Original Research Article

Efficacy of a combination of sildenafil and magnesium sulfate in the treatment of persistent pulmonary hypertension of the newborn, and its influence on hemodynamics

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Abstract

Purpose: *To investigate the efficacy of the combined use of sildenafil and magnesium sulfate in the treatment of persistent pulmonary hypertension of the newborn (PPHN), and its influence on hemodynamics.*

Methods: *A total of 174 children with persistent pulmonary hypertension who were treated in Ganzhou People's Hospital, Ganzhou, China were selected and randomly assigned to joint group (JG) and control group (CG), with 87 patients in each group. The CG group received magnesium sulfate, while the JG group received sildenafil plus magnesium sulphate. The respiratory parameters of the children were analyzed using blood gas analyzer, while their hemodynamic indices were evaluated using color Doppler echocardiography. The levels of cytokines and inflammatory factors were determined by enzyme-linked immunosorbent assay (ELISA).*

Results: *Time taken for symptom disappearance, oxygen therapy, and hospitalization period were shorter in JG than in CG (p < 0.05). Post-treatment, the respiratory parameters (PaO_2, $PaCO_2$, and SaO_2) in both groups s improved, with lower levels of PaO_2 and $PaCO_2$, and a higher level of SaO_2 in JG (p < 0.05). Following treatment, the levels of systemic vascular resistance (SVR), posterior pulmonary vascular resistance (PVR) and pulmonary artery pressure (PA) in JG were significantly reduced, relative to CG (p < 0.05). Similarly, the expression of endothelin -1 (ET-1), brain natriuretic peptide (BNP), and angiotensin 1 (ANG-1) improved, with lower levels of ET-1 and BNP, and a higher level of ANG-1 in JG (p < 0.05). There was post-treatment reduction as well in IL-6 and TNF-α, with lower levels in JG (p < 0.05). Patients in JG showed higher total treatment effectiveness and a lower incidence of adverse reactions than those in CG (p < 0.05).*

Conclusion: *The combined use of sildenafil and magnesium sulfate enhances the management of PPHN, ameliorates respiratory parameters, hemodynamics, and levels of cytokines and inflammatory factors. These findings provide evidence-based medical references for a new treatment strategy for PPHN.*

Keywords: *Sildenafil, Magnesium sulfate, Persistent pulmonary hypertension, Newborn, Hemodynamics*

INTRODUCTION

Persistent pulmonary hypertension of the newborn (PPHN) is a common disease with extremely high morbidity and mortality. Pulmonary hypertension leads to right-to-left shunt, thereby triggering hypoxemia and life-threatening circulatory failure in children [1]. Persistent pulmonary hypertension of the newborn is related to meconium aspiration, congenital pneumonia, respiratory distress syndrome, and birth asphyxia [2]. The current clinical treatment for PPHN centers mainly on the treatment of the underlying etiology, including maintenance of adequate systemic blood pressure, respiratory support for lung recruitment and alveolar ventilation, relaxation of pulmonary vessels, and reduction of pulmonary vascular resistance using pharmacological measures [3]. However, in mechanical ventilation treatment, 11 % of children suffer from chronic lung diseases, and about 9 % experience serious neurological complications [4]. Therefore, it is important to evolve safer and more effective treatment methods for PPHN.

Sildenafil is a selective phosphodiesterase type-5 inhibitor which effectively improves the athletic ability of patients with pulmonary hypertension or cardiopulmonary diseases. Several studies have revealed that sildenafil increases the local concentration of cyclic guanosine monophosphate (cGMP), thereby causing vasodilation during relaxation of smooth muscle cells in arterial walls, cavernous bodies, and lungs [5]. In addition, sildenafil is used for the treatment of various diseases such as heart failure, stroke and myocardial infarction. Rashid *et al.* [6] have reported that sildenafil mitigated the clinical symptoms of pulmonary hypertension and the hemodynamics of patients. Magnesium sulfate is a vasodilator which represses angiotensin-converting enzymes through the synthesis of PGI2, thereby controlling hypertension and protecting the nerves. Research has shown that magnesium sulfate is effective in anti-inflammatory activity and antioxidation, and that it also effectively regulates mitochondrial dysfunction, apoptosis, inflammatory corpuscles, and pulmonary vascular resistance, with a regulatory effect against pulmonary hypertension [7].

In this research, the combination of sildenafil and magnesium sulfate was used to treat PPHN. The efficacy of the combination treatment was compared with that of magnesium sulfate alone, in addition to levels of inflammatory factors, and hemodynamics of children with PPHN.

METHODS

Subjects

A total of 174 children with persistent pulmonary hypertension admitted into our hospital from May 2018 to August 2019 served as the research subjects. They were randomly assigned to the joint group (JG) and the control group (CG), with 87 patients in each group. This research was done in line with the International Human Research Guidelines [8], and it received approval from the ethical committee of Ganzhou People's Hospital (approval no. gz18031154). Patients and their family members received information on the study protocols, and they submitted signed attestations of consent to participate.

Inclusion criteria

The included patients were those who met the diagnosis of persistent pulmonary hypertension by the American Heart Association (AHA) and American Thoracic Society (ATS) [9]; patients with persistent hypoxemia, obvious dyspnea, and severe cyanosis; those who had not received any chemotherapy treatment; patients who underwent follow-up therapy post-diagnosis, and patients who were willing to cooperate with the study.

Exclusion criteria

Patients without dysfunctions in major organs, patients who withdrew from the experiment halfway, those with chronic diseases, patients with organ failure, patients with drug allergies, and those lost to follow-up, were excluded from the study.

Treatments

The two categories of children were placed on the radiation thermal table after admission, followed by continuous monitoring of blood pressure, blood oxygen, and sedation intake. With acidosis corrected and electrolyte balance, dopamine was used to maintain systemic circulation pressure. Infection prevention and control treatment were also performed.

Children in CG were treated with magnesium sulfate (Wenyi Biotechnology Co. Ltd., Guangzhou, China, WY20131104). The magnesium sulfate was dissolved in glucose injection intravenous drip, and was administered at an initial dose of 0.2 g/kg which was adjusted as appropriate according to the actual conditions of the patient. The patients' conditions were observed after 30 minutes of treatment. The drip

rate was adjusted to 25-50 mg/kg/h in case of improvement in patients' conditions, and the intravenous drip was terminated when the transcutaneous oxygen saturation of the children ≥ 90 %. The treatment for children in CG spanned 5 days.

Patients in JG were treated with sildenafil tablets (Pfizer, Dalian, China, H20020527), in addition to the treatment in the control group. The doses of sildenafil tablets were in the range of 0.5 - 2.0 mg/kg (based on the weight of the children) given once every 6 - 8 h for 5 consecutive days.

Parameters determined

Clinical symptoms

Time of symptom disappearance, oxygen therapy, and hospitalization duration) of children in both groups were recorded.

Respiratory indices

The arterial partial pressure of oxygen (PaO_2), partial pressure of carbon dioxide ($PaCO_2$), and saturation of blood oxygen (SaO_2) in the two groups before and 24 h post-therapy were determined with blood gas analyzer using 5 mL of blood taken into a heparin cap, and transferred into a cooling package (the temperature was lower than 8 °C).

Hemodynamic indices

The systemic vascular resistance (SVR), posterior pulmonary vascular resistance (PVR), and pulmonary artery pressure (PA) of children were determined using color Doppler echocardiography (Hanfei Medical Devices Co. Ltd., Shanghai, China, DC-N2S), and calculated according to Fick's formula.

Cytokines and inflammatory factors

Plasma samples from elbow venous blood (5 mL) before and 24 h after treatment, were subjected to assay of cytokines and other factors using their respective ELISA kits [13]: endothelin -1 (ET-1), brain natriuretic peptide (BNP), angiotensin 1 (ANG-1), interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α), and ET-2 (Hengfei Biotechnology Co. Ltd., Shanghai, China, bs-0954R-1); BNP (Zhenyu Biotechnology Co. Ltd, Shanghai, China, Bs-4865R-1); IL-6 [MultiSciences (Lianke) Biotech Co. Ltd., Hangzhou, China, 70-EK106/2), and TNF-α (Yuanmu Biotechnology Co. Ltd., Shanghai,

China, YM-S0122H), in accordance with the instructions on the kit manuals.

Efficacy

Treatment outcome was categorized as *markedly effective* if symptoms disappeared post-therapy, and the respiratory parameters and hemodynamic indexes returned to normal, or *effective* if the clinical symptoms, respiratory parameters, and hemodynamic indexes were improved after treatment. However, outcome was *ineffective* if the clinical symptoms of children after treatment were not mitigated, or were even aggravated. *Total effectiveness* was taken as the sum of the number of patients in whom the treatment produced marked effectiveness and those in whom there was effectiveness, expressed as a percentage of the total number of patients.

Adverse reactions

Adverse reactions in children of both groups during treatment were recorded and compared.

Statistical analysis

Count data are presented as numbers and percentages [n (%)]. Inter-group comparison was done with χ^2 test, and at frequency <5, corrected χ^2 test was applied. Measured data are presented as mean ± SD and paired *t*-test was applied for intra-group comparison before and after treatment. The comparison among multiple groups was done with one-way ANOVA, while that between both groups was analyzed with LSD *t*-test. SPSS software (version 21.0) was applied for all statistical analyses. Differences were considered statistically significant at *p* < 0.05.

RESULTS

General profile of children

There were no marked differences in gender, place of residence, nationality, gestational age (weeks), birth weight (g), delivery mode, disease type, and history of drinking and smoking between both groups (Table 1).

Clinical indices

Clinical indices such as time of symptom disappearance, oxygen therapy, and hospitalization time were shorter in JG than in controls (Table 2).

Table 1: General profile of children studied

Variable	Joint group (JG) (n=87)	Control group (CG) (n=87)	t/χ^2	P-value
Gender (male/female)	47/40	52/35	0.586	0.444
Gestational age (weeks)	35.23±3.13	35.74±3.16	1.070	0.286
Birth weight (g)	3037±343	3028±865	0.090	0.928
Delivery mode			0.829	0.363
Eutocia	45	39		
Cesarean section	42	48		
Disease type			0.944	0.815
Intrauterine distress	20	21		
Meconium aspiration	28	33		
Pneumonia	22	19		
Respiratory distress	17	14		
Alcohol (yes/no)	48/39	41/46	1.127	0.288
Smoking (yes/no)	55/32	46/41	1.912	0.167

Table 2: Clinical indices of children in both groups, post-treatment (days, mean ± SD, n = 87)

Group	Symptom disappearance time	Oxygen therapy time	Length of stay
JG	5.74±0.41	9.03±0.63	12.55±1.01
CG	7.79±0.63	13.21±1.03	18.36±1.56
t	25.44	32.29	29.16
P-value	<0.001	<0.001	<0.001

Respiratory parameters

The pre-drug exposure levels of respiratory parameters (PaO_2, $PaCO_2$, and SaO_2) in the 2 groups were not different. However, the respiratory parameters were improved after treatment. The PaO_2 and $PaCO_2$ levels were lower in JG than in CG, while SaO2 level was markedly higher in JG. These results are shown in Table 3.

Hemodynamic indices

Before treatment, levels of SVR, PVR, and PA were comparable in both groups. After treatment, the levels of these indexes decreased markedly, with SVR, PVR, and PA markedly lower in JG than in CG ($p < 0.05$; Table 4).

Cytokine levels

There were no marked differences in expressions of ET-1, BNP, and ANG-1 between both groups before treatment ($p > 0.05$). Both groups had improvements in the levels of cytokines after treatment, with significantly lower levels of ET-1 and BNP and a significantly higher level of ANG-1 in JG ($p < 0.05$; Figure 1).

Inflammatory factors

There were no marked differences in the levels of inflammatory factors like IL-6 and TNF-α before treatment. The expressions of inflammatory factors were improved after treatment, with down-regulated expressions of IL-6 and TNF-α in JG (Figure 2).

Table 3: Respiratory parameters of children pre- and post-treatment (mmHg, mean ± SD, n = 87)

Group	PaO_2		$PaCO_2$		SaO_2	
	Before	After	Before	After	Before	After
JG	29.45±3.35	48.33±3.23	64.86±5.45	35.28±3.87	55.46±4.63	90.76±8.68
CG	28.76±3.34	85.45±7.65	65.32±5.42	50.14±5.36	54.75±4.68	68.34±5.45
t	1.360	41.69	0.558	20.97	1.006	20.40
P-value	0.175	<0.001	0.577	<0.001	0.316	<0.001

Table 4: Hemodynamic indexes of children pre- and post-treatment (mean ± SD, n = 87)

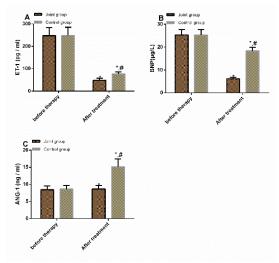| Group	SVR (dyns/cm^{-5})		PVR (dyns/cm^{-5})		PA (mmHg)	
	Before	After	Before	After	Before	After
JG	1482.32±156.45	1125.04±132.45	1032.12±152.34	719.47±100.87	69.28±10.22	25.89±3.22
CG	1479.94±155.87	1243.75±152.88	1029.96±152.01	948.34±101.45	68.96±10.04	42.52±3.18
t	0.101	5.474	0.094	14.92	0.208	34.28
P	0.920	<0.001	0.925	<0.001	0.835	<0.001

Figure 1: Comparison of cytokine levels between JG and CG pre- and post-treatment. (A): Pre-treatment ET-1 levels were similar in both groups. After treatment, ET-1 level was decreased in JG. (B): Before treatment, BNP expression was comparable in both groups. After treatment, the BNP level was decreased in JG, relative to CG. (C): ANG-1 expressions in the two groups were similar before treatment. However, post-treatment, the ANG-1 level was higher in JG. *P < 0.05; vs control pre-treatment; #p < 0.05, vs control post-treatment

Figure 2: Levels of inflammatory factors in both groups pre- and post-treatment. (A): Pre-treatment IL-6 expression was similar in the 2 groups. After treatment, IL-6 level was down-regulated in JG. (B): TNF-α expression in both groups before treatment were similar. After treatment, TNF-α level in the JG was reduced. *P < 0.05, pre-treatment; #p < 0.05, vs CG post-therapy

Therapeutic efficacy

The total effectiveness of children in JG was 94.25 % after treatment, while the effectiveness in the CG (83.91 %) was lower than that in the JG (p < 0.05; Table 5).

DISCUSSION

Pulmonary hypertension is a life-threatening disease characterized by right heart failure and death due to increased pulmonary vascular resis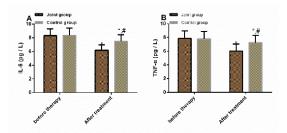tance. Clinically, the main treatment aims at the reduction of pulmonary vascular resistance, optimization of lung recruitment, and the use of pulmonary vasodilators. [10]. In this research, the combined use of sildenafil with magnesium sulfate was applied for treatment, with its effectiveness on the patients evaluated and compared.

Many clinical studies have reported the treatment methods and their effects on persistent pulmonary hypertension. For example, Sayed *et al* [11] reported that sildenafil improved the oxygenation indices of children with persistent pulmonary hypertension, with high safety, and that Oral Sildenafil (SIL) could be used as a pulmonary vasodilator in the case of nil response to inhaled nitric oxide.

Other studies have shown that magnesium sulfate reduced the incidence of complications such as arterial blood pressure, arrhythmia, and pulmonary artery pressure caused by tracheal intubation, and ameliorated the hemodynamics of patients [12]. In this study, the post-therapy improvement in clinical symptom indices of

Table 5: Comparison of treatment effectiveness between children in the two groups after treatment (n = 87)

Group	Markedly effective	Effective	Ineffective	Total effectiveness (%)
JG	35	47	5	94.25
CG	30	43	14	83.91
t				4.786
P-value				0.029

Table 6: Incidence of unwanted side effects after treatment of children

Group	Mild frequent micturition	Mild nausea	Tachycardia	Drowsiness	Total incidence (%)
JG	0	1	2	3	6.90
CG	1	3	3	4	12.64
t					1.630
P-value					0.202

children was more effective in JG than in control patients, indicating that the combined use of sildenafil with magnesium sulfate could better mitigate the clinical symptoms and dilate blood vessels. The respiratory parameters of JG after treatment were also better than those of the CG, which indicate that the combined use of sildenafil with magnesium sulfate could enhance lung perfusion in children, maintain appropriate ventilation, increase oxygen transport, and effectively alleviate hypoxic symptoms. Research has shown that the severity of pulmonary artery disease is related to pulmonary vascular resistance, and pulmonary artery disease may negatively affect hemodynamics [13].

In this study, the expressions of SVR, PVR, and PA in the JG after treatment were markedly decreased, relative to control values, suggesting that sildenafil plus magnesium sulfate was better at dilating the blood vessels and reducing pulmonary hypertension, pulmonary vascular resistance and artery pressure in children, thereby improving their hemodynamics.

Studies have shown that endothelial dysfunction can lead to pulmonary hypertension. For example, abnormal expressions of ET-1 and ANG-1 are tied to death and heart failure in patients with pulmonary hypertension [14]. Brain natriuretic peptide is a functional peptide synthesized by myocardial cells, and it plays a vital role in diagnosis of hypertension, heart failure, and pulmonary hypertension [15]. Zhou *et al* [16] have pointed out that sildenafil protects the endothelial function by inhibiting the NF-κB signaling pathway in patients with congenital heart disease complicated with pulmonary hypertension. In the present study, the results obtained demonstrate that the ET-1 and BNP expressions in JG after treatment were lower than those in the CG, while the ANG-1 expression was up-regulated in JG, indicating that sildenafil combined with magnesium sulfate reduced the cytokines in children, and the disease control effect after treatment was more satisfactory than that in controls. It has been reported that in pulmonary hypertension, the levels of inflammatory factors such as IL-6 and TNF-α increased in patients, and the prognosis of patients with pulmonary hypertension was related to the abnormal expressions of these inflammatory factors [17]. This research found that the levels of TNF-α and IL-6 were decreased in JG after treatment, indicating that combined use of sildenafil with magnesium sulfate was more effective in the mitigation of inflammatory response in children. The total effectiveness in children was markedly higher in JG after

treatment than in the controls. Adverse reactions in both groups during treatment were comparable, indicating better effectiveness in the combined use of sildenafil and magnesium sulfate.

Although this research has confirmed that the combined use of sildenafil and magnesium sulfate promoted the recovery of children with persistent pulmonary hypertension better than monotherapy with magnesium sulfate, more work needs to be done. For instance, basic experiments on the therapeutic mechanism of the two therapeutic methods can be used to identify the risk factors that affect treatment efficacy in children at the molecular level. Furthermore, the quality of life of patients can be investigated to support the results of this study.

CONCLUSION

The combined use of sildenafil and magnesium sulfate has a significant therapeutic effect on PPHN and this may be attributed to reduction in levels of inflammatory factors. Therefore, the combination therapy can potentially improve the management of children with PPHN.

DECLARATIONS

Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

We declare that this work was done by the authors named in this article, and all liabilities pertaining to claims relating to the content will be borne by the authors. Shuping Huang conceived the study and drafted the manuscript. Tao Zhong collected and analyzed the data and revised the manuscript. Both authors read and approved the final manuscript.

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