Cough frequency in children with mild asthma correlates with sputum neutrophil count

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Abstract

Objectives: To (1) measure cough frequency in children with stable asthma using a validated monitoring device, and (2) assess the correlation between cough frequency with the degree and type of airway inflammation.

Methods: Thirty-six children with a median age of 11.5 years (IQR: 9 – 14) with stable asthma were recruited. They underwent spirometric testing, exhaled nitric oxide (eNO) measurement, sputum induction for differential cell count, and ambulatory cough monitoring for 24 hours. Coughing episodes were counted both as individual spikes and as clusters.

Results: All children had mild intermittent asthma and their median forced expiratory volume in 1 second (FEV₁) and eNO were 83.3% (IQR 81.1–97.6) and 56.1 ppb (IQR 37.4–105) respectively. The median number of cough episodes per day was 25.5 (IQR 16–42.8). Sputum induction was successful in 69% of the subjects and cough frequency was found to have a significant positive correlation with sputum neutrophil count \((r=0.833, p=0.0001)\).

Conclusions: Children with stable mild asthma have increased cough frequency that might be driven by a neutrophilic inflammatory pathway.

Introduction

Cough is a very common symptom of respiratory diseases. It is audible, can cause sleep disturbance and may also represent serious underlying disorders. It is therefore
unsurprising that parents are frequently anxious about their children’s cough and often seek medical advice and remedies.[1] It has been reported that lack of sleep and choking were the two most common concerns expressed by parents in relation to their children’s cough.[1] Children with asthma can present with cough as the only symptom and in some cases, cough has been shown to be a dominant feature of asthma exacerbations.[2] In a study from South Africa, only 2.2% of children with classical asthma wheeze but do not cough.[3] In an unselected group of children hospitalized for an asthma exacerbation, almost 50% of parents reported that cough was usually or always present with an asthma exacerbation.[2] When cough was objectively measured, nocturnal cough was present in all children recovering from an acute exacerbation and in 39% of children with stable asthma.[4,5] In a study involving children with severe asthma, Li et al.[6], were able to find increased cough frequency in their subjects compared to normal controls. Whether cough frequency in children with milder form of asthma is also increased and whether the frequency of cough correlates with underlying airway inflammation is unclear.

There are several difficulties in quantifying clinically relevant cough. Subjective recording of cough by means of diary cards and patients’ report of cough frequency can be very variable and its reliability has been questioned.[7-11] Conventional tape recorders have been used to quantify cough objectively.[8] However, tape recorders are bulky and the lack of portability precludes their use during normal daily activities. In addition, they rely only on a single audio signal. Recently the use of an ambulatory cough monitor has been validated in both adults and children.[12-14] The device is highly acceptable to children and no adverse effects during recording have been reported.[6,13-15]

In this study, we aimed to (1) measure cough frequency in children with stable asthma using a validated objective cough monitoring device, and (2) assess the correlation between measured cough frequency with the degree and type of airway inflammation.

Methods

Patient selection

All children with stable mild asthma attending the Paediatric Chest Clinic of the Prince of Wales Hospital between December 2003 and October 2004 were recruited. The ages of the children were between 7 and 18 years and all were able to cooperate with the tests. The diagnosis of asthma was made on standard grounds.[16] We defined stable mild asthmatics as those (1) with no disease exacerbation in the preceding 4 weeks necessitating oral corticosteroids or an increased use of inhaled corticosteroids, (2) use of rescue treatment for not more than three times a week, (3) with no clinical indication for change in treatment medication, and (4) < 1 time a week of daytime symptoms and < 2 times a week of nocturnal symptoms. We excluded children who had other concomitant non-asthmatic chronic airway diseases such as bronchiectasis; those who used any prescription or over-the-counter medication that might have affected the course of asthma or its treatment (such as traditional Chinese herbal medicine); and children who were currently involved in any other asthma treatment trial. We obtained approval for the study from the Ethics Committee of the Chinese University of Hong Kong, and the children’s parents or guardians gave written informed consent.
Study design

This was a prospective observational study. A detailed history was taken and thorough physical examination performed on each child. Weight and height were recorded using standardized equipment. The recruited subjects were required to undergo the following assessments as part of their asthma management.

(1) Asthma severity by visual analogue score, a subjective score from 0 to 10 (0 = asymptomatic, 10 = severe disabling asthma) was obtained from the subjects. Similar visual analogue score to assess patients’ daytime and nighttime cough severity (0 = no cough, 10 = severe disabling cough) was also obtained.

(2) Skin prick test to five groups of aeroallergens (house dust mite, cat and dog dander, cockroaches, grass and tree pollens and mixed moulds) was performed using purified allergen extracts. A child was considered atopic if he had at least one skin test result that showed an induration with a diameter of at least 3mm greater than the saline control.

(3) Measurement of exhaled nitric oxide (eNO) using a chemiluminescence analyzer (NOA280i, Sievers Instruments, Boulder, CO, USA), sensitive to NO from 1 ppb to 200 ppm and with a resolution of 1 ppb and an accuracy of ±1 ppb, according to the American Thoracic Society guidelines.[17] The subject was comfortably seated without a nose clip, and inhaled NO-free air from a reservoir and subsequently exhaled against a resistor. The flow rate was set at 50 ml/s. This on-line measurement was taken in triplicate and the average recorded.

(4) Spirometry (Spirolab II, MIR, Italy) using standard technique measuring forced expiratory volume in the first second (FEV₁) and forced vital capacity (FVC). The obtained best of three efforts was compared with local age- and sex-matched reference values.[18]

(5) Sputum induction (SI). Hypertonic saline (4.5% HS) was used and sputum induction carried out using the standard technique.[19,20] The detailed methodology has been described in our previous publication.[6] The sputum sample was processed within 4 hours by an individual who was unaware of the clinical data of the subjects. Specimens containing squamous epithelial cells of less than 50% of the total inflammatory cell number were considered adequate, i.e. successful induction. At least 400 inflammatory cells were counted for each specimen. Eosinophil and neutrophil counts were expressed as percentage of total cell count.

(6) Cough monitoring. The LR 102 cough recorder used in this study is a modification of the ambulatory cough monitoring device (LR 100) which has been validated for use in children.[6,14] The only difference is that the new device is able to record over a 24-hour period. LR 102 is a multiparametric recording device, worn in a waist bag, connected to the chest by three electromyographic (EMG) leads and a microphone. Two signals are recorded, a surface EMG and an audio signal. Cough is defined as before by a combination of rapid phasic bursts in both signals and the detailed processing of the recorded signals has been described in our previous publication.[6] Coughing events were counted both as individual spikes, and as clusters or bouts. We arbitrarily defined each cluster (a cough epoch) as a close succession of coughs (< 2 seconds between individual
coughs) recorded by each trigger of the recorder. The cough data were expressed as total number of cough episodes (individual spike + cough cluster) per unit recording time. The subjects had the monitor put on at 4 pm on the study day and were asked to return at the same time the next day for its removal and would then undergo sputum induction.

**Statistical analysis**

The data is presented as medians with interquartile ranges (IQR). Spearman’s rank correlation coefficient was used to assess the association between the various parameters. SPSS for Windows statistical software (Release 11.0, SPSS Inc., Chicago, Illinois) was used in the analyses. The level of significance was set at 5% in all comparisons, and all statistical testings were two-sided.

**Results**

Thirty-six children (aged 7 – 17 years) with a median age of 11.5 years (IQR: 9 – 14) were recruited in this study. There were 24 boys and 12 girls. All subjects by definition were atopic. Eleven (31%) had sensitivity to more than one allergen, and all of them were sensitive to house dust mite. Five patients were using inhaled corticosteroids and the range of beclomethasone equivalent dosage was between 200mcg and 400mcg, (assuming budesonide is equally potent while fluticasone is twice as potent as beclomethasone). The remaining 31 were using short-acting beta agonists on an as required basis. All subjects were only symptomatic with respiratory tract infections and none complained of interval symptoms of asthma, for example wheeze with weather change. Twenty-four subjects had normal spirometry. Ten subjects had mild obstructive deficit with FEV₁ >70% predicted and the remaining two had severe obstructive deficits, FEV₁ was 46% and 49% predicted. Nonetheless, they all considered their asthma to be mild, and the median asthma severity score was 0.3. In addition, they did not consider cough to be a significant symptom at the time of the study. The median daytime and nighttime cough scores were both reported as 0.2. Seven children had passive tobacco smoke exposure. The baseline characteristics of the subjects are summarized in Table 1.

<table>
<thead>
<tr>
<th>Table 1. Demographic characteristics and investigation results of the subjects</th>
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<td><strong>Median</strong></td>
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<td><strong>Severity of asthma score</strong></td>
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<td><strong>Day Time Cough</strong></td>
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IQR – interquartile range
Cough recording was successful in all children and no adverse events or sleep disturbance were reported. The median number of cough episodes per day was 25.5 (IR 16 - 43). The distribution of cough episodes over the whole recording period is shown in Figure 1. We were not able to measure sleep stage, so we asked all parents to report the time when the patients slept and woke up in the morning. We found that all subjects went to sleep between ten and midnight and woke up between seven and nine in the morning. So we arbitrarily defined night cough as cough occurring between eleven in the evening and seven in the morning and daytime cough as cough occurring at any other time. Median daytime coughs / hour was significantly greater than nighttime coughs [71.5 / hour (IQR 56.50 – 76.25) vs 6.5 / hour (IQR 3.75 – 29.75), p<0.001].

Twenty-five out of 36 children (69.4%) were able to produce an adequate sputum sample on induction. The median eosinophil and neutrophil counts were 1.8% and 8.3% respectively. Using Spearman’s correlation, cough frequency was found to have a significant positive correlation with sputum neutrophil count. The results are summarized in Table 2 and a scatterplot showing the relationship between cough frequency and sputum neutrophils is shown in figure 2.

| Table 2. Correlation between cough and other parameters. |
|----------------------------------------|----------------|----------------|
|                                       | Correlation Coefficient | P value         |
| eNO                                    | -0.185           | 0.279          |
| FEV1                                    | -0.215           | 0.208          |
| FVC                                     | -0.119           | 0.491          |
| % Eosinophil                            | 0.02             | 0.924          |
| % Neutrophil                            | 0.833            | 0.0001*        |
| Severity of asthma score                | -0.077           | 0.679          |
| Self Reported Day Time Cough            | -0.090           | 0.630          |
| Self Reported Night Time Cough          | -0.068           | 0.717          |

Discussion

In this study, our aims were to assess the cough frequency in a group of children with mild asthma and to examine the relationship between measured cough frequency and airway inflammation. We were able to show that despite recruiting at a time of apparent stability, our cohort of mild asthmatics had increased cough frequency compared to that reported for normal Caucasian children.[13] Traditional teaching has led us to believe that children with asthma would cough more than normal subjects only during exacerbations, but not during remission.[21] This is in sharp contrast to findings of our current study where children with stable asthma had increased cough frequency. Munyard et al. used a similar monitoring device on forty-four normal Caucasian children.
aged between 8 – 12 years and found a median cough frequency of 11 episodes per day.[13] Our cohort of mild asthmatics had a median cough frequency of 25.5 episodes over a 24-hour period. This increased cough frequency was recorded at a time when the patients were in remission. Furthermore, they did not consider cough to be much of a troublesome symptom at the time of study as the median cough severity score was only 0.2. Increased cough frequency has also been reported for stable severe asthmatics, where their median cough frequency was found to be 19 episodes per day.[6] In addition, we have found that cough frequency is greater during the day than at night, despite conventional teaching on the importance of nocturnal cough in asthma.

We did not find any correlation between the subjective self reported daytime cough score ($p = 0.630$) and nighttime cough score ($p = 0.717$) with the actual cough frequency recorded. This finding is similar to what has been reported in the literature, questioning the reliability of subjectively reported cough intensity and severity.[1,5,7-11] Although subjective evaluation of patient perception of cough is the most convenient and commonly used tool, it remains an unvalidated, and apparently unreliable measure. In addition, we do not know whether the cough severity scale is linear and how much the reported symptoms are affected by the patient’s mental state.

Persistent airway inflammation in stable asthmatics may explain our finding of increased total cough. A recent study showed that persistent airway inflammation was still evident in asthmatics that were asymptomatic with normal lung function.[22] Ongoing airway inflammation in our group of stable asthmatics is supported by the finding that the median sputum eosinophil count was 1.8%, compared to the value of 0.3% reported for normal controls.[23] Chang et al. reported that an increase in cough and eosinophilic inflammation were characteristics of mild asthma exacerbation.[24] Li et al. in their study of 32 children with stable severe asthma demonstrated a positive correlation between cough frequency and eNO.[6] Our group of patients are however, very different from those involved in the above two studies. Our patients were in remission and their asthma has been in a stable state for at least 4 weeks prior to this study. They were all mild asthmatics and only became symptomatic with respiratory tract infections. Many of them were using only inhaled beta-agonists on an infrequent basis. The failure to demonstrate a positive correlation between measured cough frequency and eosinophilic inflammation as quantified by percentage sputum eosinophil and eNO may be related to the small sample size. On the other hand, our result could raise the question of whether cough is a reliable marker of the degree of eosinophilic inflammation in mild asthmatics during the non-acute state. Interestingly a significant correlation between increased cough with percentage sputum neutrophil count was demonstrated. We did not find any direct relationship between increased cough or percentage sputum neutrophil count with tobacco smoke exposure or environmental pollution. The daily pollution index was no different a week before the subjects underwent assessment and on the day of the study. Besides, none of the subjects complained of a change in their cough frequency during the study period. In children with asthma, neutrophilic inflammation is classically seen in acute severe exacerbation.[25] Whether the detected increased cough actually heralds the onset of an asthma exacerbation cannot be answered by this study as measurements were performed only at a single time point. In adults, neutrophilic inflammation typically occurs in the more severe form of asthma and reflects an infective aetiology.[26,27] We did not examine for a possible infective cause for the increased cough found in our cohort of patients. None of them had complained of any symptoms suggestive of respiratory
infections prior to and during the study. However, we are not able to exclude the presence of a sub-clinical ongoing infection causing the increased cough frequency. Nonetheless, there is accumulating evidence to suggest that neutrophilic inflammation is involved in enhanced bronchial reactivity and exacerbation of asthma.[28,29] The importance of neutrophil as the dominant inflammatory cell in more severe phenotypes of asthma has also been well established in adults.[30] There is evidence to suggest that children with mild intermittent asthma do not have classical eosinophilic airway inflammation as seen in those with more severe disease.[31,32] Stevenson et al. [31] demonstrated low or absent eosinophil counts in a group of children with symptoms only precipitated by viral infections and with predominantly mild disease, and in many ways similar to our population. Fitch et al. [32] showed that in children with cough as a prominent symptom, there appeared to be a significant increase in neutrophil count in bronchoalveolar material compared to other symptomatic and control populations. In this study, we have provided further evidence that neutrophilic rather than eosinophilic inflammation might be a more important mechanism in driving on-going symptoms in children with mild intermittent asthma.

There are several limitations of this study. Firstly, we recruited only 36 patients. Whether an increased sample size would have demonstrated a significant correlation between cough frequency and other parameters is not known. We did not have normal Chinese children to serve as comparison to our cohort of patients but it was not the intention of this study to compare cough frequency between asthmatics and normal children. Using data from a previous study that included Caucasians [13] may not be appropriate, as environmental and racial characteristics of the two cohorts could have accounted for their differences in cough frequency. Secondly, this study was a cross sectional study at a specific time point. We did not attempt to assess whether the current medications taken by the patients were optimal or not. Longitudinal studies to determine whether increasing asthma treatment or the use of anti-neutrophilic measures (such as antibiotics) are useful strategies to treat increased cough should be carried out and would provide important insight into the pathophysiology of cough in asthma. The main strength of this study however, was the use of an objective cough monitor relying on two signals to confirm genuine coughing of the child. Sound quality may be affected by the sleep position of the patient and the actual recordings may be contaminated by sounds made by people nearby. Therefore, by utilizing two signal modalities to confirm an episode of cough greatly improved reliability and accuracy of the recording.

In summary, we were able to demonstrate increased cough frequency in children with mild asthma despite their disease being in remission. The increased cough might be driven by a neutrophilic inflammatory pathway, the significance of which will require further studies to elucidate.

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Competing Interests – nil declared.
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References


Figure 1. Cough frequency distribution

Figure 2

Scatterplot showing relationship between cough frequency and sputum neutrophils