. *Eur J Cardiothorac Surg* 2014; **46**: e82.
34. Lavoie A, Hall JB, Olson DM, Wylam ME. Life-threatening effects of discontinuing inhaled nitric oxide in severe respiratory failure. *Am J Respir Crit Care Med* 1996; **153**: 1985.
35. Snell GI, Yusen RD, Weill D, *et al.* Report of the ISHLT Working Group on Primary Lung Graft Dysfunction, part I: definition and grading-A 2016 Consensus Group statement of the International Society for Heart and Lung Transplantation. *J Heart Lung Transplant* 2017; **36**: 1097.

APPENDIX

Standardized mean differences before and after weighting