ERJ Express. Published on October 18, 2006 as doi: 10.1183/09031936.00029206

Clarithromycin in The Treatment of RSV Bronchiolitis:

A Double-Blind, Randomized, Placebo-Controlled Trial

Fulya Tahan[a], Ahmet Ozcan[b] , Nedret Koc[c]

a: Erciyes University School of Medicine, Department of Pediatric Allergy, Kayseri, TURKEY

b: Erciyes University School of Medicine, Department of Pediatry, Kayseri, TURKEY

c: Erciyes University School of Medicine, Department of Microbiology, Kayseri, TURKEY

Address for correspondence:

Fulya Tahan, MD,

Erciyes University School of Medicine, Department of Pediatric Allergy, Kayseri, TURKEY.

e-mail: tahanfulya@yahoo.com

phone: + 90 352 437 4901/22117

fax: +90 352 437 5825

Abstract

Respiratory syncytial virus bronchiolitis is the most common lower respiratory tract infection in infancy. There is no effective therapy for RSV bronchiolitis.

To investigate the efficacy of clarithromycin in the treatment of RSV bronchiolitis, we conducted a randomized, double-blind, placebo controlled trial comparing clarithromycin with placebo in 21 infants with a diagnosis of RSV bronchiolitis. The infants were

randomized to receive clarithromycin daily for 3 weeks or placebo. Levels of IL-4, IL-8, eotaxin, and IFN-gamma were determined in plasma before, and after treatment using ELISA. Six months following treatment, parents were surveyed as to whether or not their child had had wheezing within the previous 6 months.

Treatment with clarithromycin was associated with a statistically significant reduction in length of hospital stay , duration of need for supplemental oxygen and need for beta-2 agonist treatment. There were significant decreases in plasma IL-4, IL-8 and eotaxin levels after 3 weeks of treatment with clarithromycin. Readmission to the hospital within six months after discharge was significantly lower in the clarithromycin group.

Clarithromycin had statistical significant effects on the clinical and laboratory findings in RSV bronchiolitis. Clarithromycin treatment may be helpful in reducing the shortterm effects of RSV bronchiolitis.

Key Words: Respiratory syncytial virus, bronchiolitis, IL-4, IL-8, eotaxin, clarithromycin

Introduction

Respiratory syncytial virus (RSV) bronchiolitis is the most common lower respiratory tract infection in infancy, occurring in 90 % of children before the age of 2 years (1, 2). Severe RSV infection in the first 6 months of life is often followed by recurrent childhood wheezing (3).

Airway inflammation in RSV bronchiolitis is a multicellular process involving epithelial cells, eosinophils, and neutrophils. Epithelial cells have always been thought of as major contributors to the inflammatory process of the airways during RSV infection. RSV causes

widespread damage to bronchial epithelium and stimulates epithelial cells to secrete a wide range of pro-inflammatory cytokines and chemokines (4). The increase in virus-induced chemokines recruits leukocytes to the airway and could increase binding of neutrophils and eosinophils to epithelial cells.

IL-8 is a key chemokine produced by RSV infected airway cells and is involved in the activation and recruitment of neutrophils (5). Neutrophils play a major role in the pathophysiology of RSV bronchiolitis (6).

RSV infection can enhance the influx of eosinophils into the lungs (7). Kristjansson et al showed that RSV infections during early infancy promotes infiltration and activation of eosinophils (8). In vitro studies have indicated that RSV infection results in the release of high concentrations of RANTES and MIP-1α (9-12).

RSV infection may influence the development of asthma by enhancing allergic sensitisation in the developing lung. The normal respiratory tract has evolved in close contact with aeroallergens. Sensitization to aeroallergens is unlikely to occur through an intact mucosal epithelium. Disturbance of the respiratory mucosal surface during viral infection may allow allergens to gain access to the subepithelial layer and interact with antigen-presenting cells and inflammatory cells, leading to allergic sensitization (13).

To break the apparent link that exists between RSV bronchiolitis and childhood asthma, effective therapy aganist its short-term effects is necessary.

The treatment of infants with bronchiolitis has been largely supportive, with supplemental oxygen, minimal handling of the infant, and the use of intravenous fluids and ventilatory support where necessary (14). The role of bronchodilators is controversial (14). The American Academy of Pediatrics (AAP) does not recommend using corticosteroids for the treatment of RSV symptoms. Despite many attempts to find effective treatments for patients with RSV bronchiolitis, no consistently effective therapy has been described.

Macrolides are widely used in the treatment of infectious diseases, including respiratory infections (15). There is increasing evidence of an anti-inflammatory effect of macrolides.

Clarithromycin, one of the newer macrolides, has been shown to have immunomodulatory effects. Possible mechanisms of the antiinflammatory effects of clarithromycin include inhibition of neutrophil migration and proinflammatory cytokines, increase in phagocytosis and natural killer cell activity, and induction of eosinophil apoptosis (16-20). Clarithromycin suppresses the production of proinflammatory cytokines via inhibition of NF-KB activation (21).

Because RSV infection initiates an immune inflammatory response that may produce long lasting harmfull effects, we hypothesized that we could modify the course of the disease and prevent wheezing after bronchiolitis by administering macrolides to infants during an acute episode of RSV bronchiolitis. To investigate this, we studied the use of 3 weeks of macrolide therapy in the treatment of RSV bronchiolitis in a double-blind, randomized, placebo-controlled trial. We measured the eosinophilic CC chemokine eotaxin (which is thought to have a role in eosinophilic inflammation), and the CXC chemokine IL-8 (thought to have a central role in neutrophilic inflammation). Knowing that severe RSV infections during early infancy are associated with the excessive production of Th2 cytokines (22, 23), we measured the Th2 cytokine IL-4 (thought to have a central role in Th2 mediated diseases) and the Th1 cytokine IFN-gamma.

We hypothesized that a three week course of clarithromycin therapy would result in a reduction in hospital length of stay (LOS) and plasma IL-4, IL-8 and eotaxin levels and enhanced production of IFN-gamma.

Material and Methods

Patients

In the RSV season of January to April 2005, 30 infants ≤ 7 months of age with documented respiratory tract infection with RSV admitted to the Department of Pediatrics at the Erciyes University Hospital in Kayseri, Turkey were enrolled in the study. Infants with a first episode of wheezing requiring hospitalization and with a clinical diagnosis of bronchiolitis were considered for entry into the study. Bronchiolitis was diagnosed based on clinical findings , including wheezing or wheezing with crackles and respiratory distress with retractions. Infants with cardiac disease, cystic fibrosis, or chronic neonatal lung disease associated with

prematurity were excluded. Infants were also excluded if they had received corticosteroids within 24 hours before presentation or had received bronchodilators within 4 hours before presentation. The study was approved by the Erciyes University Hospital Ethics Committee. Written informed consent was obtained from the parents before enrollment.

Bronchiolitis Diagnosis and Treatment

All children admitted to hospital with bronchiolitis were treated according to the same clinical pathway to minimize the variability of the results. A nasopharyngeal aspiration sample (NPA) was obtained routinely from all patients for detection of respiratory syncytial virus. RSV infection was diagnosed by direct immunofluorescent staining of the NPA. An infant was considered ready for discharge if he or she had not received supplemental oxygen for 10 hours, had minimal or no chest retractions, and was feeding adequately without the need for intravenous fluids. Supplemental oxygen was administered for oxygen saturation (SpO_2) less than 94 % as determined by pulse oximetry (Trusat pulse oximeter, Datex-Ohmeda, Louisville, CO, USA). Supplemental oxygen was discontinued when SpO_2 was consistently above 93 %, or when the infant's condition had been stable for four hours and he/she was starting to tolerate oral feeding.

Intravenous fluids were administered if supplemental oxygen was required, respiratory rate was above 60 per minute, or oral intake was inadequate. When the infant was able to tolerate oral feeding, the use of intravenous fluids was stopped.

The infants received beta-2 agonist treatment based on oxygen saturation, respiratory rate, and respiratory effort. Infants received beta-2 agonist treatment if oxygen saturation was less than 94 percent, respiratory rate was above 60 per minute, there was the presence of wheezing on auscultation of the chest or respiratory distress with retractions.

Randomization and Investigational Therapy

After written informed parental consent had been obtained, and the NPA had been found to be positive for RSV, the infants were randomized by a single study nurse to receive clarithromycin (15 mg/kg) or placebo daily for 3 weeks. We used simple randomization (24). Patients, parents and investigators were kept blinded to the randomization until the completion of the study. The primary outcome was hospital length of stay (LOS). Secondary outcomes included changes in the IL-4, IL-8, eotaxin and IFN-gamma levels, readmission rate and wheezing after discharge.

Each infant was assigned one bottle of solution containing either clarithromycin (clarithromycin, silicon dioxide, saccharose, kxantan zamk, tutti frutti aroma, potassium sorbate, citric acid, titanium dioxide, maltodextrin, water) or placebo (silicon dioxide, saccharose, kxantan zamk, tutti frutti aroma, potassium sorbate, citric acid, titanium dioxide, maltodextrin, water).

Clinical Data

Detailed clinical histories in the clinical pathway, including the duration of symptoms before presentation at the hospital, the medical history, the infant's ability to feed, previous medications, parental smoking history and family history of atopy were recorded. Observations at admission included respiratory rate and heart rate while the infant was quiet, temperature, respiratory effort, SpO_2 while breathing room air, presence or absence of wheezing or crackles on auscultation of the chest, and level of hydration. Each infant's condition was classified as mild, moderate, or severe according to a severity score (14) calculated from the SpO_2, respiratory rate, and respiratory effort observed at admission (Table

1). Six months following completion of clarithromycin or placebo therapy parents were asked whether or not their child had experienced wheezing during the previous 6 months.

Laboratory Studies

The kit used to detect RSV by direct immunofluorescent staining of nasopharyngeal aspirate was obtained from Bio-Rad (Monofluo screen RSV, Marnes-la-Coquette, France).

Peripheral blood (5 ml) was obtained from the all children pretreatment, and after 3 weeks of macrolide or placebo treatment. Samples were centrifuged at 2000 rpm for 30 min and serum frozen at –20 $^{\circ}$C for later ELISA assay. Serum samples were analysed both during the acute phase of the disease and after 3 weeks of treatment for IL-4, IL-8, eotaxin and IFN-gamma.

Total IgE and eosinophil counts were obtained and skin testing was done with a battery of 25 antigens with appropriate histamine positive and saline/diluent negative controls on the upper back of the children at presentation. Reactions with an induration >3 mm that of the negative control were considered positive.

IgE levels were measured with Uni-Cap technology in accordance with the specifications of the manufacturer (Pharmacia, Kalamazoo, MI). Eosinophil counts were determined from Coulter Counter leukocyte measurements.

Chemokine measurements

The ELISA kits used to detect IL-4, IL-8, eotaxin and IFN-gamma levels were obtained from Biosource (Camarillo, CA, USA). The sensitivity of kits for IL-4, IL-8, Eotaxin and IFN-gamma were 2 pg/ml, 5 pg/ml, 2.2 pg/ml, 4 pg/ml respectively.

Statistical Analyses

Chemokine levels in the plasma were compared by Mann Whitney U test and Wilcoxon tests, depending upon the distribution of the data. Gender, parental smoking, readmission to the

hospital, duration of wheezing at admission, severity of disease, requiring supplemental oxygen, skin test positivity and family history of atopy were compared by chi square test. A p value of less than 0.05 was considered significant.

Results

A total of 30 infants were assigned to treatment: 15 to clarithromycin and 15 to placebo. During the study period nine patients (three in the clarithromycin group, six placebo) were excluded from the study because of steroid treatment during hospitalization. There were no significant differences between the groups at randomization in terms of demographic variables. There were no significant differences at admission between the groups in the duration of wheezing and severity of the disease (Table 1). Likewise, there were no significant differences between the groups with respect to the number of eosinophils and plasma IgE levels ($p>0.05$ Mann Whitney U test, Table 1).

Primary end points

Treatment with clarithromycin was associated with a significant reduction in LOS (51 hours vs 88 hours, $p<0.05$, Table 2). Duration of need for supplemental oxygen and intravenous fluids was higher in the placebo group (31 vs 72 hours, <0.05, and 26 vs 56 respectively, Mann Whitney U test, Table 2).

There was a significant difference in use of beta-2 agonist treatment between the clarithromycin and placebo group. Among infants receiving beta-2 agonist treatment, those in the placebo group received it longer than those in the clarithromycin group (5 days vs 7days, <0.05 Mann Whitney U test, Table 2).

Secondary end points

Comparison of pretreatment plasma chemokine levels revealed that there was no significant difference between the clarithromycin and placebo groups ($p>0.05$, Mann Whitney U test).

Three weeks of clarithromycin therapy was associated with significant changes in the plasma chemokine levels. There were significant decreases in the plasma IL-4, IL-8 and eotaxin levels following clarithromycin therapy ($p<0.05$, Wilcoxon, Figure 1, 2, 3). There were no

differences in plasma eotaxin, IL-4 and IL-8 levels ($p>0.05$, Wilcoxon, Figure 1, 2, 3). IFN-gamma levels were below the limit of detection in all infants.

Five patients were readmitted to the hospital with wheezing within six months after discharge, four (44 %) in the placebo group, one (8.3 %) in the clarithromycin group (<0.05, chisquare, Table 2). For the children that were not readmitted to the hospital, their parents were asked about wheeze at 6 months by phone. None reported wheezing during that time.

Discussion

To our knowledge, this is the first study investigating the effect of clarithromycin treatment in RSV bronchiolitis. The study was designed to determine whether clarithromycin treatment had any clinical and/or laboratory effect on clinical and/or biomarker outcomes in RSV bronchiolitis. In order to explore a potential immunomodulatory effect of clarithromycin, we measured plasma levels of IL-4, IL-8, eotaxin, IFN-gamma. We have shown that clarithromycin, when compared to placebo, is capable of significant changes in some of these parameters.

In RSV bronchiolitis, treatment with clarithromycin had a statistically significant effect on LOS, use of beta-2 agonist treatment and plasma IL-4, IL-8 and eotaxin levels.

Clarithromycin is widely used in the treatment of infectious diseases, and has antibacterial effect. The question is whether or not the clinical improvement is related to its antibacterial effect. It's known that acute bronchiolitis is predominantly a viral disease. RSV is responsible for more than 50 % of cases (25). Other agents include parainfluenza, adenovirus, and occasionally other viruses. There is no evidence of a bacterial cause for bronchiolitis (25). In our study group, we did not investigate bacterial infection, because all subject's leukocyte counts were normal, CRP levels were negative, and all were RSV positive.

How could clarithromycin act in the short term to effect changes in LOS and use of beta 2 agonist treatment in bronchiolitis? We considered that suppressive effects of clarithromycin on the plasma IL-4, IL-8 and eotaxin levels may have a role in suppression of airway hyperresponsiveness, or may inhibit cholinergic neuroeffector transmission in human airway smooth muscle, thereby influencing bronchial tone.

We know that RSV could potentially cause increased airway responsiveness by enhancing parasympathetic bronchoconstrictive responses (26). The documentation that macrolides attenuate the contractile response of human isolated bronchial strips to electrical field

stimulation (18) leads us to hypothesize that macrolides might influence bronchial tone by inhibiting cholinergic neuroeffector transmission in human airway smooth muscle.

Airway hyperresponsiveness appears to be one manifestation of the airway inflammation induced by RSV. It has been shown that a correlation exists between numbers of mast cells, eosinophils and neutrophils and the degree of airway hyperresponsiveness (27). It has been further shown that enhanced IL-4, IL-8 and eotaxin has a role in development of airway inflammation and hyperresponsiveness (28, 29, 30). Pitrez, et al suggested that there were significant correlations between IL-4 levels in blood and airway secretions (31). We found significant decreases in plasma IL-4 levels after 3 weeks of clarithromycin therapy. IL-4 is a critical cytokine in the mediation of allergic airway inflammation. IL-4 is critical in the switching of B cells to IgE production. It also promotes mucus hypersecretion, and vascular cell adhesion molecule-1 (VCAM-I) expression in endothelial cells, resulting in the recruitment of eosinophils (32).

It is known that enhanced IL-4 has a critical role in development of airway hyperresponsiveness (28). Inhalation of IL-4 causes the development of sputum eosinophilia and increased airway hyperreactivity (33). We hypothesized that clarithromycin, by decreasing plasma IL-4 levels, may reduce need for beta-2 agonist therapy in RSV bronchiolitis. Indeed, infants in the placebo group required significantly longer duration of beta-2 agonist therapy than infants in the clarithromycin group.

Neutrophil mediated inflammation is involved in the augmentation of bronchial reactivity in RSV bronchiolitis (26, 29, 34, 35). IL-8 and LTB_4 are known neutrophil chemotactic factors and play an important role in neutrophilic airway inflammation (36). Previous studies demonstrated that one of the antiinflammatory mechanisms of macrolides relates to the inhibition of IL-8 production (36, 37). Macrolides also inhibit formation of LTB_4 and neutrophil infiltration into lung tissue and reduce the formation of superoxide by neutrophils

(38-42). Steroids fail to down-regulate RSV-induced IL-8 secretion in infants. This may explain why steroid therapy is unsuccessful in RSV bronchiolitis (43).

We found significant decreases in plasma eotaxin levels following clarithromycin therapy. Eotaxin is highly specific for eosinophil recruitment (30). Macrolides attenuate the release of eotaxin, GM-CSF, and RANTES (30). Lung fibroblasts are an important source of eosinophil chemotactic activity; inhibitory effects of erythromycin on eosinophil chemotactic cytokine release by lung fibroblasts may be one of the mechanisms of decreased airway hyper-responsiveness and the resulting amelioration of disease activity following therapy with that agent (30). Macrolides may also protect epithelial cells at inflammed sites by inhibiting the release of reactive oxygen species from eosinophils (44).

Severe RSV infections during early infancy are associated with the excessive production of Th2 cytokines (22, 23), which has been suggessted as a risk factor for the development of asthma and allergic sensitization (45). Macrolides may normalize the T helper type lymphocyte balance (38). They regulate immunologic activities by enhancing production of IFN-gamma and by reducing production of IL-4, IL-5 (46). Treatment that restores the Th1/Th2 cytokine balance to the relative type 1 predominance may ameliorate shorth term and long term effects of RSV disease.

We have no data concerning the long term effects of clarithromycin in RSV bronchiolitis. Further investigations will be required to understand the long term effects of clarithromycin, especially reducing recurrent wheezing, allergic sensitisation and asthma,.

Numerous studies have shown that 75 % patients with RSV bronchiolitis exhibit recurrent wheezing or pulmonary function abnormalities years later (47, 48). As discussed earlier, RSV infection may enhance allergic sensitisation in the developing lung through disruption of the respiratory epithelium. Macrolides promote the reparative process in the chronically inflamed upper and lower respiratory tract, and are associated with salutary tissue reparative effects in

patients with chronic inflammatory sinopulmonary diseases such as chronic sinusitis, asthma, bronchiectasis, cystic fibrosis, and diffuse panbronchiolitis (49). Suppressing IgE (49) and tissue reparative effects of macrolides may partly protect against allergic sensitisation.

Further investigations of larger groups of children will be required to elucidate potential effects of macrolides in this area.

In conclussion, treatment with clarithromycin had statistically significant effects on LOS, duration of need for supplemental oxygen, and rate of readmission to the hospital within six months after discharge in RSV bronchiolitis. Suppressive effects of clarithromycin on the plasma IL-4, IL-8 and eotaxin levels may have a role in suppression of airway hyperresponsiveness and epithelial cell damage leading to a reduction in postbronchiolitic symptoms and allergic sensitisation. Effective therapy against the short-term effects of RSV bronchiolitis could be important in reducing subsequent morbidity. This study should encourage further studies to confirm the use of clarithromycin in RSV bronchiolitis, especially infants under 6 months of age.

References

1. Phelan P, Olinsky A, Robertson C. Clinical patterns of acute respiratory infections. Oxford, England: Blackwell Science, 1994;52-93.
2. Glezen WP, Denny FW. Epidemiology of acute lower respiratory disease in children. N Engl J Med 1973;288:498-505.
3. Martinez FD, Wright AL, Taussing LM, Holberg CJ, Hallonen M, Morgan WJ. Asthma and wheezing in the first six years of life. N Engl J Med 1995;332:133-138.
4. Folkerts G, Busse WW, Nijkamp FP, Sorkness R, Gern JE. Virus-induced airway hyperresponsiveness and asthma. Am J Respir Crit Care Med 1998;157:1708-1720.
5. Bataki EL, Evans GS, Everard ML. Respiratory syncytial virus and neutrophil activation. Clin Exp Immunol 2005;140;470-477.
6. Evans GS, Pettitt E, Elphick HE, Jones A, Everard ML. Identification of netrophil elastase as the Major uninhibited protease in the airways of children with RSV positive bronchiolitis. Am J Respiratory Crit Care Med 1999;159:189.
7. Schwarze J, Hamelmann E, Bradley KL, Takeda K, Gelfand EW. Respiratory syncytial virus infection results in airway hyperresponsiveness and enhanced airway sensitization to allergen. J Clin Invest 1997;100:226.
8. Kristjansson S, Bjarnarson SP, Wennergren G, Palsdottir AH, Arnadottir T, Haraldsson A, Jonsdottir I. Respiratory syncytial virus and other respiratory viruses during the first 3 months of life promote a local Th2 like response. J Allergy Clin Immunol 2005;116:805-811.

9. Becker S, Quay J, Soukup J. Cytokine (tumor necrosis factor, IL-6, IL-8) production by respiratuvar syncytial virus-infected human alveolar macrophages. J Immunol 1991;147:4307-4312.

10. Noah TL, Becker S. Respiratory syncytial virus-induced cytokine production by a human bronchial epithelial cell line. Am J Physiol 1993;265:472-478.

11. Arnold R, Humbert B, Werchau H, Gallati H, Konig W. Interleukin-8 interleukin-6, and soluble tumor necrosis receptor type I release from a human pulmonary epithelial cell line (A549) exposed to respiratory syncytial virus. Immunol 1994;82:126-133.

12. Olszewska-Pazdrak B, Casola A, Saito T, Alam R, Crowe SE, Mei F, Ogra PL, Garofalo RP. Cell-specific expression of RANTES, MCP-1, and MIP-1α by lower airway epithelial cells and eosinophils infected with respiratory syncytial virus. J Virol 1998;72:4756-4764.

13. Mallia P, Johnston SL. Respiratory viruses: do they protect from or induce asthma? Allergy 2002;57:1118-1129.

14. Wainwright C, Altamirano L, Medico-Cirujano, Cheney M, Cheney J, Barber S, Price D, Moloney S, Kimberley A, Woolfield N, Cadzow S, Fiumara F, Wilson P, Mego S, Vandevelde D, Francis P. A multicenter, randomized, double-blind, controlled trial of nebulized epinephrine in infants with acute bronchiolitis. N Engl J Med 2003;349:27-35.

15. Ferrara G, Losi M, Franco F, Corbetta L, Fabbri LM, Richeldi L. Macrolides in the treatment of asthma and cystic fibrosis. Respir Med 2005;99:1-10.

16. Ianaro A, Ialenti A, Maffia P, Sautebin L, Rombola L, Carnuccio R. Anti-inflammatory activity of macrolide antibiotics. J Pharmacol Exp Ther 2000;;292:156-163.

17. Anderson R, Theron AJ, Feldman C. Membran-stabilizing, anti-inflammatory interactions of macrolides with human neutrophils. Inflammation 1996;20:693-705.

18. Labro MT, Anti-inflammatory activity of macrolides: a new therapeutic potential? J Antimicrob Chemother 1998;41:37-46.

19. Morikawa K, Watabe H, Araake M, Morikawa S. Modulatory effect of antibiotics on cytokine production by human monocytes in vitro. Antimicrob Agents Chemother 1996;40:1366-1370.

20. Adachi T, Motojima S, Hirata A, Fukuda T, Kihara N, Kosadu A. Eosinophil apoptosis caused by theophylline, glucocorticosteroids, and macrolides after stimulation wiith IL-5. J Allergy Clin Immunol 1996;98:207-215.

21. Ichiyama T, Nishikawa M, Yoshitomi T. Clarithromycin inhibits NF-KB activation in human peripheral blood mononuclear cells and pulmonary epithelial cells. Antimicrob Agents Chemother 2001;45:44-47.

22. Roman M, Calhoun WJ, Hinton KL, Avendano LF, Simon V, Escobar AM. Respiratory syncytial virus infection in infants is associated with predominant Th2 like response. Am J Respir Crit Care Med 1997;156:190-195.

23. Kristjansson S, Bjarnarson SP, Wennergren G, Palsdottir AH, Arnadottir T, Haraldsson A, Jonsdottir I. Respiratory syncytial virus and other respiratory viruses during the first 3 months of life promote a local Th2 like response. J Allergy Clin Immunol 2005;116:805-811.

24. Beller EM, Gebski V, Keech AC. Randomisation in clinical trials. MJA 2002;177:565-567.

25. Goodman D. Inflammatory disorders of the small airways. In Behrman RE, Kliegman RM, Jenson HB, editors. Textbook of Pediatrics. 17th edition. Philadelphia, Saunders, 2003. p. 1414-1416.

26. Everard MI, Swarbrick A, Wrightham M, McIntyre J, Dunkley C, James PD, Sewell HF, Milner AD. Analysis of cells obtained by bronchial lavage of infants with respiratory syncytial virus infection. Arch Dis Child 1994;71:428-432.

27. Paul M O'Byrne. Mechanisms of airway hyperresponsiveness.In Holgate ST, Busse WW, editors. Inflammatory mechanisms in asthma. New York: Marcel Dekker, Inc;1998.p. 891-901.

28. Schwarze J, Cieslewicz G, Joetham A, Ikemura T, Makela MJ, Dakhama A, Shultz LD, Lamers MC, Gelfand EW. Critical roles for interleukin-4 and interleukin-5 during respiratory syncytial virus infection in the development of airway hyperresponsiveness after airway sensitization. Am J Respir Crit Care Med 2000;162:380-386.

29. Abu-Harb M, Bell F, Finn A, Rao WH, Nixon L, Shale D, Everard ML. Interleukin-8 and neutrophil elastase levels in the respiratory tract of infants with RSV bronchiolitis. Eur Respir J 1999;14:139-143.

30. Sato E, Nelson DK, Koyama S, Hoyt JC, Robbins RA. Erythromycin modulates eosinophil chemotactic cytokine production by human lung fibroblasts in vitro. Antimicrobial Agents Chemotherapy 2000;45:401-406.

31. Pitrez PM, Ponzi D, Machado DC, Bauer ME, Jones MH, Stein RT. Discrepancy between cytokine production from peripheral blood mononuclear cells and nasal secretions among infants with acute bronchiolitis. Ann Allergy Asthma Immunol 2004;92:659-662.

32. Christodoulopoulos P, Tulic MK, Kontolemos M, Hamid Q. Immunopatholgy of allergic airway inflammation. In Adkinson FN Jr, Yunginger JW, Busse WW, Bochner BS, Holgate ST, Simons FER, editors. Allergy, Principles and Practice.6th ed. Philadelphia: Mosby; 2003.p.501-514.

33. Shi HZ, Deng JM, Xu H. Effect of inhaled interleukin-4 on airway hyperreactivity in asthmatics. Am Rev Respir Dis 1998;157:1818.

34. Faden H, Kaul TN, Ogra PL. Activation of oxidative and arachidonic acid metabolism in neutrophils by respiratory syncytial virus antibody complexes: possible role in disease. J Infect Dis 1983;148:110-116.

35. Jaovisidha P, Peeples ME, Brees AA, Carpenter LR, Moy JN. Respiratory syncytial virus stimulates neutrophil degranulationand chemokine release. J Immunol 1999;163:2816-2820.

36. Basyigit I, Yildiz F, Ozkara SK, Yildirim E, Boyaci H, Ilgazli A. The effect of clarithromycin on inflammatory markers in chronic obstructive pulmonary disease: Preliminary data. Ann Pharmacother 2004;38:1400-1405.

37. Abe S, Nakamura H, Inoue S, Takeda H, Saito H, Kato S, Mukaida N, Matsushima K, Tomoike H. Interleukin-8 gene repression by clarithromycin is mediated by the activator protein-1 binding site in human bronchial epithelial cells. Am J Respir Cell Mol Biol 2000;22:51-60.

38. Parnham MJ. Immunomodulatory effects of antimicrobials in the therapy of respiratory tract infections. Curr Opin Infect Dis 2005;18:125-131.

39. Tamaoki J, Kadota J, Takizawa H. Am J Med 2004;117:5-11.

40. Roche Y, Gougerot-Pocidalo MA, Fay M, Forest N, Pocidalo JJ. Macrolides and immunity: effects of erythromycin and spiramycin on human mononuclear cell proliferation. J Antimicrob Chemother 1986;17:195-203.

41. Labro MT, el Benna J, Babin-Chevaye C. Comparison of the in vitro effect of several macrolides on the oxidative burst of human neutrophils. J Antimicrob Chemother 1989;24:561-572.

42. Konno S, Adachi M, Asano K, Okamoto K, Takahashi T. Anti-allergic activity of roxithromycin: inhibition of IL-5 production from Mouse T lymphocytes. Life Sci 1993;52:25-30.

43. Thomas LH, Sharland M, Friedland JS. Steroids fail to down-regulate respiratory syncytial viru induce IL-8 secretion in infants. Pediatr Res 2002;52:368-372.

44. Cui CH, Honda K, Saito N, Yamada Y, Sannohe S, Ueki S, Hamada K, Yamaguchi K, Kobayachi Y, Adachi T, Kayaba H, Chihara J. Effect of roxithromycin on eotaxin-primed reactive oxygen species from eosinophils. Int Arch Allergy Immunol 2001;125:38-41.

45. Sigurs N, Gustafsson PM, Bjarnason R, Lundberg F, Schmidt S, Sigurbersson F. Severe respiratory syncytail virus bronchiolitis in infancy and asthma and allergy at age 13. Am J Respir Crit Care Med 2005;171:137-141.

46. Noma T, Aoki K, Hayashi M, Yoshizawa I, Kawano Y. Effect of roxithromycin on T lymphocyte proliferation and cytokine production elicited by mite antigen. International Immunopharmacology 2001;1:201-210.

47. Wennergren G, Kristjansson S. Relationship between respiratory syncytial virus bronchiolitis and future obstructive airway diseases. Eur Respir J 2001;18:1044-1058.

48. Stein RT, Sherill D, Morgan WJ, Holberg CJ, Halonen M, Taussig L, Wright AL. Respiratory syncytial virus in early life and risk of wheeze and allergy by age 13 years. Lancet 1999;354:541-545.

49. Garey KW, Alwani A, Danziger LH, Rubinstein I. Tissue reparative effects of macrolide antibiotics in chronic inflammatory sinopulmonary diseases. Chest 2003;123:261-265.

Table 1. Demographic Characteristic of the Infants

Characteristic	Clarithromycin (n=12)	Plasebo (n=9)	P value
Age (months)*	2 (1-6)	2 (1-7)	>0.05†
Gender			
Male n (%)	8 (66)	4 (45)	>0.05‡
Female n (%)	4 (34)	5 (55)	
Parental smoking n (%)	4 (33)	3 (33)	>0.05‡
Duration of wheezing at admission n (%)			
No wheezing	0	0	
<3 days	8 (66)	6 (66)	>0.05‡
3-6 days	4 (34)	3 (34)	
>6 days	0	0	
Severity of disease n (%)			
Mild			
Moderate	2 (16)	2 (22)	
Severe	9 (72)	6 (66)	>0.05‡
	1 (12)	1 (11)	
Supplemental oxygen n (%)			
No oxygen	2 (16)	2 (22)	>0.05‡
Oxygen	10 (84)	7 (78)	
Skin test positivity %	0	0	>0.05‡
Eosinophil count *	185 (20-544)	150 (30-250)	>0.05†
IgE (ku/L)*	12.5 (1-94)	15 (1-30)	>0.05†
Family history of atopy n (%)	0	0	>0.05‡

* Median (Interquartile range)
† Mann Whitney U test
‡ Chi-square test

Table 2.

Variable	Clarithromycin n=12	Placebo n=9	p value
LOS (hr) *	51 (48-68)	88 (72-100)	<0.05†
Duration of supplemental oxygen need (hr) *	31 (28-42)	72 (52-80)	<0.05†
Duration of supplemental intravenous fluid (hr) *	26 (22-36)	56 (46-66)	<0.05†
Duration beta agonist therapy (day)	5 (4-7)	7 (5-7)	<0.05†
Readmission to the hospital within six months after discharge n (%)	1 (8.3)	4 (44.4)	<0.05‡

* Median (Interquartile range)
† Mann Whitney U test
‡ Chi-square test

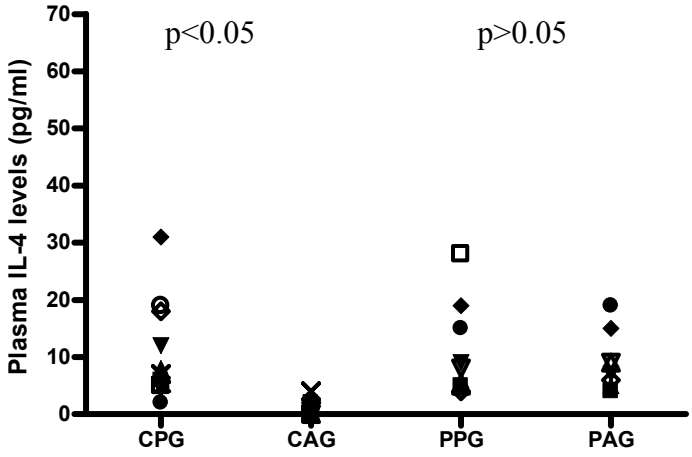
p<0.05
p>0.05
Plasma IL-4 levels (pg/ml)
70
60
50
40
30
20
10
0
CPG
CAG
PPG
PAG

Figure 1.

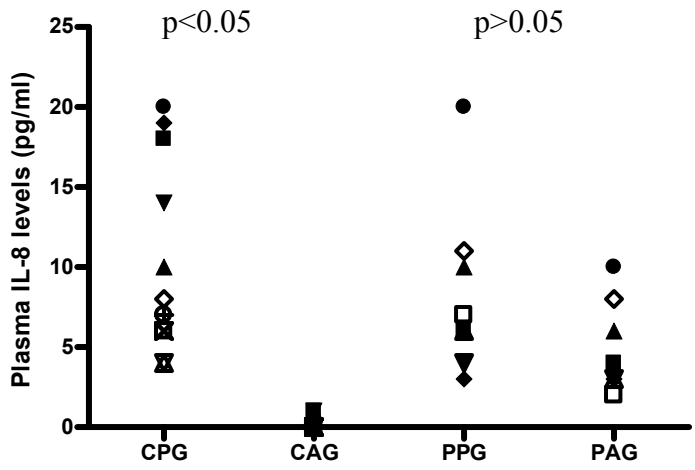

Figure 2.

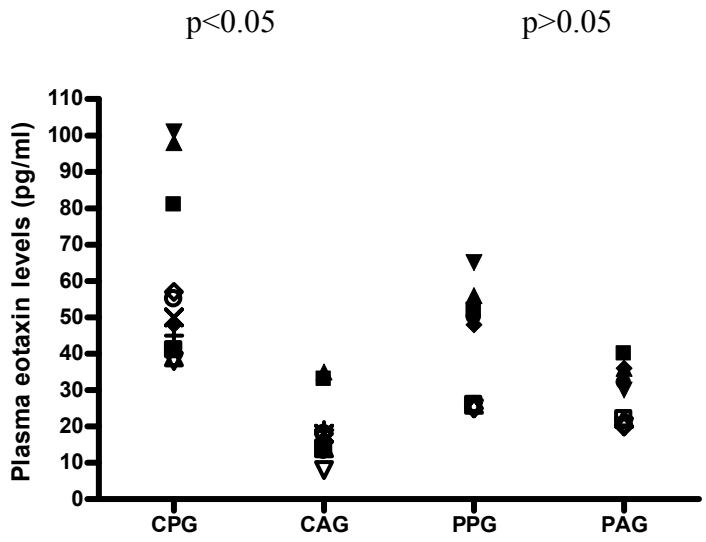
p<0.05
p>0.05
Plasma eotaxin levels (pg/ml)
110
100
90
80
70
60
50
40
30
20
10
0
CPG
CAG
PPG
PAG

Figure 3.

Figure Legends

Figure 1. Plasma IL-4 levels (pg/ml)

CPG : Clarithromycin Pretreatment Group

CAG : Clarithromycin After Treatment Group

PPG : Placebo Pretreatment Group

PAG : Placebo After Treatment Group

Figure 2. Plasma IL-8 levels (pg/ml)

CPG : Clarithromycin Pretreatment Group

CAG : Clarithromycin After Treatment Group

PPG : Placebo Pretreatment Group

PAG : Placebo After Treatment Group

Figure 3. Plasma eotaxin levels (pg/ml)

CPG : Clarithromycin Pretreatment Group

CAG : Clarithromycin After Treatment Group

PPG : Placebo Pretreatment Group

PAG : Placebo After Treatment Group