



Acute effects of the combination of sildenafil and inhaled treprostinil on haemodynamics and gas exchange in pulmonary hypertension

Robert Voswinckel, Frank Reichenberger, Beate Enke, Andre Kreckel, Stefanie Krick, Henning Gall, Ralph Theo Schermuly, Friedrich Grimminger, Lewis J. Rubin, Horst Olschewski, et al.

► To cite this version:

Robert Voswinckel, Frank Reichenberger, Beate Enke, Andre Kreckel, Stefanie Krick, et al.. Acute effects of the combination of sildenafil and inhaled treprostinil on haemodynamics and gas exchange in pulmonary hypertension. *Pulmonary Pharmacology & Therapeutics*, 2008, 21 (5), pp.824. 10.1016/j.pupt.2008.07.003 . hal-00499160

HAL Id: hal-00499160

<https://hal.science/hal-00499160>

Submitted on 9 Jul 2010

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Author's Accepted Manuscript

Acute effects of the combination of sildenafil and inhaled treprostinil on haemodynamics and gas exchange in pulmonary hypertension

Robert Voswinckel, Frank Reichenberger, Beate Enke, Andre Kreckel, Stefanie Krick, Henning Gall, Ralph Theo Schermuly, Friedrich Grimminger, Lewis J. Rubin, Horst Olschewski, Werner Seeger, Hossein A. Ghofrani

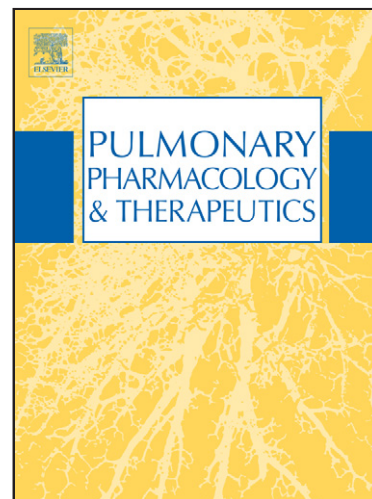
PII: S1094-5539(08)00073-4
DOI: doi:10.1016/j.pupt.2008.07.003
Reference: YPUPT 852

To appear in: *Pulmonary Pharmacology & Therapeutics*

Received date: 25 February 2008
Revised date: 6 May 2008
Accepted date: 6 July 2008

Cite this article as: Robert Voswinckel, Frank Reichenberger, Beate Enke, Andre Kreckel, Stefanie Krick, Henning Gall, Ralph Theo Schermuly, Friedrich Grimminger, Lewis J. Rubin, Horst Olschewski, Werner Seeger and Hossein A. Ghofrani, Acute effects of the combination of sildenafil and inhaled treprostinil on haemodynamics and gas exchange in pulmonary hypertension, *Pulmonary Pharmacology & Therapeutics* (2008), doi:10.1016/j.pupt.2008.07.003

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting galley proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.



www.elsevier.com/locate/ypupt

Acute effects of the combination of sildenafil and inhaled treprostinil on haemodynamics and gas exchange in pulmonary hypertension

Robert Voswinckel¹, Frank Reichenberger¹, Beate Enke¹, Andre Kreckel¹, Stefanie Krick^{1,4}, Henning Gall¹, Ralph Theo Schermuly¹, Friedrich Grimminger¹, Lewis J. Rubin², Horst Olschewski^{1,3}, Werner Seeger¹, Hossein A. Ghofrani¹

¹ University of Giessen Lung Center, Department of Internal Medicine, University Hospital Giessen and Marburg GmbH, Giessen, Germany.

² Division of Pulmonary and Critical Care Medicine, University of California, San Diego School of Medicine, La Jolla, California, USA

³ Division of Pulmonology, Medical University Graz, Austria

⁴ Mount Sinai Medical School, New York, USA

Running title: Sildenafil and inhaled treprostinil in PHT

Corresponding author: Robert Voswinckel, M.D.

University Hospital Giessen and Marburg GmbH

Department of Internal Medicine

Klinikstrasse 36

D-35392 Giessen

e-mail: robert.voswinckel@innere.med.uni-giessen.de

Phone: +49-1792923202

Fax: +49-6032705419

Abstract

Background: Inhaled treprostinil was recently developed for the treatment of pulmonary arterial hypertension. We investigated the safety and acute haemodynamic effects of the combination oral sildenafil and inhaled treprostinil in an open label study in patients with precapillary pulmonary hypertension.

Methods and Patients: Inhaled nitric oxide (20 ppm; n=50), sildenafil (50mg; n=50) and inhaled treprostinil (15µg; n=25 or 30µg; n=25) were applied in subsequent order during right heart catheter investigation to consecutive patients with pulmonary arterial hypertension (PAH; n=28), non-operable chronic thromboembolic pulmonary hypertension (CTEPH; n=17) and pulmonary fibrosis associated pulmonary hypertension (n=5).

Results: Inhaled nitric oxide reduced pulmonary vascular resistance (PVR) to $87.3 \pm 5.1\%$ of baseline values, reduced mean pulmonary arterial pressure (PAP) to $89.7 \pm 3.5\%$ and increased cardiac output (CO) to $102.4 \pm 2.9\%$. Sildenafil reduced PVR to $80.1 \pm 5.0\%$, mPAP to 86.5 ± 2.9 and increased CO to $103.8 \pm 3.2\%$. Treprostinil, inhaled one hour after sildenafil, reduced PVR to $66.3 \pm 3.8\%$, mPAP to $77.8 \pm 3.3\%$, and increased CO to $107.1 \pm 3.3\%$ (mean $\pm$ 95% confidence interval). Subgroup analysis showed similar acute haemodynamic effects in PAH and CTEPH patients. Ventilation/perfusion distribution measurement in six patients with pre-existing gas exchange limitations was not changed by sildenafil and treprostinil. Relevant side effects were not observed.

Conclusion: The combination of sildenafil and inhaled treprostinil was well tolerated and induced additive, pulmonary selective vasodilatation in pulmonary hypertension patients. This could be of relevance also for long term treatment of PAH and CTEPH patients.

Key words: prostaglandins; pulmonary arterial hypertension; phosphodiesterase inhibitors; pulmonary heart disease; treprostinil

INTRODUCTION

Pulmonary arterial hypertension (PAH) as well as non-operable chronic thromboembolic pulmonary hypertension (CTEPH) are progressive diseases with a poor prognosis if not specifically treated [1,2]. The development of drugs that target the nitric oxide pathway (phosphodiesterase-5 inhibitors), endothelin receptors (endothelin receptor antagonists) or the prostacyclin pathway (PGI₂ analogues) has led to significant improvement in exercise capacity, quality of life and survival of PAH patients [3-8]. With the exception of inhaled iloprost, these drugs are not approved for treatment of pulmonary hypertension (PHT) in non-operable chronic thromboembolic disease. So far several uncontrolled trials suggested that these drugs may also be beneficial for these patients [9-11].

Despite the therapies at hand, many patients still develop disease progression and are in need of combination therapy and finally lung transplantation [12]. Controlled clinical trials for combinations of the approved drugs are ongoing. Open label combination therapy is already widely used and resulted in better patient improvement than mono-therapy [13-17]. Combination therapy may be complicated by accumulating side effects and by increasing complexity of medical treatment that may lead to non-compliance of the patients. Prostanoids have been widely used for treatment of PAH with excellent results. Potent pulmonary vasodilatation, platelet inhibition and long term anti-remodeling effects have been attributed to prostanoid therapy. The inhalation of iloprost facilitated the first effective non-parenteral prostanoid therapy for PAH, offering a reduced risk profile compared to the i.v. application [3,13,18,19]. However, the frequency of 6-9 inhalations per day and the relatively long inhalation time of 5 to 10 minutes restricts the early use of inhaled prostanoids in current treatment regimens. Further optimisation of inhaled prostanoid treatment should therefore aim for better patient convenience by lower inhalation frequency and quick drug application.

Treprostinil is a prostacyclin analogue with prolonged half life that is approved for treatment of pulmonary arterial hypertension (PAH) via intravenous or subcutaneous infusion [8,20,21]. In addition to the vasodilatory effects it was shown that treprostinil potently reduces vascular smooth muscle cell proliferation [22]. Inhaled treprostinil induces sustained pulmonary vasodilatation and therefore needs only 4 inhalations per day. The inhalation of up to 60µg treprostinil with an ultrasonic nebuliser is done in less than one minute and can be accomplished even in one single breath. It leads to pulmonary selective vasodilation without relevant systemic side effects [23,24]. Long term open label treatment with inhaled treprostinil was noted to be effective and without relevant side effects [23-25].

In this open label study we addressed the safety, tolerability and haemodynamic effects of the combination of sildenafil and inhaled treprostinil.

METHODS AND PATIENTS

All studies were approved by the institutional review board of the University of Giessen. Written informed consent was obtained from all patients. All inhalations were performed with the Optineb ultrasonic nebulizer (Nebutech, Elsenfeld, Germany).

Acute effects of nitric oxide, sildenafil and inhaled treprostinil

A total number of 50 patients with moderate to severe precapillary pulmonary hypertension were investigated. Baseline values were determined 20 minutes after catheterisation. Heart rate, pulmonary and systemic blood pressures were recorded and cardiac output was measured by thermodilution. Blood gases (central venous blood and arterialised earlobe blood) were taken at baseline and during each pharmacological intervention at defined time points. After evaluation of baseline parameters, all patients inhaled 20 ppm nitric oxide for 5 minutes through a mouthpiece

while using a nose clamp (n=50). When haemodynamic values had returned to baseline values after termination of NO-inhalation, 50 mg oral sildenafil was given (n=50). One hour after the sildenafil application the patients were randomly assigned to receive either 15 µg or 30 µg inhaled treprostinil (TRE; n=25 for each dose). Haemodynamic parameters and blood gases were further recorded for 120 minutes after TRE inhalation.

Ventilation-perfusion matching was assessed in 6 patients with impaired oxygenation by use of the multiple-inert-gas elimination technique as described before [26,27]. In brief, six inert gases (sulphur hexafluoride, ethane, cyclopropane, halothane, diethyl ether, acetone) with widely varying blood-gas partition coefficient were continuously infused in a peripheral vein for at least 30 minutes before the first measurement was done to allow for equilibration of the gases in the body compartments. For one measurement, 10 ml blood was drawn in parallel into 50ml gas tight heparinised glass syringes from an arterial line placed in the radial artery and from the right heart catheter tip. In addition, 30 ml exhaled gas collected at the same time point was drawn from a gas reservoir that was heated to 40 ° C to prevent condensation. The blood was then equilibrated in the syringes with 30 ml nitrogen gas at body temperature for 60 minutes. The equilibrated nitrogen and the exhaled gas were further analysed by gas chromatography. Ventilation/perfusion ratios were calculated based on a specific algorithm [28].

Data analysis and statistical procedures

Mean values, standard deviation, standard error of the mean and 95% confidence intervals were calculated. To test for significant differences between groups, the 2 sided t- test was applied. Treatment effects were analysed for significant differences compared to baseline using 2 sided paired t-test.

Drugs

Treprostinil sodium was obtained from LungRX (Silver Spring, USA) as part of a clinical research collaboration. Sildenafil (Viagra®) is commercially available (Pfizer) and was obtained via the institutional pharmacy. Nitric oxide gas (2000ppm in nitrogen) was obtained from Linde AG (Pullach, Germany) and dosed to achieve an inspiratory concentration of 20-40 ppm.

RESULTS

Acute haemodynamic effects of nitric oxide, sildenafil and inhaled treprostinil

Patient characteristics

Patients were 27 female, 23 male, age 57 ± 2.1 years, mean pulmonary arterial pressure (PAP) 48.0 ± 1.8 mmHg, pulmonary vascular resistance (PVR) 838 ± 53 dynes*s*cm⁻⁵, pulmonary capillary wedge pressure (PCWP) 8.8 ± 0.4 mmHg, central venous pressure (CVP) 9.0 ± 0.7 mmHg, cardiac output (CO) 4.1 ± 0.2 l/min, central venous oxygen saturation (SvO₂) 62.6 ± 1.3 mmHg (mean $\pm$ SEM). Disease aetiologies according to the last WHO classification were idiopathic PAH (n=14), PAH other (n=14), CTEPH (n=17) and pulmonary hypertension associated with pulmonary fibrosis (n=5).

Pooled data of all 50 patients

No significant differences in baseline haemodynamics or gas exchange were noted between the two groups that received either 15µg or 30µg treprostinil. Analysis of the pooled data from all 50 patients demonstrated that inhaled nitric oxide, oral sildenafil and add-on inhaled treprostinil changed pulmonary vascular resistance to 87.3 ± 5.1 %, 80.1 ± 5.0 % and 66.3 ± 3.8 % of baseline values, pulmonary arterial pressure to 89.7 ± 3.5 %, 86.5 ± 2.9 % and 77.8 ± 3.3 %, systemic arterial pressure to 100.8 ± 2.2 %, 92.9 ± 2.2 % and 90.8 ± 2.2 %, and cardiac output to

102.4 $\pm$ 2.9 %, 103.8 $\pm$ 3.2 and 107.1 $\pm$ 3.3 %, respectively (mean $\pm$ 95 % confidence interval, baseline = 100%; Figures 1 and 2). The inhalation of treprostinil on top of the sustained effect of sildenafil thus induced a significant additional, pulmonary selective vasodilatation that did not return to the level of maximal sildenafil effect during the 2 hour observation period. Gas exchange was not changed (Figure 2).

Separate data analysis for the group receiving 15 μ g TRE

Nitric oxide decreased PVR to 82.8 $\pm$ 3.9 % of baseline values, 50mg sildenafil reduced PVR to a minimum of 81.2 $\pm$ 5.1 % and add-on treprostinil additionally reduced PVR to 66.6 $\pm$ 3.6 % of baseline. Systemic arterial pressure was reduced to 92.6 $\pm$ 1.0 % by sildenafil and to 90.2 $\pm$ 1.3 % by additional treprostinil (Figure 3).

Separate data analysis for the group receiving 30 μ g TRE

Nitric oxide decreased PVR to 91.8 $\pm$ 3.2 %, 50mg sildenafil reduced PVR to a minimum of 77.9 $\pm$ 2.5 % and add-on treprostinil reduced PVR to 65.7 $\pm$ 2.7 % of baseline. Systemic arterial pressure was reduced to 92.2 $\pm$ 2.4 % by sildenafil and to 89.9 $\pm$ 1.7 % by treprostinil (Figure 3).

Area under the curve for pulmonary vascular resistance

To compare the impact of the two treprostinil doses on haemodynamics, the area under the curve (AUC) was calculated for the treprostinil effect from the beginning of inhalation (values at start of inhalation set as 100%) to the end of the 120 minute observation time for treprostinil by multiplication of PVR reduction and time, divided by the observation time. Inhalation of 15 μ g or 30 μ g treprostinil on top of the sildenafil effect resulted in an AUC for PVR of -14.0 $\pm$ 4.7 % and -9.6 $\pm$ 5.7 %, PAP -6.7 $\pm$ 2.4 % and -7.5 $\pm$ 3.8 %, SAP -2.1 $\pm$ 3.1 % and -3.1 $\pm$ 2.9 %, SVR -

$5.6 \pm 4.5 \%$ and $-3.5 \pm 5.2 \%$, CO $+5.6 \pm 3.4 \%$ and $+1.6 \pm 4.0 \%$ respectively. A dose dependent effect was thus not observed.

Subgroup analysis for PAH and CTEPH patients

The largest subgroups of patients of this study were PAH patients (n=28) and CTEPH patients (n=17). They were separately analysed to demonstrate their response in more detail. The PAH patient characteristics were: f/m 16/12, age 52 ± 2.9 years, mPAP 51.2 ± 2.8 mmHg, PVR 869 ± 79 dynes*sec*cm⁻⁵, CVP 9.1 ± 1.2 mmHg, PAWP 8.8 ± 0.5 mmHg and CO 4.2 ± 0.2 l/min. The CTEPH patient characteristics were: f/m 12/5, age 66 ± 2.3 years, mPAP 46.3 ± 2.6 mmHg, PVR 892 ± 89 dynes*sec*cm⁻⁵, CVP 9.8 ± 1.1 mmHg, PAWP 9.8 ± 0.7 mmHg and CO 3.6 ± 0.3 l/min. The haemodynamic changes induced by either sildenafil alone or by the combination of sildenafil and inhaled treprostinil were comparable in both collectives, demonstrating a favourable acute effect of this drug combination also in CTEPH patients (Figure 4).

The subset of pulmonary fibrosis patients was not separately evaluated.

Side effects of acute vasodilator challenges

Flush was sometimes observed with the PDE-5 inhibitor sildenafil but was not induced or aggravated by the inhaled prostanoid treprostinil. Other typical prostanoid side effects like headache, ankle pain, dizziness, nausea, diarrhoea or drop of systemic blood pressure were not observed. The only side effect caused by treprostinil was mild and transient cough in less than 10% of the acutely challenged patients.

Multiple inert gas elimination technique

The assessment of ventilation/perfusion distribution in the lungs of six patients with gas exchange limitations prior to the measurements showed without specific treatment at baseline $2.3 \pm 1.9\%$ shuntflow and $4.3 \pm 3.5\%$ low V/Q areas (defined by V/Q ratios of 0-0.1). Upon nitric oxide inhalation shunt was $2.5 \pm 2.0\%$ and low V/Q areas $6.5 \pm 4.0\%$. One hour after sildenafil treatment shunt flow was $3.1 \pm 2.9\%$ and low V/Q areas $4.4 \pm 3.9\%$. Thirty minutes following add-on inhaled treprostinil shunt was measured as $3.2 \pm 2.3\%$ and low V/Q areas $4.7 \pm 5.1\%$ (Mean $\pm$ 95% confidence interval). Therefore the changes of shunt fraction and low V/Q areas were not significant for sildenafil as well as for sildenafil and inhaled treprostinil in combination. Of note, there was no difference between the absolutely pulmonary selective agent inhaled NO and the two other substances in terms of ventilation perfusion matching.

DISCUSSION

The treatment options for pulmonary arterial hypertension increased over the recent years due to the development of targeted therapies that induce pulmonary vasodilatation and reduce pulmonary vascular remodelling. Prostanoids have been the first specific treatment at hand and combine several positive effects for treatment of PAH. They are not only potent vasodilators but also reduce platelet aggregation and exert antiproliferative effects [22,29]. The endogenous prostacyclin PGI₂ leads via binding to its cognant receptors to the production of the second messenger cyclic adenosine monophosphate (cAMP) and induces vascular smooth muscle relaxation and vasodilation of the pulmonary arteries [30]. Cyclic AMP generation by prostanoids and selective inhibition of cAMP degradation by phosphodiesterase (PDE) -3/4-inhibition synergistically increase pulmonary vasodilatation [31]. Unfortunately, a selective PDE-3/4 inhibitor is currently not available for clinical use. The application of prostacyclin or its more stable analogues iloprost and treprostinil demonstrated clinical efficacy and improved survival of

patients with precapillary pulmonary hypertension [3,6,8,32]. The phosphodiesterase 5 (PDE-5) comprises the major degradation pathway of cyclic guanosine monophosphate (cGMP), the second messenger of nitric oxide [33,34]. PDE-5 is abundantly expressed in lung tissue and is a critical enzyme for the regulation of pulmonary vascular tone [35]. The feasibility of PDE-5 inhibition for pulmonary selective vasodilatation has been shown for sildenafil [10;19] and its clinical efficacy in PAH was recently demonstrated in the SUPER-1 trial [5]. The combination of inhaled iloprost and sildenafil has been shown to induce synergistic effects with respect to acute haemodynamic changes. An uncontrolled clinical trial showed that in patients who deteriorated with inhaled iloprost monotherapy adjunct sildenafil treatment improved and stabilised exercise capacity above the range previously achieved by iloprost alone [13,19]. Currently a randomized, placebo controlled, phase III trial investigates the efficacy of inhaled iloprost on top of previous sildenafil treatment (VISION trial). Inhaled treprostinil can be applied in only four daily doses due to its prolonged half life [36]. The inhalation of an effective treprostinil dose can be done in literally one single breath without systemic side effects [23].

The current study addressed the safety, tolerability and acute pharmacodynamic effects of the combination of sildenafil and inhaled treprostinil.

Out of the 50 patients undergoing acute vasodilator-challenge in our study, 4 patients (8%) were classified as nitric oxide-responders according to the current consensus criteria (drop of mean PAP more than 10 mmHg and below 40mmHg, normalisation of cardiac index) [37]. The oral dose of 50mg sildenafil showed a tendency to induce a lower pulmonary arterial resistance as compared to the preceding nitric oxide inhalation ($p=0.07$).

Treprostinil inhalation following the sildenafil application induced additional pulmonary vasodilatation ($p<0.05$ for PVR). The reduction of PVR by the combination of sildenafil and

inhaled treprostinil was significantly stronger than the NO-effect on PVR ($p < 0.001$). The additional effect of inhaled treprostinil outlasted the observation period of 120 minutes after inhalation (Figure 1), longer observations were not done in order to keep total catheter time tolerable. The 120 minute AUC of the treprostinil add-on effect (PVR at start of treprostinil inhalation set as 100%) was comparable to the previously recorded 120 minute AUC after treprostinil inhalation without sildenafil pre-treatment [23]. Given the precaution of a lack of an intra-individual comparison of treprostinil monotherapy and sildenafil/treprostinil combination, the observed additive effects of inhaled treprostinil and sildenafil in our view still document a translation of the theoretical benefits of parallel activation of the cAMP and cGMP pathways into the clinical setting.

For specific treatment of chronic thromboembolic pulmonary hypertension patients only inhaled iloprost is currently approved in Australia and New Zealand. In our non-operable chronic thromboembolic patients sildenafil and inhaled treprostinil induced additive pulmonary vasodilatation that did not differ in magnitude from the effects observed in PAH patients (Figure 4). We could recently show for open label treatment of more than 100 patients that sildenafil is beneficial in CTEPH [10,38]. Although it is not known if a favourable acute vasoresponse to this combination of sildenafil and treprostinil will translate into long term clinical effects for CTEPH patients, we propose that this drug combination will be useful also for continuous treatment of CTEPH patients and a controlled trial should be performed to solve that question.

A significant dose effect of inhaled treprostinil could not be observed in this study with 15µg and 30µg doses. However, the higher dose induced a longer lasting reduction of PVR (Figure 3). Dose dependent treprostinil effects on exercise capacity and haemodynamics were observed recently [25].

The effects on systemic arterial pressure of sildenafil alone and sildenafil plus inhaled treprostinil were minor, the combination of both drugs did not lead to increasing side effects. Mild side effects like flush were sometimes observed with the drug combination. The safety reported here only relates to a single and short term application of this drug combination and can not be extrapolated to long term use.

Relevant changes in gas exchange were not provoked by either drug. The multiple-inert-gas elimination technique was employed to directly assess intrapulmonary ventilation-perfusion matching in six patients with pre-existing gas exchange problems. Drug actions of sildenafil and inhaled treprostinil were compared to nitric oxide inhalation. Inhaled nitric oxide is the prototype of an intrapulmonary selective vasodilator due to its rapid inactivation after binding to haemoglobin [26]. Shunt flow fraction or low ventilation/perfusion areas were not increased by the combination of sildenafil and inhaled treprostinil compared to baseline and to nitric oxide inhalation. This finding is in favour for intrapulmonary selective acute vasodilatation by both applied agents in combination. Intrapulmonary selective vasodilatation has already been demonstrated for sildenafil [9]. This is considered to be due to the higher NO production in well ventilated areas resulting in higher cGMP concentrations and preferential sildenafil action at these sites. The intrapulmonary selectivity of inhaled treprostinil can be explained by preferential aerosol deposition in well ventilated areas.

Study limitations

Our study had limitations. The study was not placebo controlled. However, previous extensive work on acute haemodynamics in pulmonary hypertension patients consistently showed stable or increasing values for mPAP and PVR over time of catheterisation. Therefore the significant decreases of these values in our study must not directly be compared to placebo. The study did

not include an intra-individual comparison of treprostinil vs. sildenafil + treprostinil because of the long clinical effect inhaled treprostinil induces. To do this a washout time of at least 4 hours and a overall catheter time of approximately 8 hours would be needed which we judged inappropriate. The study was not a prospective trial on continuous treatment with this drug combination. It was not the intent of this study to derive long term treatment recommendations and such may not be deduced from these acute haemodynamics. But based on our experience with the clinical development of sildenafil and inhaled iloprost in this field, there is a high probability for a translation of the favourable acute haemodynamic improvements observed with this drug combination into clinical efficacy for PAH and possibly also non-operable CTEPH patients.

Perspectives

Pulmonary arterial hypertension and other forms of significant precapillary pulmonary hypertension, such as the also here investigated non-operable chronic thromboembolic PH and PH associated with chronic lung diseases, are generally assumed to take a progressive and fatal course if not specifically treated. Since a few years specific medical treatments are available that may slow down the natural history of disease and stabilise patients for a certain time but that needs to be expanded to a combination of approved drugs for most patients in order to prevent relapse of disease progression. Oral and inhaled drugs are preferable over intravenous or subcutaneous continuous drug infusion with respect to convenience and side effects. Sildenafil is widely used since its release for PAH. Prostacyclin analogues are well known for their beneficial effects in PAH. However, their use has so far been hampered by the need of continuous infusion or frequent inhalation. Inhaled treprostinil is currently in clinical development and may soon expand our medical armamentarium with an inhalation frequency of only q.i.d. The data provided

in this manuscript clearly indicate an additive effect of sildenafil and inhaled treprostinil without significant side effects both in PAH as well as in CTEPH patients. The potent pulmonary selective vasodilatation did not lead to gas exchange alterations. Long term treatment effects of this combination will have to be assessed in controlled trials.

ACKNOWLEDGEMENTS:

We would like to thank Melanie Thamm, Burcu Karadas, Peter Haredza and Sebastian Klemm (Department of Internal Medicine, University of Giessen) for technical assistance. We thank Rory Morty, PhD, for proofreading.

CONFLICT OF INTEREST DECLARATIONS

Robert Voswinckel, Beate Enke, Frank Reichenberger, Andree Kreckel, Stefanie Krick, Ralph Theo Schermuly and Henning Gall have nothing to declare. H.A. Ghofrani receives grant and contract support by Pfizer Ltd., Altana Pharma AG, Schering AG; in addition, he serves on advisory board of Pfizer Ltd. Friedrich Grimminger receives grant and contract support by Pfizer Ltd. and Altana Pharma AG. W. Seeger receives grant and contract support by Schering, Altana Pharma, Myogen Inc. Westminster, LungRX and Aventis Pharma. Horst Olschewski, is consultant and investigator for LungRX and Encysive and received research grants from Schering AG. Lewis J. Rubin is investigator and consultant for Actelion, Gilead, Pfizer, Lung Rx, and Schering. The work was supported with a clinical research grant by LungRX.

Reference List

- [1] Barst RJ, Rubin LJ, McGoon MD, Caldwell EJ, Long WA, Levy PS. Survival in primary pulmonary hypertension with long-term continuous intravenous prostacyclin. *Ann Intern Med* 1994 Sep 15;121(6):409-15.
- [2] Galie N, Torbicki A, Barst R, Darteville P, Haworth S, Higenbottam T, et al. Guidelines on diagnosis and treatment of pulmonary arterial hypertension. The Task Force on Diagnosis and Treatment of Pulmonary Arterial Hypertension of the European Society of Cardiology. *Eur Heart J* 2004 Dec;25(24):2243-78.
- [3] Olschewski H, Simonneau G, Galie N, Higenbottam T, Naeije R, Rubin LJ, et al. Inhaled iloprost for severe pulmonary hypertension. *N Engl J Med* 2002 Aug 1;347(5):322-9.
- [4] Rubin LJ, Badesch DB, Barst RJ, Galie N, Black CM, Keogh A, et al. Bosentan therapy for pulmonary arterial hypertension. *N Engl J Med* 2002 Mar 21;346(12):896-903.
- [5] Galie N, Ghofrani HA, Torbicki A, Barst RJ, Rubin LJ, Badesch D, et al. Sildenafil citrate therapy for pulmonary arterial hypertension. *N Engl J Med* 2005 Nov 17;353(20):2148-57.
- [6] Barst RJ, Rubin LJ, Long WA, McGoon MD, Rich S, Badesch DB, et al. A comparison of continuous intravenous epoprostenol (prostacyclin) with conventional therapy for primary pulmonary hypertension. The Primary Pulmonary Hypertension Study Group. *N Engl J Med* 1996 Feb 1;334(5):296-302.

- [7] Barst RJ, Langleben D, Frost A, Horn EM, Oudiz R, Shapiro S, et al. Sitaxsentan therapy for pulmonary arterial hypertension. *Am J Respir Crit Care Med* 2004 Feb 15;169(4):441-7.
- [8] Simonneau G, Barst RJ, Galie N, Naeije R, Rich S, Bourge RC, et al. Continuous subcutaneous infusion of treprostinil, a prostacyclin analogue, in patients with pulmonary arterial hypertension: a double-blind, randomized, placebo-controlled trial. *Am J Respir Crit Care Med* 2002 Mar 15;165(6):800-4.
- [9] Ghofrani HA, Wiedemann R, Rose F, Schermuly RT, Olschewski H, Weissmann N, et al. Sildenafil for treatment of lung fibrosis and pulmonary hypertension: a randomised controlled trial. *Lancet* 2002 Sep 21;360(9337):895-900.
- [10] Ghofrani HA, Schermuly RT, Rose F, Wiedemann R, Kohstall MG, Kreckel A, et al. Sildenafil for long-term treatment of nonoperable chronic thromboembolic pulmonary hypertension. *Am J Respir Crit Care Med* 2003 Apr 15;167(8):1139-41.
- [11] Hoeper MM, Kramm T, Wilkens H, Schulze C, Schafers HJ, Welte T, et al. Bosentan therapy for inoperable chronic thromboembolic pulmonary hypertension. *Chest* 2005 Oct;128(4):2363-7.
- [12] Humbert M, Sitbon O, Chaouat A, Bertocchi M, Habib G, Gressin V, et al. Pulmonary arterial hypertension in France: results from a national registry. *Am J Respir Crit Care Med* 2006 May 1;173(9):1023-30.
- [13] Ghofrani HA, Rose F, Schermuly RT, Olschewski H, Wiedemann R, Kreckel A, et al. Oral sildenafil as long-term adjunct therapy to inhaled iloprost in severe pulmonary arterial hypertension. *J Am Coll Cardiol* 2003 Jul 2;42(1):158-64.

- [14] Hoeper MM, Faulenbach C, Golpon H, Winkler J, Welte T, Niedermeyer J. Combination therapy with bosentan and sildenafil in idiopathic pulmonary arterial hypertension. *Eur Respir J* 2004 Dec;24(6):1007-10.
- [15] Hoeper MM, Taha N, Bekjarova A, Gatzke R, Spiekerkoetter E. Bosentan treatment in patients with primary pulmonary hypertension receiving nonparenteral prostanoids. *Eur Respir J* 2003 Aug;22(2):330-4.
- [16] Gomberg-Maitland M, McLaughlin V, Gulati M, Rich S. Efficacy and safety of sildenafil added to treprostinil in pulmonary hypertension. *Am J Cardiol* 2005 Nov 1;96(9):1334-6.
- [17] Schermuly RT, Roehl A, Weissmann N, Ghofrani HA, Schudt C, Tenor H, et al. Subthreshold doses of specific phosphodiesterase type 3 and 4 inhibitors enhance the pulmonary vasodilatory response to nebulized prostacyclin with improvement in gas exchange. *J Pharmacol Exp Ther* 2000 Feb;292(2):512-20.
- [18] Hoeper MM, Schwarze M, Ehlerding S, Adler-Schuermeyer A, Spiekerkoetter E, Niedermeyer J, et al. Long-term treatment of primary pulmonary hypertension with aerosolized iloprost, a prostacyclin analogue. *N Engl J Med* 2000 Jun 22;342(25):1866-70.
- [19] Ghofrani HA, Wiedemann R, Rose F, Olschewski H, Schermuly RT, Weissmann N, et al. Combination therapy with oral sildenafil and inhaled iloprost for severe pulmonary hypertension. *Ann Intern Med* 2002 Apr 2;136(7):515-22.
- [20] McLaughlin VV, Gaine SP, Barst RJ, Oudiz RJ, Bourge RC, Frost A, et al. Efficacy and safety of treprostinil: an epoprostenol analog for primary pulmonary hypertension. *J Cardiovasc Pharmacol* 2003 Feb;41(2):293-9.

- [21] Laliberte K, Arneson C, Jeffs R, Hunt T, Wade M. Pharmacokinetics and Steady-State Bioequivalence of Treprostinil Sodium (Remodulin(R)) Administered by the Intravenous and Subcutaneous Route to Normal Volunteers. *J Cardiovasc Pharmacol* 2004 Aug;44(2):209-14.
- [22] Clapp LH, Finney P, Turcato S, Tran S, Rubin LJ, Tinker A. Differential effects of stable prostacyclin analogs on smooth muscle proliferation and cyclic AMP generation in human pulmonary artery. *Am J Respir Cell Mol Biol* 2002 Feb;26(2):194-201.
- [23] Voswinckel R, Enke B, Reichenberger F, Kohstall M, Kreckel A, Krick S, et al. Favorable effects of inhaled treprostinil in severe pulmonary hypertension: results from randomized controlled pilot studies. *J Am Coll Cardiol* 2006 Oct 17;48(8):1672-81.
- [24] Voswinckel R, Ghofrani HA, Grimminger F, Seeger W, Olschewski H. Inhaled treprostinil [corrected] for treatment of chronic pulmonary arterial hypertension. *Ann Intern Med* 2006 Jan 17;144(2):149-50.
- [25] Channick RN, Olschewski H, Seeger W, Staub T, Voswinckel R, Rubin LJ. Safety and efficacy of inhaled treprostinil as add-on therapy to bosentan in pulmonary arterial hypertension. *J Am Coll Cardiol* 2006 Oct 3;48(7):1433-7.
- [26] Walmrath D, Schermuly R, Pilch J, Grimminger F, Seeger W. Effects of inhaled versus intravenous vasodilators in experimental pulmonary hypertension. *Eur Respir J* 1997 May;10(5):1084-92.

- [27] Walmrath D, Schneider T, Pilch J, Schermuly R, Grimminger F, Seeger W. Effects of aerosolized prostacyclin in severe pneumonia. Impact of fibrosis. *Am J Respir Crit Care Med* 1995 Mar;151(3 Pt 1):724-30.
- [28] Wagner PD, Saltzman HA, West JB. Measurement of continuous distributions of ventilation-perfusion ratios: theory. *J Appl Physiol* 1974 May;36(5):588-99.
- [29] Moncada S, Higgs EA. Prostaglandins in the pathogenesis and prevention of vascular disease. *Blood Rev* 1987 Jun;1(2):141-5.
- [30] Narumiya S, Sugimoto Y, Ushikubi F. Prostanoid receptors: structures, properties, and functions. *Physiol Rev* 1999 Oct;79(4):1193-226.
- [31] Ghofrani HA, Rose F, Schermuly RT, Olschewski H, Wiedemann R, Weissmann N, et al. Amplification of the pulmonary vasodilatory response to inhaled iloprost by subthreshold phosphodiesterase types 3 and 4 inhibition in severe pulmonary hypertension. *Crit Care Med* 2002 Nov;30(11):2489-92.
- [32] McLaughlin VV, Shillington A, Rich S. Survival in primary pulmonary hypertension: the impact of epoprostenol therapy. *Circulation* 2002 Sep 17;106(12):1477-82.
- [33] Ghofrani HA, Osterloh IH, Grimminger F. Sildenafil: from angina to erectile dysfunction to pulmonary hypertension and beyond. *Nat Rev Drug Discov* 2006 Aug;5(8):689-702.
- [34] Giordano D, De Stefano ME, Citro G, Modica A, Giorgi M. Expression of cGMP-binding cGMP-specific phosphodiesterase (PDE5) in mouse tissues and cell lines using an antibody against the enzyme amino-terminal domain. *Biochim Biophys Acta* 2001 May 28;1539(1-2):16-27.

- [35] Corbin JD, Beasley A, Blount MA, Francis SH. High lung PDE5: A strong basis for treating pulmonary hypertension with PDE5 inhibitors. *Biochem Biophys Res Commun* 2005 Sep 2;334(3):930-8.
- [36] Wade M, Baker FJ, Roscigno R, DellaMaestra W, Arneson CP, Hunt TL, et al. Pharmacokinetics of treprostinil sodium administered by 28-day chronic continuous subcutaneous infusion. *J Clin Pharmacol* 2004 May;44(5):503-9.
- [37] Sitbon O, Humbert M, Jais X, Ioos V, Hamid AM, Provencher S, et al. Long-term response to calcium channel blockers in idiopathic pulmonary arterial hypertension. *Circulation* 2005 Jun 14;111(23):3105-11.
- [38] Reichenberger F, Voswinckel R, Enke B, Rutsch M, El FE, Schmehl T, et al. Long-term treatment with sildenafil in chronic thromboembolic pulmonary hypertension. *Eur Respir J* 2007 Nov;30(5):922-7.

Figure 1

Acute haemodynamic responses of PHT patients to inhaled nitric oxide (bar graph), oral sildenafil and inhaled treprostinil (line graph). The graphs show the pooled data of all 50 patients. The arrows depict the time of pharmacological challenges. Sildenafil induced a sustained reduction of PVR and PAP, as well as a rise in cardiac output. Inhaled treprostinil in addition to sildenafil induced a significant additional reduction of PVR and PAP that outlasted the observation period. Treprostinil also induced a sustained increase in cardiac output. No relevant changes of heart rate were observed. Data are given as mean $\pm$ 95% confidence interval. PVR = pulmonary vascular resistance, PAP = mean pulmonary artery pressure, CO = cardiac output, HR = heart rate, NO = nitric oxide inhalation (20 ppm), SIL = sildenafil (50 mg), TRE = treprostinil inhalation (15 or 30 μ g).

Figure 2

Acute haemodynamic responses of PHT patients to inhaled nitric oxide (bar graph), oral sildenafil and inhaled treprostinil (line graph). The graphs show the pooled data of all 50 patients. The arrows depict the moments of pharmacological challenges. SAP and SVR were not changed by nitric oxide (NO), the prototype of pulmonary selective vasodilators. The combination of oral sildenafil and inhaled treprostinil evoked only minor changes in SAP and SVR, demonstrating pulmonary selectivity of both drugs. The effects of sildenafil and treprostinil on gas exchange and central venous oxygen saturation, as parameters for intrapulmonary selectivity and overall improvement of circulation, were comparable to nitric oxide effects. Data are given as mean $\pm$ 95% confidence interval. SAP = mean systemic arterial pressure, SVR = systemic vascular resistance, SaO₂ = arterial oxygen saturation, SvO₂ = central venous oxygen saturation, NO = nitric oxide inhalation (20 ppm), SIL = sildenafil (50 mg), TRE = treprostinil inhalation (15 or 30 μ g).

Figure 3

Acute haemodynamic responses to inhaled nitric oxide, oral sildenafil and inhaled treprostinil separated for patients that inhaled 15µg treprostinil (white bars and circles; n=25) or 30µg treprostinil (black bars and circles; n=25). The arrows depict the time of pharmacological challenges. In the 15µg treprostinil group, a more pronounced PVR-response to nitric oxide inhalation was observed. However, this did not correlate with the response to sildenafil or treprostinil. The higher treprostinil dose (30µg) induced a more sustained pulmonary vasodilatation. The differences observed for cardiac output between the groups were not significant, the changes in SvO₂ were comparable in both groups. Data are given as mean ± 95% confidence interval. PVR = pulmonary vascular resistance, PAP = mean pulmonary arterial pressure, CO = cardiac output, SvO₂ = central venous oxygen saturation, NO = nitric oxide inhalation (20 ppm), SIL = sildenafil (50 mg), TRE = treprostinil inhalation (15 or 30µg).

Figure 4

Acute haemodynamic changes due to sildenafil and inhaled treprostinil in pulmonary arterial hypertension patients (PAH, n=26) and chronic thromboembolic pulmonary hypertension patients (CTEPH, n=17). In both groups, oral sildenafil induced a reduction of pulmonary vascular resistance (PVR) and pulmonary artery pressure (PAP), as well as a significant increase in cardiac output (CO). Inhaled treprostinil in addition provoked additive effects that lasted beyond the observation time in both groups. Systemic vascular resistance (SVR) was only marginally affected and not clinically relevant. No significant differences of acute haemodynamic responses to the combination treatment were observed between both patient collectives. Data are given as mean $\pm$ 95% confidence interval.

Figure 5

Ventilation/perfusion matching assessed with the multiple-inert-gas elimination technique.

Six patients with pre-existing gas exchange limitations were investigated for changes in ventilation-perfusion ratios. All patients demonstrated shunt flow in absence of pharmacological challenge (baseline). Shunt-flow and low V/Q areas were not significantly changed by nitric oxide (NO) inhalation, oral sildenafil (SIL) or the combination of sildenafil and treprostinil inhalation (SIL+TRE). Data are given as mean $\pm$ 95% confidence interval.

Accepted manuscript

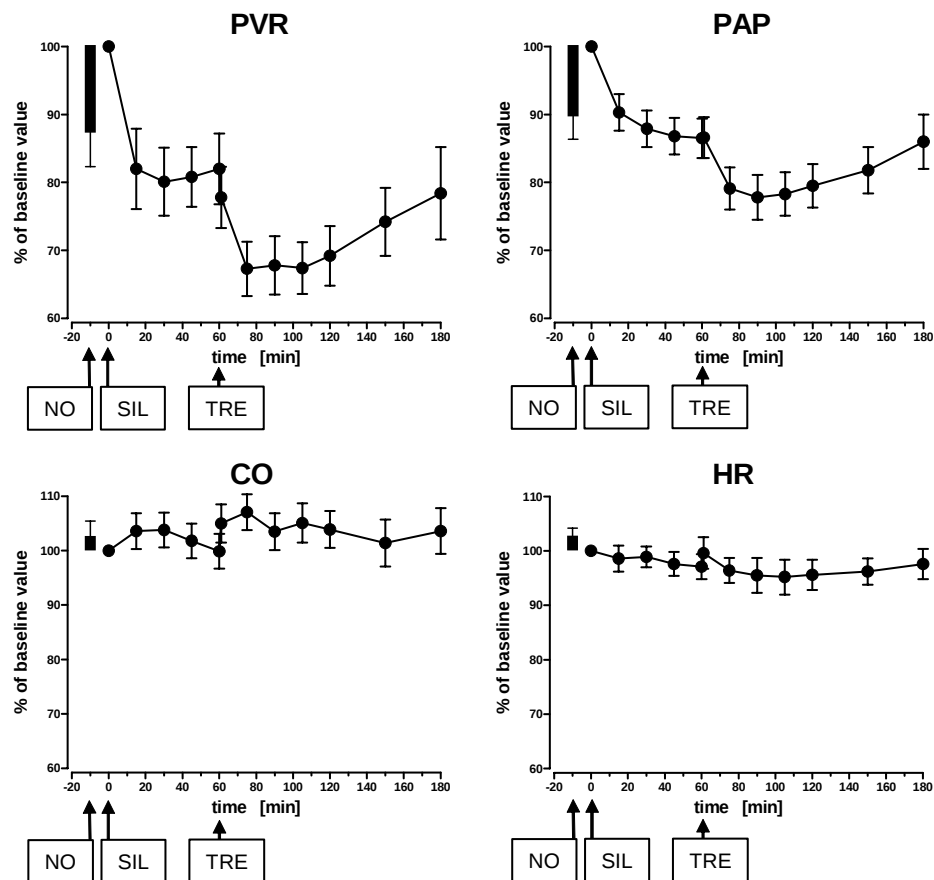


Figure 1

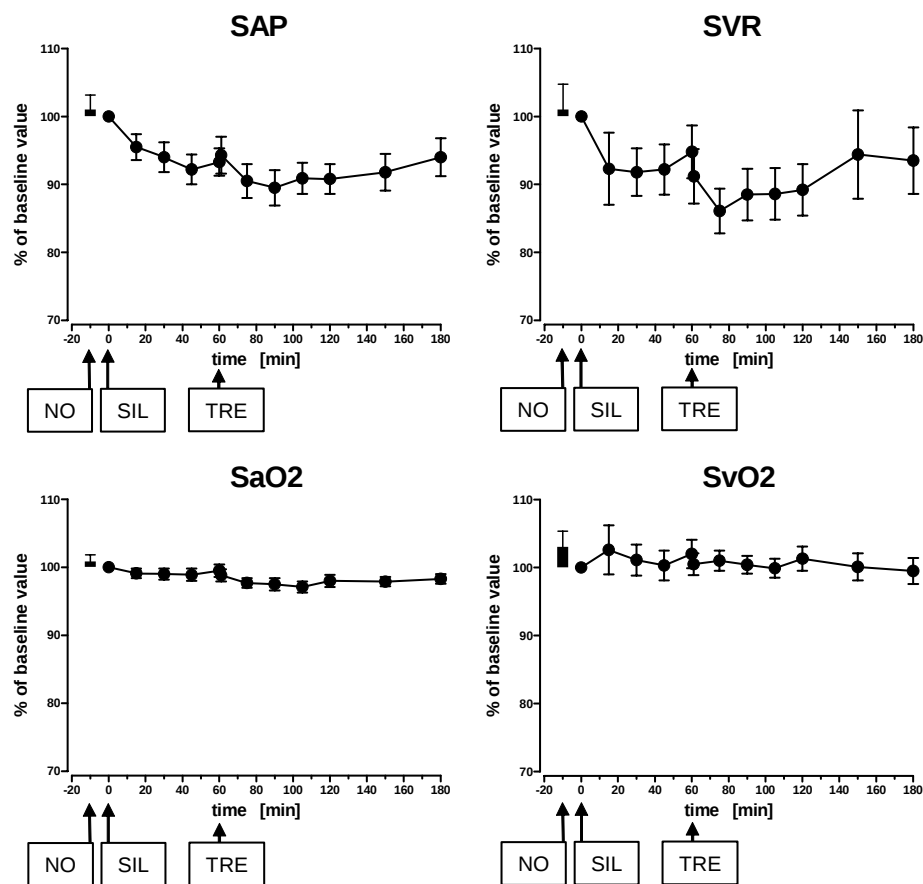


Figure 2

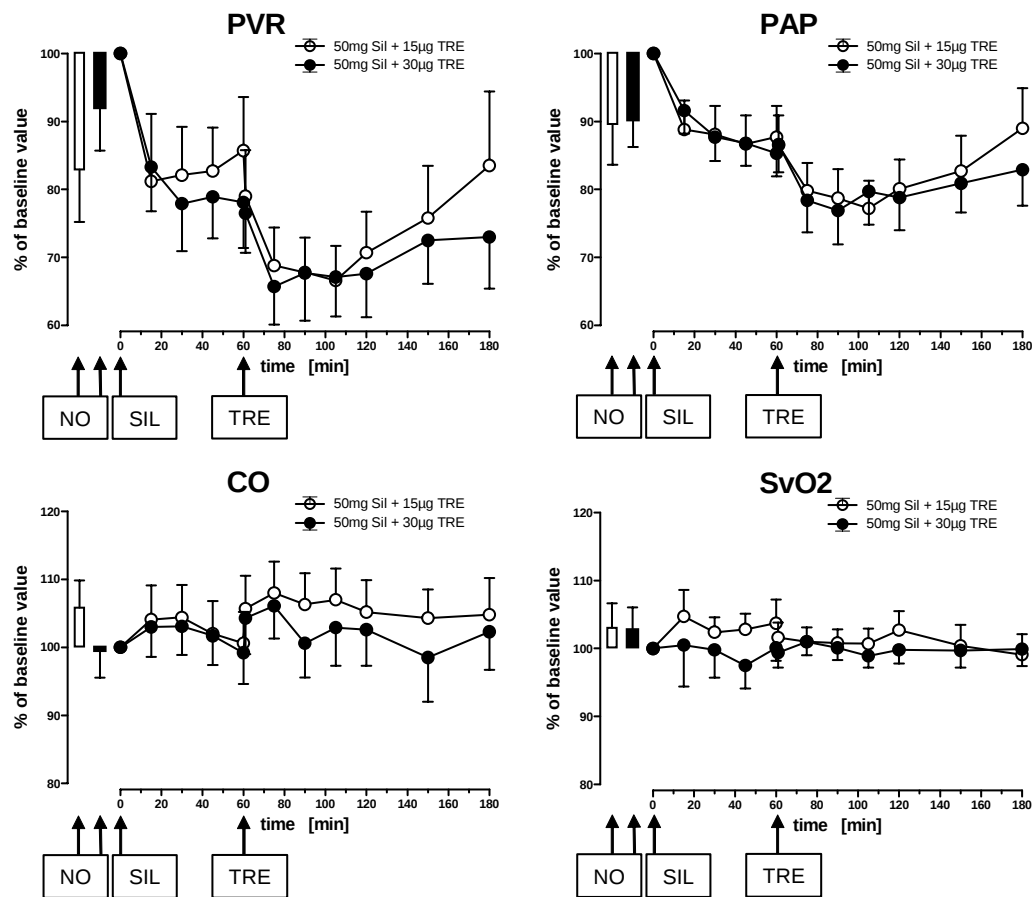


Figure 3

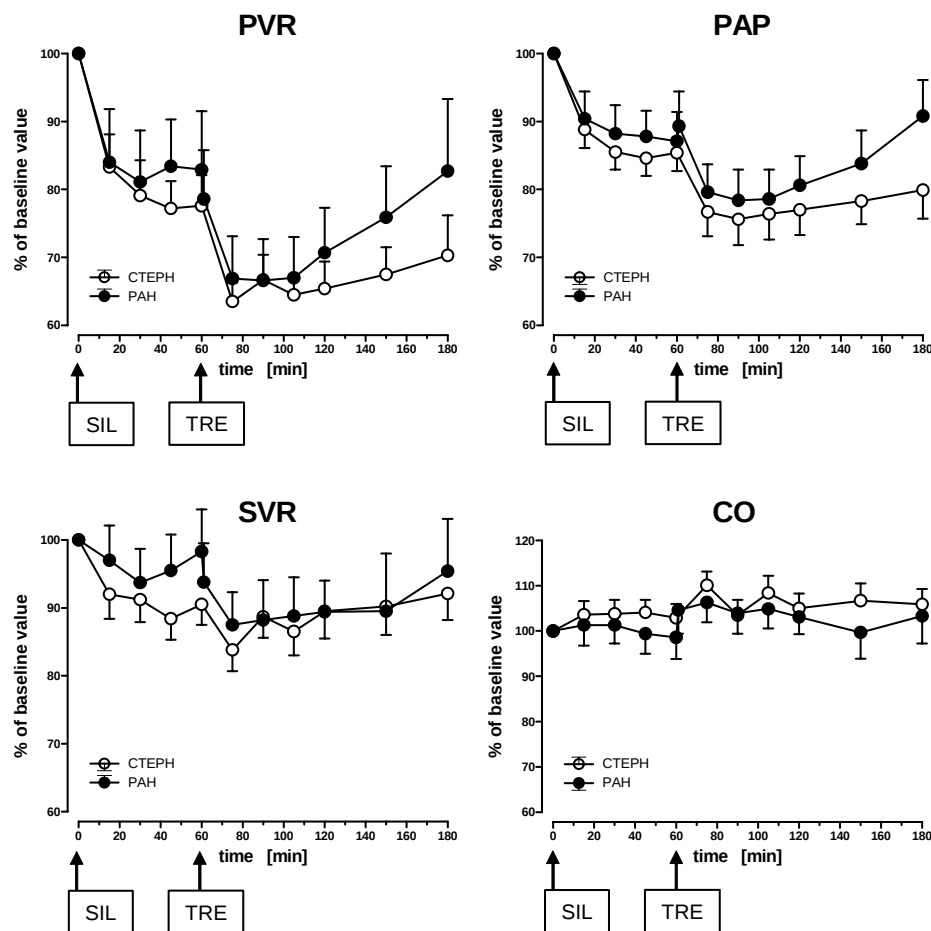


Figure 4

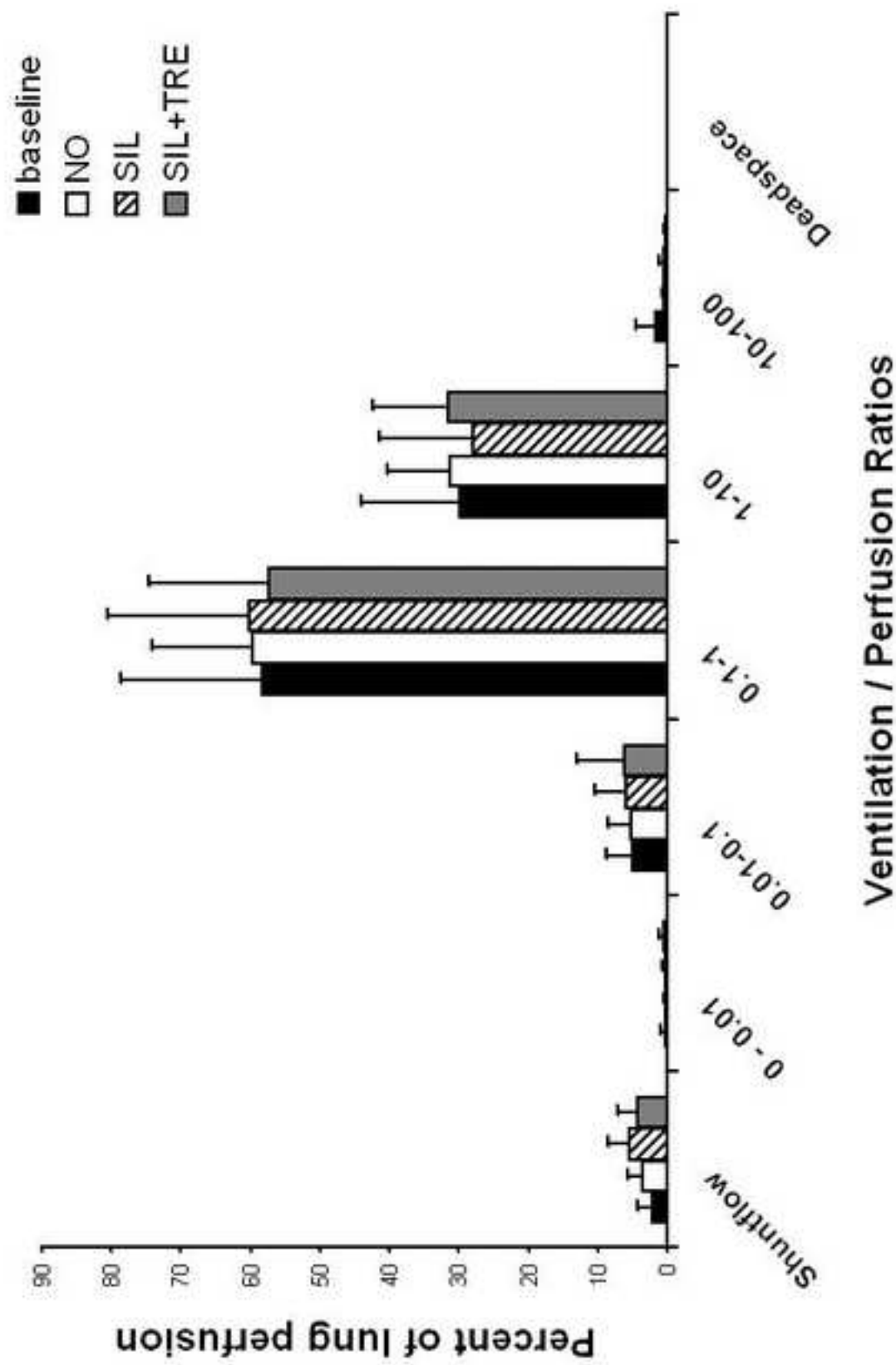


Figure 5