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Atopic Characteristics of Wheezing Children and Responses to Prednisolone

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Summary. We wanted to test the hypothesis that the efficacy of systemic corticosteroid is associated with atopic characteristics in wheezing children. A randomized controlled trial comparing oral prednisolone (2 mg/kg/day in 3 divided doses for 3 days) with placebo in hospitalized wheezing children (n = 266, median 1.6 years, range 3 months to 15.2 years) was conducted. In this post-hoc analysis, we assessed the link between the efficacy of prednisolone and several atopic characteristics, such as atopy, aeroallergen sensitization, total IgE level, number of sensitizations, eczema, atopic eczema, blood or nasal eosinophils, exhaled nitric oxide, positive modified asthma predictive index/asthma, inhaled corticosteroid medication and parental asthma/allergy. Virology was studied comprehensively. Our primary endpoint was the time until ready for discharge, and the most important secondary endpoint was the occurrence of relapses during the following 2 months. For statistics, we used interaction analyses in uni- and multivariate regression models. Overall, prednisolone did not decrease any of our predefined clinical endpoints. Neither was the efficacy of prednisolone associated with atopy. However, prednisolone significantly decreased the time until ready for discharge in children with positive modified asthma predictive index/asthma, inhaled corticosteroids, or rhinovirus infection and/or in children without azithromycin treatment. Prednisolone significantly decreased relapses in children with eczema, nasal eosinophilia and rhinovirus infection. The multiple clinical, inflammatory and viral markers associating with the efficacy of prednisolone should be confirmed in prospective trials. It is important that corticosteroid intervention trials have strict design for these potentially confounding factors. **Pediatr Pulmonol. 2007; 42:1125–1133.** © 2007 Wiley-Liss, Inc.

Key words: prednisolone; child; wheezing; asthma; atopy; eosinophils; exhaled nitric oxide; rhinovirus; virus; macrolides.

INTRODUCTION

Acute wheezing affects 30–50% of children before school age.[1,2] One third of them suffer from recurrent wheezing. The main risk factors for recurrent wheezing include atopy, eczema, male gender, older siblings, maternal smoking and parental history of atopy or asthma.[1–3] Studies how viral etiology of early wheezing may contribute to later development of asthma have focused almost exclusively on respiratory syncytial virus (RSV), but recent studies suggest that rhinovirus may contribute as well.[3–5]

It would be important to identify children at high risk for recurrent wheezing and provide them with effective treatment.[5] Only a limited number of factors predicting a favorable response to systemic corticosteroids in wheezing children have been identified. Previous studies have found that early episodes of RSV-induced wheezing do not respond to systemic corticosteroids at standard doses[6,7], but rhinovirus detection was not included in these studies. In two studies, non-viral factors, family history of atopy or

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systemic eosinophil priming were not associated with efficacy of systemic corticosteroids in young wheezing children.[6,8] We have previously reported provocative findings on the efficacy of prednisolone in virus-positive cases of early wheezing (<3 episodes) and recurrent wheezing (≥3 episodes).[9,10] Prednisolone consistently decreased relapses in children affected by respiratory picornavirus infection.

In our previous reports, statistical power to detect efficacy in relation to other important risk factors, such as atopy or its correlates, was lost when the populations of interest were narrowed down to relatively small sample sizes (i.e., n = 59 and n = 78) according to specific viral etiology and/or number of previous wheezing episodes.[9,10] Here, we report our full data (n = 266) used to obtain maximum power to assess the link between the efficacy of prednisolone and several atopic characteristics of the patients as exploratory endpoints. We wanted to test the hypothesis that the efficacy of prednisolone is associated with atopy and its correlates. We also recently updated our viral detection methods adding polymerase chain reaction tests up to 16 different viruses to conventional diagnostic tests, and our total detection rate is now 93% in this study population.[11]

METHODS

Subjects

Children aged from 3 months to 16 years admitted to the Department of Pediatrics of Turku University Hospital (9/2000–5/2002) for acute wheezing were considered for the VINKU-study (see Fig. 1). The methods have been described in detail earlier.[9–12] Predefined exclusion criteria included systemic corticosteroid treatment in the preceding 4 weeks, any chronic disease (other than asthma, allergy, or eczema), severe wheezing necessitating intensive care unit treatment or previous participation in this study. The study protocol was approved by the Ethics Committee of the Turku University Hospital.

ABBREVIATIONS

RSV	respiratory syncytial virus
PCR	polymerase chain reaction
API	asthma predictive index
PIV	parainfluenza virus
hMPV	human metapneumovirus
IgE	immunoglobulin E
ICS	inhaled corticosteroid
ER	emergency room
RSS	respiratory symptom score
RR	risk ratio
OR	odds ratio
SD	standard deviation
CI	confidence interval
RV	rhinovirus

Definitions

Atopy was defied as positive IgE antibodies for any of the common allergens (cut-off level 0.35 kU/L for codfish, cow's milk, egg, peanut, soybean, wheat, cat, dog, horse, birch, mugwort, timothy, *Cladosporium herbarum*, and *Dermatophagoides pteronyssinus*; fluoro-enzyme immunoassay, CAP FEIA, Phadiatop Combi®, Phadia, Uppsala, Sweden). Aeroallergen sensitization was defined as positive IgE antibodies for any of the latter 8 allergens, and perennial aeroallergen sensitivity was defined as sensitization to dog, cat or *Dermatophagoides pteronyssinus*. In children aged ≥6 years, the diagnosis of asthma was based on criteria suggested by the guidelines of the National Heart, Lung, and Blood Institute, USA.[13,14] Briefly, children needed to have a history of episodic symptoms of airflow obstruction, and the airflow obstruction had been documented as at least partially reversible (increase in forced expiratory flow in 1 sec ≥12% of predicted after using a short-acting inhaled $beta_2$-agonist). In children aged ≤5 years, instead of asthma diagnosis, we used slightly modified asthma predictive index (API) in agreement with the guidelines, that is, ≥4 wheezing episodes within the past year, of which at least one confirmed by a physician and ≥1 major risk factors (atopic dermatitis or parental asthma) and ≥2 minor risk factors (allergic rhinitis, wheezing apart from colds or blood eosinophilia $\geq 0.40 \times 10^9$/L) originally reported by Castro-Rodriguez et al.[14,15]

Study Design

The children were randomly assigned in a double-blind fashion to receive either oral prednisolone (first dose 2 mg/kg, then 2 mg/kg/day in 3 divided doses for 3 days, maximum dose 60 mg/day, Prednisolon® 5 mg tablets, Leiras, Finland) or placebo after informed consent had been obtained. All patients received nebulized salbutamol according to the study protocol in the hospital (26 patients also received nebulized racemic epinephrine on demand).[9] After discharge, $beta_2$-agonists were used on demand.

The patients were examined twice daily during their hospital stay by the study physicians (T.J. and P.L.). After the child's discharge, the guardian recorded the child's symptoms in a diary for 2 weeks after which the child was seen at the clinic. After 2 months, the parents were contacted by telephone to record any relapses necessitating a visit to a physician or hospitalization.

Outcome Measures

A pre-specified primary endpoint was the time until ready for discharge, which was defined as a duration of respiratory symptoms score >3 during hospital stay.[9,10] Prespecified secondary endpoints were oxygen saturation and exhaled nitric oxide during hospital stay, wheeze and

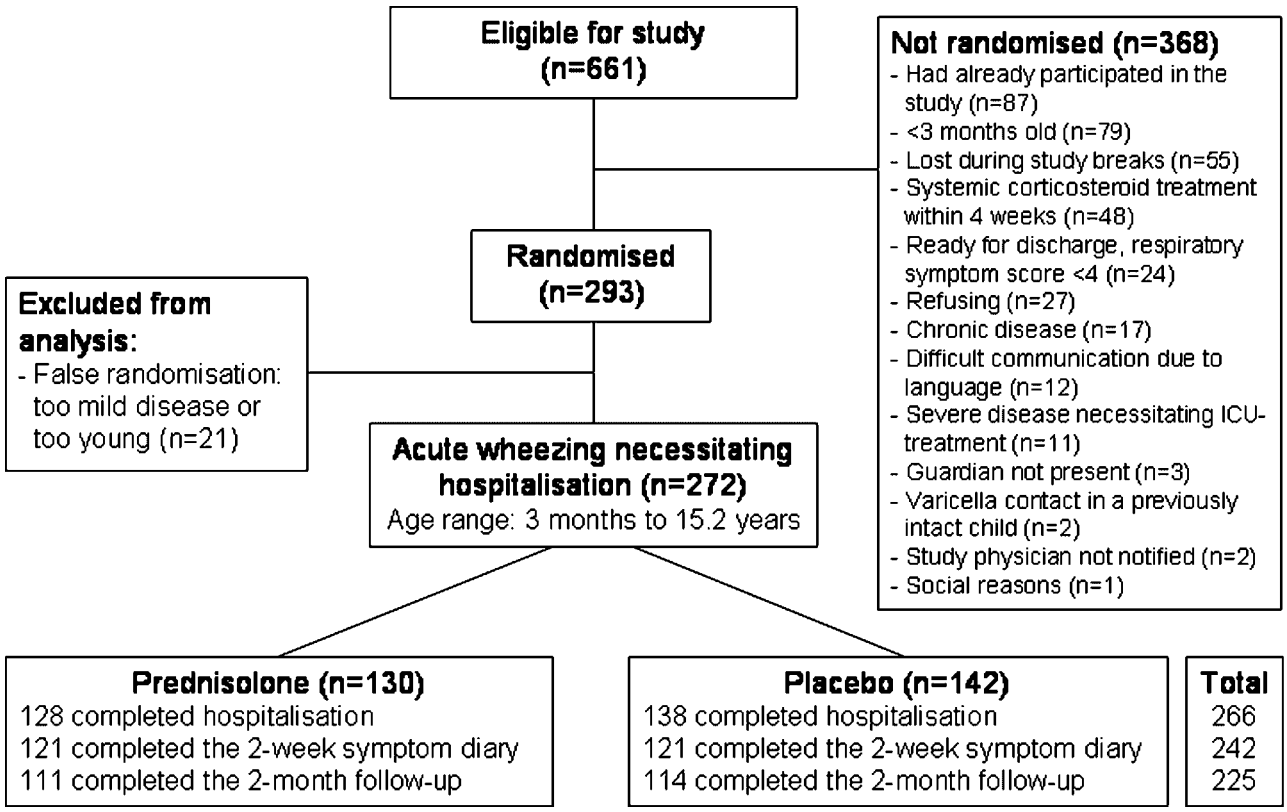

Fig. 1. Study flow chart.

cough during 2 weeks after discharge, impulse oscillometry with bronchodilation 2 weeks after discharge, blood eosinophil counts at discharge and 2 weeks later and readmission to the out-patient clinic or hospital for recurrent wheezing within a 2-month period after discharge.

Sample Collection and Analysis

On admission, a nasopharyngeal aspirate for viral diagnostics was drawn in a standardized procedure.[16] Blood samples were collected on the patient's admission, at discharge from hospital and 2–3 weeks after discharge.

Virus culture was done for adenovirus, influenza A and B viruses, parainfluenza virus (PIV) types 1–3, RSV, enteroviruses, rhinovirus and human metapneumovirus (hMPV).[11,16] Viral antigens were detected for adenovirus, influenza A and B viruses, PIV 1–3, and RSV. Levels of IgG antibodies specific for adenovirus, enteroviruses, influenza A and B viruses, parainfluenza virus types 1/3, RSV were analyzed in paired serum samples, in addition to IgM antibodies for enteroviruses. PCR was used for the detection of entero- and rhinoviruses, RSV, coronaviruses (229E, OC43, NL63, and HKU1), hMPV, human bocavirus, influenza A, and B viruses, adenovirus and PIV 1–4.

Exhaled nitric oxide was successfully collected in the tidal breathing offline collection procedure from 162 children.[9] Triple measurements of respiratory resistance at 5, 20, and 35 Hz were made in 65 children 2 weeks after discharge.[10]

Statistics

Primary power calculation served our original aim to study efficacy in relation to viral infections.[9] The normality of data distribution was tested using the Kolmogorov–Smirnov test. The *t*-test, Mann Whitney *U* test and Chi square test were used when appropriate. The efficacy of prednisolone was analyzed using multivariate regression analysis (generalized linear model) or multivariate logistic regression analysis with binomial distribution and logit-link. Interaction analysis between prednisolone and different variables was first done with one interaction term with respective subterms in the model (here called univariate model) and then repeated with adjustments to age, gender, atopy, positive modified API/asthma, inhaled corticosteroid (ICS) on admission, rhinovirus infection and antibiotic treatment (which were not included as interaction terms) in a backward stepwise model (here called multivariate model). Only significant adjustments as shown were kept in the model. Furthermore,

to estimate drug effect in certain patient groups, contrast estimates were formulated. The effects of prednisolone on oxygen saturation during the first 48 hr of hospital stay were tested at 12 hr intervals using the repeated measurements analysis of variance. Statistical significance was established at a *P*-value lower than 0.05. The statistical analyses were carried out using SAS/STAT(r) software, Version 9.1.3 SP4 of the SAS System for Windows, SAS Institute Inc. (Cary, NC).

RESULTS

Characteristics of the Children

The recruitment, randomization and follow-up of study subjects are shown in Figure 1. The median age of the 266 children completing hospitalization was 1.6 years (interquartile range, 11 months to 2.5 years). Of these, 115/265 (43%) children had a history of eczema, 93/260 (36%) children had specific IgE sensitization, 49/260 (19%) children had aeroallergen sensitization and 37/264 (14%) children had positive modified API/asthma. Only 25/265 (9%) children were using ICS at study entry. Twenty-two (59%) children with positive modified API/asthma had eczema, 18 (49%) of them were sensitized using the cut-off level of >0.35 kU/L and 12 (32%) of them were sensitized using the cut-off level of >0.70 kU/L.

No significant differences were found in baseline values between the treatment groups except in the number of children receiving miscellaneus/dual antibiotic treatments ($P = 0.027$, Tables 1 and 2). No other macrolides than azithromycin were used.

Efficacy of Prednisolone

Overall, prednisolone showed no effect on our primary endpoint, time until ready for discharge (medians [interquartile range], prednisolone vs. placebo, 18 [6,43] vs. 30 hr [18,50], $P = 0.51$, Table 3). The study drug, however, significantly decreased the time until ready for discharge in children with positive API/asthma (API/asthma+, risk ratio [RR] 1.99, 95% CI 1.18–3.35, $P = 0.0094$; API/asthma-, RR 1.11, 95% CI 0.90–1.37, $P = 0.33$; adjusted to age, antibiotic treatment and rhinovirus infection), and in children with ICS at study entry (ICS+, RR 2.23, 95% CI 1.12–4.41, $P = 0.022$; ICS-, RR 1.02, 95% CI 0.82–1.26, $P = 0.89$; no adjustments). As for viral etiology, prednisolone decreased the time until ready for discharge in RSV negative children (RSV-, RR 1.38, 95% CI 1.09–1.75, $P = 0.0078$; RSV+, RR 0.79, 95% CI 0.54–1.15, $P = 0.21$; no adjustments), and in rhinovirus positive children (rhinovirus+, RR 1.69, 95% CI 1.14–2.50, $P = 0.0092$; rhinovirus−, RR 1.07, 95% CI 0.85–1.33, $P = 0.57$; adjusted to age and antibiotic treatment). Children without antibiotic treatments also benefited from prednisolone (antibiotics−, RR 1.56, 95% CI 1.19–2.06, $P = 0.0015$; antibiotics+, RR 0.95, 95% CI 0.73–1.24, $P = 0.70$; adjusted to age and rhinovirus infection), especially those without azithromycin (azithromycin−, RR 1.30, 95% CI 1.06–1.60, $P = 0.011$; azithromycin+, RR 0.67, 95% CI 0.37–1.19, $P = 0.17$; adjusted to antibiotic treatment, age and rhinovirus infection).

No overall difference in the effect on the occurrence of relapses during the 2 months after discharge was found

TABLE 1— Patient Characteristics in Prednisolone and Placebo Groups

Factor	Prednisolone (n = 128)[1]	Placebo (n = 138)	*P*
Age, years	1.4 (0.8, 2.4)	1.6 (1.0, 2.9)	0.14
Male, No.	86 (67%)	92 (67%)	0.93
Atopy (specific IgE > 0.35 Ku/L), No.	43/125 (34%)	50/135 (37%)	0.66
Atopy (specific IgE > 0.70 kU/L), No.	33/125 (26%)	36/135 (27%)	0.96
Aeroallergen sensitization (IgE > 0.35 kU/L), No.	24/125 (19%)	25/135 (19%)	0.89
Aeroallergen sensitization (IgE > 0.70 kU/L), No.	20/125 (16%)	40/127 (16%)	0.95
Perennial aeroallergen sensitization (IgE > 0.35 kU/L), No.	22/125 (18%)	19/135 (14%)	0.44
Perennial aeroallergen sensitization (IgE > 0.70 kU/L), No.	16/125 (13%)	17/135 (13%)	0.96
Eczema, No.	50 (39%)	65/137 (47%)	0.17
Atopic eczema, No.	20/127 (16%)	29/137 (21%)	0.26
Positive API/asthma, No.	17/126 (13%)	20 (14%)	0.82
Number of wheezy episodes, No.	1 (1, 2)	1 (1, 3)	0.72
ICS at study entry, No.	13/127 (10%)	12 (9%)	0.67
ICS started/continued after discharge, No.	24/124 (19%)	28/131 (21%)	0.69
ICS started/continued 2 weeks after discharge, No.	37/124 (30%)	42/131 (32%)	0.70
Parental asthma, No.	25/126 (20%)	31/135 (23%)	0.54
Parental allergy, No.	78/126 (62%)	74/135 (55%)	0.25
Previous lower respiratory symptoms, days	3 (1, 7)	3 (1, 4)	0.30

Values are medians (interquartile range), unless otherwise noted. Analysis using the Mann–Whitney *U* test or Chi-square test. IgE, Immunoglubulin E; API, asthma predictive index; ICS, inhaled corticosteroid.
[1]Subjects completing hospitalization.

TABLE 2— Clinical and Laboratory Data at Study Entry in Prednisolone and Placebo Groups

Factor	Prednisolone (n = 128)[1]	Placebo (n = 138)	*P*
RSS, points[2]	5.3 (6.5)	6.5 (1.6)	0.89
O_2-saturation, %	95.9 (2.2)	95.6 (2.5)	0.30
Acute otitis media, No.	61 (48%)	56/137 (41%)	0.27
Blood eosinophils, $\times 10^9$/L	0.20 (0.10, 0.40)	0.20 (0.10, 0.50)	0.40
Blood eosinophils $\geq 0.40 \times 10^9$/L	57/124 (46%)	66/133 (50%)	0.55
Nasal eosinophils, 4-point scale 0–3[3]	0 (0, 1)	0 (0, 1)	0.86
Exhaled nitric oxide, ppb	6.2 (4.2, 8.2)	5.9 (3.8, 9.5)	0.87
Exhaled nitric oxide >10 ppb, No.	12/77 (16%)	17/85 (20%)	0.46
Any virus, No.	120 (94%)	128 (93%)	0.75
Rhinovirus, No.	34 (27%)	30 (22%)	0.36
Enteroviruses, No.	29/120 (24%)	39/129 (30%)	0.28
Nontypable rhino- or enterovirus, No.	14 (11%)	26 (19%)	0.072
Respiratory syncytial virus, No.	38 (30%)	35 (25%)	0.43
Multiple viruses, No.	38 (30%)	47 (34%)	0.45
Albuterol at ER, mg/kg	0.15 (0.11, 0.39)	0.15 (0.10, 0.40)	0.72
Antibiotic treatment, No.[4]	68/127 (54%)	68 (49%)	0.45
Penicillin or amoxicillin, No.	25/127 (20%)	33/137 (24%)	0.39
Azithromycin, No.[5]	13/127 (10%)	16/137 (12%)	0.71
Other antibiotic or dual, No.[6]	30/127 (24%)	18/137 (13%)	0.027

Values are means (SD) or medians (interquartile range), unless otherwise noted. Analysis using the *t*-test, Mann–Whitney *U* test or Chi-square test. ER, emergency room (before entry to the study).
[1]Subjects completing hospitalization.
[2]Respiratory symptom score (RSS) were assessed on a scale from 0 (none) to 12 (severe).
[3]Prednisolone group, n = 114; placebo group, n = 125.
[4]Name of the antibiotic not available from two cases.
[5]No other macrolides were used.
[6]In the prednisolone group: single-dose ceftriaxone i.m. 22 (73%), amoxicillin-clavunate p.o. 4 (13%), cephalosporin p.o. 3 (10%), and nitrofurantoin p.o. 1 (3%). In the placebo group: single-dose ceftriaxone i.m. 13 (72%), amoxicillin-clavunate p.o. 3 (17%), sulfa-trimethoprim p.o. 1 (6%) and sulfa-trimethoprim p.o. + single dose ceftriaxone i.m. 1 (6%).

between the prednisolone and placebo groups (prednisolone vs. placebo, 23/111 [21%] vs. 29/114 [25%], $P = 0.40$, Table 3). Most strikingly, prednisolone decreased relapses in rhinovirus positive children (rhinovirus+, OR 13.00, 95% CI 2.33–72.46, $P = 0.0034$; rhinovirus−, OR 0.70, 95% CI 0.44–1.77, $P = 0.13$; adjusted to age), but also in all respiratory picornavirus positive children (picornavirus+, OR 2.92, 95% CI 1.21–7.04, $P = 0.017$; picornavirus−, OR 0.86, 95% CI 0.42–1.39, $P = 0.69$; adjusted to age). Prednisolone also decreased relapses in children with a history of eczema (eczema+, OR 3.22, 95% CI 1.16–8.94, $P = 0.025$; eczema−, OR 0.64, 95% CI 0.25 to 1.61, $P = 0.34$; adjusted to age) and in children with increased nasal eosinophil counts at study entry (one unit increase in nasal eosinophil count on 4-point scale, OR 2.17, 95% CI 1.25–3.78, $P = 0.0062$; one unit decrease in nasal eosinophil count, OR 0.78, 95% CI 0.37–1.60, $P = 0.49$; adjusted to age).

In the other endpoints, overall blood eosinophil count at discharge was strongly affected by prednisolone treatment ($P < 0.0001$). No other significant effects were found in any of the studied endpoints between prednisolone and placebo (data not shown). Prednisolone was well tolerated.

DISCUSSION

Surprisingly, allergic sensitization was rarely associated with a positive response to prednisolone treatment in wheezing children although 36% of them were sensitized. None of the studied sensitization groups or cut-off levels could significantly be linked to the response to prednisolone treatment. Of the many atopic correlates studied, history of eczema, nasal eosinophils and positive modified API/asthma were most likely to predict a beneficial response to this agent. Rhinovirus infection was, again in this full analysis, clearly associated with the usefulness of the study drug. Interestingly, azithromycin treatment appeared to mask the effects of prednisolone.

The strengths of the study include comprehensive viral diagnostics, detailed clinical follow-up, careful characterization of atopic status and relatively large sample size. Weaknesses of the analysis include multiple comparisons (risk for type 1 error, but meaningful associations argue against it), loss of randomization due to post-hoc analysis

TABLE 3—Factors Associating With the Effects of Prednisolone for the Time Until Ready for Discharge and Out-Patient Visits Due to Wheezing During 2 Months After Discharge—Results of Regression Analysis

	Time until ready for discharge					Out-patient visits due to wheezing				
	Univariate		Multivariate			Univariate		Multivariate		
Interaction term	n	*P*	n	*P*	Adjustments[1]	n	*P*	n	*P*	Adjustments[1]
None	266	0.51	262	0.10	Age, ab, rv	225	0.40	225	0.26	Age
Age, years	266	0.42	262	0.77	ab, rv	225	0.70	225	0.70	—
Male gender	266	0.14	262	0.16	Age, ab, rv	225	0.35	225	0.43	Age
Atopy (specific IgE > 0.35 kU/L)	260	0.46	260	0.55	Age, ab, rv	223	0.43	225	0.28	Age
Atopy (specific IgE > 0.70 kU/L)	260	0.054	260	0.090	Age, ab, rv	223	0.51	223	0.40	Age
Aeroallergen sensitivity (>0.35 kU/L)	260	0.41	260	0.72	Age, ab, rv	223	0.26	223	0.32	Age
Aeroallergen sensitivity (>0.70 kU/L)	260	0.35	260	0.67	Age, ab, rv	223	0.26	223	0.39	Age
Perennial aeroallergen sensitivity (>0.35 kU/L)	260	0.33	260	0.84	Age, ab, rv	223	0.75	223	0.90	Age
Perennial aeroallergen sensitivity (>0.70 kU/L)	260	0.21	260	0.49	Age, ab, rv	223	0.98	223	0.88	Age
Eczema	265	0.47	261	0.57	Age, ab, rv	225	0.051	225	0.022	Age
Atopic eczema	264	0.35	261	0.88	Age, ab, rv	225	0.40	225	0.23	Age
Blood eosinophil count, $\times 10^9$/L	257	0.89	256	0.87	Age, ab, rv	218	0.48	218	0.31	Age
Nasal eosinophil count, 4-point scale	239	0.67	236	0.52	Age, ab, rv	203	0.021	203	0.023	Age
Parental allergy	261	0.81	258	1.00	Age, ab, rv	223	0.57	223	0.59	Age
Number of wheezy episodes	258	0.96	255	0.62	Age, ab, rv	221	0.70	221	0.42	Age
Positive API/asthma	264	0.025	263	0.040	Age, ab, rv	225	0.98	225	0.97	Age
ICS on arrival	265	0.032	264	0.053	Age, ab, rv	225	0.50	225	0.24	Age
RSS on arrival, points[2]	266	0.97	262	0.99	Age, ab, rv	225	0.49	225	0.51	Age
Parental asthma	261	0.81	258	0.72	Age, ab, rv	223	0.093	223	0.15	Age
Any virus	266	0.067	265	0.35	Age, ab, rv	225	0.17	225	0.090	Age
RSV	266	0.014	265	0.064	Age, ab, rv	225	0.68	225	0.61	Age
Respiratory picornavirus	266	0.093	265	0.10	Age, ab, API/asthma	225	0.011	225	0.010	Age
Rhinovirus	266	0.024	265	0.047	Age, ab	225	0.0074	225	0.0043	Age
Enterovirus	249	0.16	248	0.50	Age, ab, rv	220	0.20	220	0.34	Age
Antibiotic treatment	265	0.0034	265	0.011	Age, rv	225	0.63	225	0.63	Age
Penicillin or amoxicillin	264	0.87	264	0.89	ab, age, rv	224	0.55	224	0.51	Age
Azithromycin[3]	264	0.17	264	0.032	ab, age, rv	224	0.97	224	0.79	Age
Other antibiotic or dual[4]	264	0.19	264	0.11	ab, age, rv	224	0.16	224	0.14	Age

Interaction analysis between prednisolone and the variables shown without adjustments (univariate) and adjusted with age, gender, atopy, asthma, ICS on arrival, rhinovirus infection and antibiotic treatment in a backward stepwise model (multivariate). Only significant ($P < 0.05$) adjustments were kept in a final model. Ab, antibiot; rv, rhinovirus; IgE, Immunoglobulin E; API, modified asthma predictive index; ICS, inhaled corticosteroid; RSS, respiratory symptom score; RSV, respiratory syncytial virus; OR, odds ratio; RR, relative risk.

[1]In addition to interaction terms.

[2]Respiratory symptom score (RSS) were assessed on a scale from 0 (none) to 12 (severe) at study entry.

[3]No other macrolides were used.

[4]No significant difference with the cephalosporin group only.

(only option, however, if full viral diagnostics is used), power calculation designed for viral etiology, wide age distribution, predominance of young children and inclusion of all wheezy phenotypes in one group. Age and number of wheezy episodes, however, were not associated with the response to prednisolone treatment.

Only a limited number of studies have focused on non-viral factors predicting a favorable response to systemic corticosteroids in wheezing children. In two studies, family history of atopy or systemic eosinophil priming were not associated with efficacy of systemic corticosteroids in young wheezing children.[6,8] The limitations of the former include a rather small sample size and of the latter a delay of up to 12 months between measurement of eosinophil priming and prednisolone treatment and parents' responsibility to initiate the treatment. The results of the studies of blood or mucosal eosinophil counts as predictors of response to corticosteroids have been variable but seem to favor their feasibility.[17,18] In a study of wheezing infants, intermittent ICS therapy was not associated with any beneficial response either in patients with or without eczema.[19] Some of the causes for the ineffectiveness of the ICS treatment may have been a 3-day delay in starting treatment, low non-systemic dosage or out-patient setting. In another recent study, young children with a positive API benefited from ICS during a 2-year treatment but it did not have any disease-modifying effect after its discontinuation.[20]

Why atopic children did not respond to prednisolone in our study? First, most of our study children were young and may not have had enough time to develop significant sensitization. This may also explain why children with eczema responded to prednisolone, since non-atopic eczema predicts sensitization in wheezing children.[21] Second, the prevalence of aero- or perennial allergies were rather low (16–19%). These have been the most important allergy risk groups for later development of asthma.[1,2] Third, exposure to allergens may be more important than the type of allergy. Higher cut-off level for specific IgE and nasal eosinophilia discriminated better steroid responders, which may suggest that those with stronger/active allergies or heavier exposure to allergens are more likely to respond to systemic corticosteroids.

The asthma predictive index has been developed for young children, since no single genetic or biochemical marker, or epidemiologic risk factor has been sufficiently sensitive to predict the development of asthma.[15] Interestingly, our slightly modified API predicted apparently well the response to prednisolone in terms of shortened time until ready for discharge. Our negative result on the association between exhaled nitric oxide levels and the efficacy of prednisolone may have been due to low levels of nitric oxide measured, a low number children with positive modified API/asthma and insufficient statistical power.[22] Currently, no standardized tidal breathing offline method is available for young children, and practical issues remain to be solved.

No other picornavirus specific efficacy studies in wheezing children are yet available apart from our reports.[9,10,12] The experimental infections in adults with mild asthma are not comparable to natural viral infections in children, especially to moderate-to-severe cases necessitating hospitalization.[23–25] We speculate that an early asthma-like inflammation could explain the beneficial effect of prednisolone in the rhinovirus group. The children susceptible to rhinovirus induced moderate-to-severe wheezing may have an underlying immunologic anomaly predisposing them to more severe outcome as recently extensively reviewed.[26,27] The predisposing factors are not precisely known, but may be related intrinsic genetic susceptibility (family history of asthma/atopy, cytokine dysregulation, or lung development), environmental factors (properties of the infecting virus/virus strain, passive smoking, or allergen exposure) and age (stage of development of immune system). These factors together may lead to altered antiviral immunity, increased inflammatory response and altered cell signaling pathways. Lower respiratory tract rhinovirus infection may even be the first sign of asthma, as suggested by Kotaniemi-Syrjänen et al.[4] Their preliminary finding is supported by a recent report of Lemanske et al.[3] showing that first-year wheezing illnesses caused by rhinovirus infection are the strongest predictor of subsequent third-year wheezing.

Our study supports the view that azithromycin (the decision of antibiotic use was made by an on-duty physician independent of the study) may have anti-inflammatory effects in viral induced lower respiratory illness. Viral infection was detected in 93% and bacterial seroconversion (to *Streptococcus pneumoniae*, *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, *Hemophilus influenzae*, or *Moraxella catarrhalis*) in 18% of virus positive children.[28] Macrolides have been shown to inhibit inflammatory cell mediator release and survival as well as to have a direct relaxant effect on airway smooth muscle.[29–31]

CONCLUSIONS

Our post-hoc analysis suggests that clinical, inflammatory and viral markers, such as history of eczema, positive modified API/asthma, nasal eosinophilia and rhinovirus infection, are likely to be associated with a favorable response to systemic corticosteroids in wheezing children. Surprisingly, allergic sensitization as such was rarely associated with a positive response to prednisolone treatment. These findings among others need to be confirmed in prospective trials which look for relationships of particular characteristics to response. It is important that corticosteroid intervention trials have strict

design for these multiple and potentially confounding factors.

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