

Effect of Ascorbic Acid on Increased Bronchial Responsiveness during Upper Airway Infection

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Abstract. We investigated the acute effect of ascorbic acid on histamine bronchial responsiveness (PC_{20} : concentration causing a 20% fall in FEV_1) in 9 hospital staff members with upper respiratory tract infection (URI) and cough. Subjects were examined within 5 days from the start of illness and 6 weeks after. On day 1, the reproducibility of PC_{20} was assessed by 2 consecutive inhalation challenges 1 h apart; the two values were closely related ($r = 0.96$, $p < 0.001$). Five subjects had bronchial hyperresponsiveness ($PC_{20} < 8$ mg/ml histamine). On the following day, PC_{20} was measured before and 1 h after oral intake of 2 g ascorbic acid. Vitamin C produced a significant increase in average PC_{20} ($p < 0.01$) from $7.8 \pm (SE) 1.2$ to $25.1 \pm (SE) 1.2$ mg/ml. None had airway hyperresponsiveness after treatment. Six weeks after the onset of URI, bronchial responsiveness was normal in all the subjects but one. The mean PC_{20} was $15.5 \pm (SE) 1.25$ mg/ml, significantly higher than during URI ($p < 0.05$); after ascorbic acid it increased nonsignificantly to $25.7 \pm (SE) 1.35$ mg/ml.

Our results indicate that vitamin C inhibits the transient increase in bronchial responsiveness occurring in otherwise normal subjects during URI.

Introduction

Infections of the upper respiratory tract (URI) may cause a transient increase in nonspecific bronchial responsiveness in normal subjects [1, 2]. This is mainly observed in those who complain of symptoms from the lower respiratory tract, such as cough [1, 3, 4]. The mechanism responsible for the increased responsiveness is

presently unknown. Histological specimens from experimental or naturally occurring infections [4, 5] indicate a relation with inflammation and disruption of the respiratory epithelium. It has been proposed that inflammatory mediators, such as cyclooxygenase products of arachidonate metabolism, may play a role in the pathogenesis of bronchial hyperresponsiveness [6, 7], even if their relative impor-

tance is still debated. Moreover, activated human phagocytes generate antimicrobial reactive oxidants [8] which, when released extracellularly, damage the surrounding tissues. Ascorbic acid (vitamin C) has been shown to neutralize such extracellular phagocyte-derived oxidants [8] and to shift the cyclooxygenase pathway from synthesis of bronchoconstrictor PGF_2 toward dilator PGE_2 [9].

In view of these metabolic properties, we wondered whether vitamin C could inhibit the transient increase in bronchial responsiveness caused by URL

We therefore carried out an open self-controlled study on the acute effects of ascorbic acid on airway responsiveness to histamine in 9 volunteers with URI and cough. The subjects were examined during their illness and 6 weeks after the onset of symptoms.

Subjects and Methods

The study was performed during the winter season. The volunteers were selected among otherwise normal members of the hospital staff with upper airway infection with cough. URI was characterized by: nasal discharge or congestion, fever, cold sores combined or not with sore throat and laryngitis. Due to lack of virus isolation facilities, a specific diagnosis was not obtained. Criteria for inclusion were: a negative history of asthma and atopy, negative skin tests to seven common aeroallergens, forced expiratory volume (FEV_1) > 90% of predicted and a measurable spirometric threshold to inhaled histamine. Subjects were informed about the aim of the study and gave written informed consent.

The volunteers were examined at the same time of day, on 3 occasions. On each study day, after baseline spirometry, they received 2 consecutive histamine bronchial challenges, 1 h apart (or more if FEV_1 had not returned to baseline).

On the first examination, performed 3-5 days after the onset of symptoms, the subjects' sensitivity to

histamine and the reproducibility of airway response was assessed. On the following day, the broncho-provocation test was performed before and 1 h after oral intake of 2 g ascorbic acid.

On the 3rd day, 6 weeks from the start of symptoms, the protocol of day 2 was repeated to control subjects' sensitivity to histamine and the acute effect of ascorbic acid after recovery. Ascorbic acid was administered in chewable tablets formulated with orange and lemon flavours.

The subjects were asked to refrain from antihistaminic drugs for the whole study period and to avoid anti-inflammatory drugs or foods and beverages containing ascorbic acid the day before each observation. Intake of coffee or tea was not allowed on a study day.

Baseline readings of lung volumes and flows, taken as the best of 5 measurements, were obtained by a computerized OHIO 840 spirometer. Bronchial challenges to inhaled histamine were performed according to a standardized procedure [10]. Histamine was delivered in doubling concentrations of 0.5, 1, 2, 4, 8, 16 and 32 mg/ml, by a compressed air nebulizer controlled by a breath actuated MEFAR dosimeter. The dosimeter was set to nebulize for 0.6 s; the AMMD of dried particles was 1.65 μm (GSD 3.3) [11]. Histamine was inhaled by 5 slow vital capacity breaths from the nebulizer. The FEV_1 was measured 2 min after each nebulization and the test was stopped when FEV_1 had dropped 20% or more or when the highest histamine concentration was reached.

Analysis of Results

The provocative concentration causing a 20% fall of FEV_1 from the control value (PC_{20}) was calculated from the dose-response curve. The histamine concentrations were plotted in a logarithmic scale on the abscissa and the percent change in FEV_1 on the ordinate.

Subjects with a PC_{20} of 8 mg/ml or less, the predictive value for symptomatic asthma [12], were considered to be hyperresponsive. Logarithmic transformation of PC_{20} was used for the statistical analysis. Reference values for volumes and flows were obtained from Knudson et al. [13]. The coefficient of variation of PC_{20} (mean \pm SD) was calculated from the baseline values obtained during URI (2 on day 1 and 1 before vitamin C on day 2).

One-way analysis of variance was used for the

Table 1. General characteristics and baseline values of FEV₁ and PC₂₀ (geometric mean of 3 measurements) of the 9 subjects

No.	Age years	Sex	Smoking	FEV ₁ l	% of predicted	MEF ₅₀ l/s	% of predicted	PC ₂₀ mg/ml	Coeff. var., %
1	19	M	+	5.31	102	8.73	115	8.3	2
2	24	F	-	3.71	95	4.60	73	5.9	3
3	23	M	+	5.62	94	7.23	107	4.5	9
4	18	F	+	3.88	107	4.77	95	7.9	4
5	48	F	-	2.83	107	4.28	102	12.9	2
6	26	M	+	4.52	102	4.80	99	8.7	4
7	51	M	-	4.20	99	3.10	52	2.8	18
8	31	F	-	3.62	96	4.80	99	7.8	3
9	19	M	-	5.37	113	7.36	102	20.4	4
Mean ± SE	28.8 ± 3.9			4.34 ± 0.31	102 ± 2.1	5.46 ± 0.58	94 ± 6.4	8.8 ± 1.7	5.4 ± 1.7

comparison of: (1) baseline values for spirometry on the 3 study days, (2) baseline measurements of PC₂₀ and spirometry during infection, (3) values of PC₂₀ before and after ascorbic acid during infection, and (4) the same after recovery. The 95% confidence intervals of PC₂₀ before and after ascorbic acid were calculated to verify the validity of the significant differences. Reproducibility of PC₂₀ was established by linear regression analysis between the first and the second baseline PC₂₀ on day 1.

Results

Among the 10 volunteers selected, we excluded a woman who had convulsive cough and a raised titre of serum antibodies for *Bordetella pertussis*.

Table 1 shows the patients' data and baseline values of FEV₁ and MEF₅₀ together with the geometric mean and the coefficient of variation of the 3 baseline PC₂₀ (recorded during infection, 2 on day

1 and 1 before ascorbic acid on day 2, in the 9 subjects who completed the study). Prechallenge lung function values were in the normal range in all but 1 subject (No. 7). No significant difference was observed in their values before or after vitamin C. Spirometric values recovered to baseline 1 h after each challenge.

Histamine Bronchial Challenges

Baseline values of histamine PC₂₀ during infection were below 8 mg/ml in 5 subjects. The reproducibility of PC₂₀ on day 1 was very satisfactory: the coefficient of correlation between the PC₂₀ of the first and second challenge was 0.96 ($p < 0.001$). The individual values of PC₂₀ before and after vitamin C recorded during illness (day 2) and after recovery (day 3) are seen in figure 1.

On day 2, ascorbic acid produced an increase in PC₂₀ greater than a single 2-fold

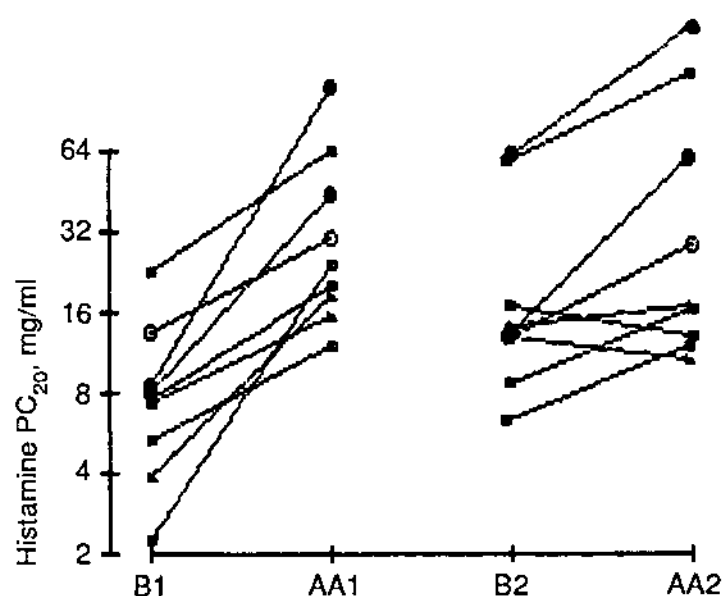


Fig. 1. Individual values of histamine PC₂₀ during URI (1) and after recovery (2), before (B) and after ascorbic acid (AA).

concentration in all the subjects (the woman with whooping cough included), and none was found to have bronchial hyperresponsiveness. The geometric means and 95% CI of PC₂₀ increased from 7.8 ± 1.2 mg/ml in baseline conditions to 25.1 ± 1.2 after ascorbic acid ($F = 17$, $p < 0.01$) with no overlapping between the relative CI.

After recovery, average baseline PC₂₀ was significantly higher than during URI (15.5 ± 1.25 mg/ml, $F = 5.23$, $p < 0.05$). One subject only was still hyperresponsive to histamine and mildly symptomatic. In 2 subjects no bronchial response could be elicited even at the highest histamine concentration. After ascorbic acid, PC₂₀ increased by more than a single 2-fold concentration in 2 subjects only; its mean value was not significantly higher than baseline (25.7 ± 1.35 mg/ml, $F = 1.7$, $p > 0.05$).

Discussion

Our results suggest that ascorbic acid acutely inhibits the transient increase in bronchial reactivity caused by URI in otherwise normal subjects. Its effectiveness is suggested by several points: (1) the increase in PC₂₀ after treatment was greater than a single 2-fold concentration, the upper limit for PC₂₀ reproducibility [14], in all the subjects, (2) the 95% CI of PC₂₀ after vitamin C did not overlap those of the baseline recordings during URI, (3) after treatment, none of the subjects was found to have airway hyperreactivity to histamine, (4) the increase in PC₂₀ after vitamin C was similar to that seen after the recovery from infection.

Six weeks after the onset of URI, PC₂₀ was over 8 mg/ml in 8 out of the 9 subjects, suggesting that its decrease was induced by the disease. At this time, the effect of ascorbic acid did not reach statistical significance. However, any further increase in PC₂₀ would have been of trivial importance as most subjects were asymptomatic and had normal bronchial responsiveness.

Even if the pathogenesis of the increase in airway responsiveness induced by URI is presently unknown, it is supposed to be the consequence of inflammation and disruption of airway mucosa [4, 5]. The increase in PC₂₀ observed in our subjects after treatment could thus depend on inhibition of toxic radicals or mediators of inflammation, such as phagocyte-derived oxidants [8] and prostaglandins [9] by vitamin C.

On the other hand, the increase in PC₂₀ by ascorbic acid during URI could not be due to a change in baseline bronchial tone,

as the vitamin was devoid of any bronchodilator effect. Likewise, it is unlikely that its effect depended on training as the 3 baseline measurements of PC_{20} were highly reproducible. This was due to the fact that our subjects were well trained members of the hospital staff. Moreover, the learning effect on lung function tests is said to be insignificant after 3 repeated measurements [15].

Although the influence of prolonged treatment with vitamin C on prevalence and duration of common cold is controversial, a beneficial effect on symptoms has been reported [9, 16, 17]. We suggest that this effect might depend on a decrease in bronchial responsiveness.

Although airway hyperresponsiveness is not invariably found in subjects with URI [1-4], it is supposed to have a higher prevalence among those who complain of symptoms from the lower respiratory tract, mainly cough [1, 3, 4]. The high prevalence of airway hyperresponsiveness in our series could thus depend on the selection of subjects with cough. On the other hand, the association of symptoms and airway responsiveness is supported by several epidemiologic surveys [18, 19]. An improper inclusion of asthmatics seems unlikely, in view of the subsequent normalization of PC_{20} in most subjects. The high air pollution of our city might also have contributed to the high number of hyperresponsive subjects in our series. High concentrations of pollutants, mostly NO_2 and SO_2 , are known to produce airway abnormalities [20] and to increase bronchial reactivity during infections [21]. The experimental observation that exposure to NO_2 may decrease lung tissue levels of ascorbic acid [22] suggests that

air pollution may increase the oxidative burden caused by infection. Support to our findings is the recent report that vitamin C protects against NO_2 -induced airway hyperresponsiveness in normal subjects [23].

The influence of vitamin C on bronchial hyperreactivity has been investigated in asthma, with inconclusive results [9, 24]. The controversies might depend on the fact that most patients were in clinical remission, so that the degree of their airway inflammation, if any, had to be minimal.

In conclusion, our results suggest that ascorbic acid may be of clinical benefit in a selected population of otherwise normal subjects who develop a transient increase in bronchial reactivity during cold. The vitamin may act by re-establishing the redox state in the inflamed airways or by modulating the release of mediators.

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