Intratracheal administration of budesonide with surfactant in very low birth weight infants to prevent bronchopulmonary dysplasia

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ABSTRACT

Background and objectives. Respiratory distress syndrome (RDS) is a major cause of early postnatal death in preterm infants. Bronchopulmonary dysplasia (BPD) is one of the most fatal chronic respiratory complications of preterm infants after management of RDS. Anti-inflammatory therapy with corticosteroid is one of the effective treatments to prevent BPD. However, systemic administration of corticosteroid is not recommended because of long-term adverse effects. We studied the effect of early intratracheal instillation of budesonide with surfactant in preterm infants with severe RDS.

Method. Very low birth weight infants (VLBWIs) weighing less than 1,500 g who were admitted to the neonatal intensive care unit (NICU) of Busan Paik Hospital between January 2018 and December 2018 and diagnosed with severe RDS were enrolled. The treatment group was given a mixture of budesonide and surfactant (calfactant) while the control group was given surfactant (calfactant) only.

Results. Surfactant re-dosing, duration of mechanical ventilation, BPD, mortality, and retinopathy of prematurity (≥ stage 2) were not different between the two groups though there were decreasing trends in the treatment group compared to those in the control group. The duration of hospital stay was longer in the control group with statistical significance.

Conclusion. Early intratracheal administration of budesonide with surfactant in preterm infants with severe RDS might decrease BPD and mortality without disturbing surfactant function. Further studies with different preparations of surfactants with a large number of preterm infants are required.

Key words: bronchopulmonary dysplasia, budesonide, preterm infant, pulmonary surfactant, respiratory distress syndrome.

Respiratory distress syndrome (RDS) is a major cause of early postnatal death in preterm infants. Bronchopulmonary dysplasia (BPD) is one of the most fatal chronic respiratory complications in preterm infants after management of RDS. The incidence of BPD has not decreased despite advances in neonatal care of RDS in the post-surfactant era.[1] BPD is a lung injury syndrome. Its pathogenesis is multifactorial. Inflammation is an important pathogenetic factor in the development of BPD.[2] Anti-inflammatory therapy with corticosteroid is one of the most effective treatments to prevent BPD.[3] However, systemic corticosteroid can cause long-term adverse effects such as poor somatic growth, small head circumference, and neurodevelopmental disabilities such as neuromotor deficits, and cognitive deficits.[4] Thus, systemic administration of corticosteroid is not recommended.[5]

If corticosteroid can be used directly to lungs, then systemic adverse effects can be decreased and local anti-inflammatory effect can be

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increased. However, there have been limitations to deliver corticosteroid to distal lungs of newborn babies even if corticosteroid is inhaled as aerosol particles.

Budesonide is a strong corticosteroid with a local anti-inflammatory effect commonly used as a nebulizing suspension for treating bronchial asthma in children.[6] However, inhalation of budesonide as aerosol particles with metered dose inhaler to newborn babies is technically difficult. Budesonide has to be deposited in lung tissues by inhalation. It needs to be successfully distributed to lung tissues and then dissolved in lung cells of newborn babies. Inhalation of budesonide to newborn babies has been shown to be ineffective because this method cannot deliver budesonide to distal lungs.[7-9] Thus, an alternative lung-targeted delivery method is needed. Some researchers have studied intratracheal administration of budesonide. However, intratracheal administration of budesonide alone is ineffective in decreasing inflammation of lungs.[10] One of the suggested methods to deliver budesonide to distal lungs is intratracheal administration of budesonide with vehicle.

Pulmonary surfactant is a mixture of phospholipids and surfactant proteins (SPs).[11,12] Phospholipids bilayers incorporated with SPs are typical structures of surfactant. These layers called surface films play an important role in adsorption and spreading of surfactant into distal lungs with excellent solubility related to the rich lipid content that can lower surface tension.[13] Intratracheal administration of drug with surfactant as a vehicle markedly facilitates the distribution and spreading of the drug compared to administration of the drug alone.[14-16] Thus, surfactant has been suggested as a vehicle to deliver drugs to distal lungs.[17] Huang et al.[18] revealed that the administration of budesonide with surfactant can facilitate pulmonary distribution of budesonide compared to budesonide alone using fluorescent dye in mice. The objective of the present study was to determine the effect of early intratracheal instillation of budesonide

with surfactant in preterm infants with severe RDS for alleviating BPD.

Material and Methods

The protocol of this study was reviewed and approved by the Institutional Review Board of Inje University Busan Paik Hospital (identification code: 19-0019) in accordance with the Declaration of Helsinki. A waiver of consent was granted for chart review without patient contact. Very low birth weight infants (VLBWIs) weighing less than 1,500 g who were admitted to the neonatal intensive care unit (NICU) of Busan Paik Hospital between January 2018 and December 2018 and diagnosed with severe RDS were enrolled. VLBWIs with severe RDS on chest radiography who required mechanical ventilator with fractional inspired oxygen (FiO$_2$) of > 0.5 were included. VLBWIs with chromosomal abnormality, congenital cardiac anomaly, or congenital pulmonary anomaly were excluded as these conditions could be confounding factors by interfering with pulmonary function and survival, making them unsuitable for evaluating the efficacy or safety of budesonide with surfactant. Included infants were allocated to two groups according to the medicine instilled: treatment group, surfactant and budesonide; and control group, surfactant only.

Study protocol

Clinical data were collected retrospectively from medical records. The diagnosis of RDS and the decision to perform surfactant replacement were left to the attending physician based on disease severity according to chest radiography and assisted ventilation.[19]

Surfactants were instilled into the trachea via an endotracheal tube using an orogastric tube. The treatment group was given a mixture of 0.25 mg/kg (1 mL/kg) of budesonide (Pulmicort nebulizing suspension, Astra Zeneca, Lund, Sweden) and 105 mg/kg (3 mL/kg) of calfactant (Infasurf®, ONY, Inc., Amherst, NY, USA) while the control group was given 105 mg/

kg of calfactant only. The concentration ratio of calfactant to budesonide was 35 mg: 0.25 mg or 140:1. The dosage of budesonide was determined according to previous studies.[20,21]

Infant demographic factors included gestational age, birth weight, gender, small for gestational age (SGA), Apgar score, and clinical risk index for babies (CRIB) II score. Maternal demographic factors included antenatal corticosteroids therapy, maternal gestational diabetes mellitus (GDM), maternal pregnancy-induced hypertension (PIH), and histologically confirmed chorioamnionitis. Outcomes associated with BPD included doses of surfactant, total duration of mechanical ventilation, duration of invasive ventilation, BPD, severity of BPD, and mortality. Outcomes associated with prematurity included patent ductus arteriosus (PDA), high grade intraventricular hemorrhage (IVH, ≥ grade 3), periventricular leukomalacia (PVL), high stage retinopathy of prematurity (ROP, ≥ stage 2), necrotizing enterocolitis (NEC, ≥ stage 2), and duration of hospital stay.

SGA was defined as a birth weight less than the tenth percentile on fetal-infant growth chart for preterm infants.[22] BPD was defined as an oxygen dependency at 36 weeks post-menstrual age with oxygen treatment for at least the first 28 days of life. BPD was subdivided to three groups according to its severity: mild BPD, breathing room air at 36 weeks post-menstrual age or discharge; moderate BPD, need for < 30% O2 at 36 weeks post-menstrual age or discharge; and severe BPD, need for ≥ 30% O2 with or without positive pressure ventilation or continuous positive pressure at 36 weeks post-menstrual age or discharge.[23] Sepsis was limited to positive blood culture with clinical signs of systemic infection.[24]

Statistical analysis

For nominal variables, Chi-square test or Fisher's exact test was performed. For continuous variables, Student's *t*-test or Mann-Whitney *U* test was performed. All statistical analyses were performed using SAS Enterprise Guide 3.0 (SAS Institute, Cary, NC, USA). *P*-values < 0.05 were considered significant. Data are given as mean ± standard deviation.

Results

Demographic factors

Among a total of 68 VLBWIs who were admitted to the NICU of Busan Paik Hospital during the period of enrollment (January 2018 through December 2018), 34 VLBWI who were diagnosed as severe RDS were enrolled: 16 in the treatment group (budesonide + surfactant) and 18 in the control group (surfactant). There were no significant differences in gestational age, birth weight, gender, SGA, Apgar score at 1 and 5 minutes, CRIB II score, antenatal corticosteroids, maternal GDM, maternal PIH, or chorioamnionitis between the treatment group and the control group (Table I).

Gestational age was $28^{+4} \pm 1^{+6}$ weeks (range: 25^{+2} - 32^{+0} weeks), and $28^{+2} \pm 2^{+0}$ weeks (range: 25^{+0} - 33^{+4} weeks), and birth weight was 1055 ± 247 g (range: 460 - 1400 g), and 1032 ± 255 g (range: 615 - 1400 g) in treatment group and control group, respectively.

BPD and mortality

Surfactant re-dosing was not different between the two groups (treatment group: 5 (31.3%); control group: 8 (44.4%); *p* = 0.429) (Table II). Total duration of mechanical ventilation (treatment group: 14.7 ± 15.7 days; control group: 26.3 ± 26.7 days; *p* = 0.162), and duration of invasive ventilation (treatment group: 10.6 ± 13.2 days; control group: 19.0 ± 25.2 days; *p* = 0.266) were not different between the two groups, though there were decreasing trends in the treatment group compared to those in the control group (reduction of 11 days and 8 days, respectively).

The incidence of BPD, mortality rate, and BPD or death were not different between two groups. There were decreasing trends of BPD

Table I. Demographic factors.

	Treatment group (n= 16)	Control group (n= 18)	p value
Gestational age, week^{+day}	$28^{+4} \pm 1^{+6}$ (25^{+2} - 32^{+0})	$28^{+2} \pm 2^{+0}$ (25^{+0} - 33^{+4})	0.641
Birth weight, g	1055 ± 247 (460 - 1400)	1032 ± 255 (615 - 1400)	0.791
Male, n (%)	8 (50.0)	9 (50.0)	1.000
SGA, n (%)	4 (25.0)	5 (27.8)	0.855
Apgar score at 1min	4.25 ± 1.53	4.44 ± 1.65	0.725
Apgar score at 5min	6.81 ± 1.05	6.67 ± 1.19	0.708
CRIB II	7.50 ± 2.71	7.56 ± 2.77	0.953
Antenatal corticosteroids, n (%)	16 (100.0)	18 (100.0)	
Maternal GDM, n (%)	0 (0)	2 (11.1)	0.169
Maternal PIH, n (%)	3 (18.8)	1 (5.6)	0.233
Chorioamnionitis, n (%)	7 (43.8)	5 (27.8)	0.331

SGA: small for gestational age, CRIB: clinical risk index for babies, GDM: gestational diabetes mellitus, PIH: pregnancy induced hypertension.

Table II. Bronchopulmonary dysplasia and mortality.

	Treatment group (n= 16)	Control group (n= 18)	p value
Surfactant re-dosing, n (%)	5 (31.3)	8 (44.4)	0.429
Total duration of mechanical ventilation, days	14.7 ± 15.7	26.3 ± 26.7	0.162
Duration of invasive ventilation, days	10.6 ± 13.2	19.0 ± 25.2	0.266
BPD, n (%)	5 (33.3)	8 (53.3)	0.269
BPD (moderate to severe), n (%)	2 (13.3)	4 (26.7)	0.361
BPD (severe), n (%)	0 (0.0)	2 (13.3)	0.143
Death, n (%)	1 (6.3)	4 (22.2)	0.189
BPD or death, n (%)	6 (37.5)	11 (61.1)	0.169

BPD: bronchopulmonary dysplasia.

(treatment group: 33.3%; control group: 53.3%; $p = 0.269$), mortality rate (treatment group: 6.3%; control group: 22.2%; $p = 0.189$), and BPD or death (treatment group: 37.5%; control group: 61.1%; $p = 0.169$). However, we couldn't find statistical differences.

There was no severe BPD in the treatment group whereas 13.3% had severe BPD in the control group

Outcomes Associated with Prematurity

Incidences of ligation of PDA, high grade IVH (≥ grade 3), PVL, and NEC (≥ stage 2) were similar between the two groups (Table III).

Incidence of high stage ROP (≥ stage 2) was not different between the two groups, though there was a decreasing trend in the treatment group compared to the control group (treatment group: 12.5%; control group: 33.3%; $p = 0.166$). The duration of hospital stay was longer in the control group with statistical significance (treatment group: 73.2 ± 19.5 days; control group: 91.9 ± 20.3 days; $p = 0.018$).

Discussion

Budesonide has a structure similar to cholesterol, a sterol component of mammalian cell membrane. Thus, budesonide is lipid

Table III. Outcomes associated with prematurity.

	Treatment group (n= 16)	Control group (n= 18)	p value
Ligation of PDA, n (%)	1 (6.3)	3 (16.7)	0.347
IVH (≥ grade 3), n (%)	1 (6.3)	1 (5.9)	0.965
PVL, n (%)	1 (6.3)	0 (0.0)	0.295
ROP (≥ stage 2), n (%)	2 (12.5)	5 (33.3)	0.166
NEC (≥ stage 2), n (%)	0 (0.0)	0 (0.0)	
Hospital stay, days	73.2 ± 19.5	91.9 ± 20.3	0.018 *

PDA: patent ductus arteriosus, IVH: intraventricular hemorrhage, PVL: periventricular leukomalacia, ROP: retinopathy of prematurity, NEC: necrotizing enterocolitis.
*p-values <0.05.

soluble. It has strong affinity to airway and lung tissues. After budesonide is absorbed in lung tissue, it can conjugate with fatty acid and make fatty acid esters of budesonide. This conjugated form of budesonide is lipophilic, thus prolonging intracellular retention with very slow release of free budesonide. Other corticosteroids such as fluticasone propionate or beclomethasone dipropionate could not make fatty acid esters, which is why budesonide has longer pharmacological effects in airway tissues than other corticosteroids.[25,26] This is also the reason why topically inhaled budesonide has markedly better uptake and retention in airway tissues than non-inhaled type of corticosteroid.

We calculated the dosage of budesonide according to a previous study reporting that if the concentration ratio of surfactant and budesonide was 50:1 or more, then budesonide would not disturb the role of surfactant to decrease surface tension.[20] Therefore, calfactant (105 mg/kg, 35 mg/ml) and budesonide (0.25 mg/kg, 0.25 mg/ml) at a concentration ratio of 35 mg:0.25 mg or 140:1 (>50:1) was chosen so that surfactant function would not be disturbed by budesonide.

Systemically administered corticosteroid can cross the blood brain barrier and result in high concentrations in brain tissues, especially in hippocampus which contains high density of corticosteroid receptors.[3] Corticosteroid in brain tissues can change cerebral blood flow abnormally. It can cause ischemia and exert adverse effects on brain growth, especially in the hippocampus which is a critical area for memory and learning. This is why systemic administered corticosteroid can cause neurodevelopmental side effects.

To improve the local effect and decrease the systemic side effect of steroid, another route of drug delivery has been studied. Shah et al.[27-29] have reviewed the effect of inhaled versus systemic corticosteroids in preterm infants to prevent BPD with meta-analysis over three times. There were differences between study protocols. However, most trials administered inhaled budesonide or beclomethasone dipropionate by metered dose inhaler and a spacer device or intravenous dexamethasone. They found that there were no significant differences in effectiveness or side effects between inhaled and systemic corticosteroids. All reviewed randomized controlled trials did not use surfactant as a vehicle. They administered inhaled corticosteroid with metered dose inhaler, a well-known ineffective delivery system of drugs to the airway and lung in preterm infants. They suggested that a better mode of delivering inhaled corticosteroid could be more effective without increasing side effects.

Researchers have investigated intratracheal administering steroid with surfactant as a vehicle in animal models. Fajardo et al.[30] have revealed that intratracheal instillation of budesonide with surfactant is more effective than intratracheal

budesonide alone or inhaled budesonide in ventilated rabbit. Chen et al.[31] have administered intratracheal high dose dexamethasone (0.5 mg/kg) with surfactant (beractant, 50 mg/kg or 100 mg/kg) in rats with acute lung injury. They concluded that intratracheal administration of high dose dexamethasone with surfactant could alleviate inflammation of lungs compared to intratracheal administration of surfactant alone. In another animal study with rats, intratracheal administered low dose dexamethasone (0.04 mg/kg, 1 mL/kg) with surfactant as a vehicle (beractant, 10 mg/kg, 5 mL/kg, diluted to 2 mg/mL) improved the distribution of dexamethasone to peripheral lungs compared to the use of saline as a vehicle. It also decreased circulating concentration of dexamethasone compared to systemic administration of dexamethasone.[32] They suggested that the most effective mode of delivering corticosteroid could be intratracheal instillation. Yang et al.[33] have reported that intratracheal administered budesonide (0.5mg/kg) with surfactant (beractant, 100mg/kg) in neonatal piglets can improve oxygenation and histological lung injuries during acute period (in four hours) compared to no treatment, although it has no difference compared to treatment with surfactant alone. However, budesonide did not disturb surfactant function of decreasing surface tension. They reported updated results[34] that intratracheal administration of budesonide (0.25 mg/kg) with surfactant (beractant, 100 mg/kg) in neonatal piglets with severe RDS could improve oxygenation and histological lung injuries over 24 hours constantly. Differences from their previous study were suggested by too short observation periods such as four hours to alleviate oxygenation and histological changes. Considering that BPD is a lung injury syndrome with chronic and multifactorial pathogenesis, observation over a longer period might be more reliable for evaluating the effectiveness of drugs to alleviate BPD.

In another study by Ricci et al.[10], intratracheal administration of 0.25 mg/kg (1 mL/kg) budesonide with 200 mg/kg (2.5 mL/kg) surfactant improved tidal volume, gas exchange, and lung compliance in premature rabbits. They used poractant alfa as a vehicle, not beractant. The concentration ratio of poractant alfa to budesonide was 80 mg: 0.25mg or 320:1 (≥ 50:1). Thus, surfactant function might not be disturbed by budesonide. Also, administration of budesonide with surfactant decreased lung inflammation compared to budesonide or surfactant alone. This meant that budesonide did not disturb surfactant function, and surfactant can be a good vehicle to deliver budesonide to distal lungs to alleviate BPD. Roberts et al.[35] have reported that, when budesonide with surfactant is administered intratracheally in premature lambs, budesonide and its metabolites (16α-hydroxy prednisolone, or budesonide-palmitate) could not be detected in the brain tissue. This means that intratracheally administered budesonide with surfactant is deposited in the airway and lung tissues mostly. It is less absorbed systemically, showing low plasma concentration. It could not cross the blood brain barrier, resulting in minimal neurologic side effects with only local anti-inflammatory effect in airway tissue

Yeh et al.[20] have conducted a prospective randomized controlled trial (RCT) with intratracheal instillation of 0.25 mg/kg (1 mL/kg) budesonide with 100 mg/kg (4 mL/kg) surfactant (beractant) in 116 VLBWIs with severe RDS. They reported that this therapy could decrease the incidence of BPD and mortality and increase survival without BPD compared to surfactant only therapy without significantly increasing adverse events. In a follow up study[21] of this trial at the age of two to three years of these enrolled preterm infants (67 VLWBIs, 35 budesonide treated and 32 control), there were no significant differences in the incidence of hospital admission by respiratory causes or physical growth such as body weight, height, or head circumference between budesonide with surfactant treated group and surfactant only treated group. There were no significant differences in Mental Development Index (MDI) or Psychomotor Development Index (PDI) by Bayley Scales of Infant Development

(BSID) II between budesonide with surfactant treated group and surfactant only treated group either. Authors concluded that there were no long-term adverse events associated with intratracheal instillation of budesonide with surfactant. In a prospective multicenter RCT,[36] 265 VLBWIs in three tertiary NICUs in USA and Taiwan were enrolled. Budesonide (0.25 mg/kg, 1 mL/kg) with surfactant (beractant, 100 mg/kg, 4 mL/kg) treatment decreased the incidence of BPD or mortality compared to surfactant (100 mg/kg) only treatment (66% to 42%, $p < 0.001$). Also, levels of interleukins (IL-1, -6, and -8) in tracheal aspirates were lower in budesonide with surfactant treatment group compared to those in surfactant only treatment group, indicating that budesonide could ameliorate inflammation. Physical growth and neurodevelopmental impairment were similar between the two groups in a follow up study at the age of two to three years of these enrolled preterm infants. Researchers reported that intratracheal administration of budesonide with surfactant decreased BPD or mortality without significantly increasing acute or long-term adverse effects.

The differences of this study from Yeh's studies[20,21,36] are that we used different preparation of surfactant (calfactant in this study vs. beractant in Yeh's studies), and different concentration ratio of surfactant to budesonide (140:1 in this study vs. 100:1 in Yeh's studies).

We couldn't find differences in the BPD, mortality rate, BPD or mortality, ventilator duration, and high stage ROP between the two groups. There were decreasing trends of those parameters in the treatment group compared to the control group without statistical differences. Hospital stay was decreased in the treatment group compared to control group with statistical significance. It might be due to decreasing trends of BPD and ventilator duration in the treatment group. The limitation of this study is that the small number of enrolled patients to prove statistical differences.

In conclusion, early intratracheal administration of budesonide and surfactant in preterm infants with severe RDS might decrease BPD and mortality without disturbing surfactant function. Further prospective randomized controlled studies with different preparations of surfactants with a large number of preterm infants are required. To the best of our knowledge, this is the first study administering budesonide with calfactant in preterm infants having severe RDS.

The protocol of this study was reviewed and approved by the Institutional Review Board of Inje University Busan Paik Hospital. A waiver of consent was granted for chart review without patient contact.

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