

Stability of the circadian alteration in lung function in asthma

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Although studies on pharmacologic interventions for nocturnal asthma are increasing, information about the long-term stability of circadian spirometric changes or bronchial responsiveness is not known. This study was undertaken to evaluate these variables in 10 patients with asthma measured quarterly during a 1-year period. We have found that the overnight decrease in peak expiratory flow rate measurements is stable (mean range, 13.8% to 16.4%) during the year, as are the quarterly 4 PM (1600-hour) and 4 AM (0400-hour) FEV₁ values. The 4 PM provocative concentration of methacholine that produced a 20% fall in FEV₁ (PC₂₀) (range of geometric mean, 0.213 to 0.359 mg/ml) and the 4 AM PC₂₀ (range, 0.057 to 0.152 mg/ml) for the group were also relatively stable. Individual variation was higher for the PC₂₀ values than for the FEV₁. We concluded that during a 1-year period, without acute respiratory events, (1) the overnight decrement in peak expiratory flow rate and the 4 AM and 4 PM FEV₁ values were constant and (2) bronchial responsiveness demonstrated some individual variability, but for the group, it remained stable. (J ALLERGY CLIN IMMUNOL 1992;89:703-8.)

Key words: Nocturnal asthma, lung function, bronchial responsiveness

We have recently demonstrated in a population with asthma¹ that the overnight fall in PEFR is correlated to the severity of the daytime airflow limitation and bronchial responsiveness measured at 4 PM (1600 hour). Additionally, individuals with larger overnight decrements in PEFR have additional marked increases in bronchial responsiveness at 4 AM (0400 hour). In patients with stable asthma, bronchial responsiveness to histamine and methacholine is similar when patients are tested within a week interval^{2,3} or when they are studied during 10 to 30 months.⁴ However, alterations or stability in the circadian changes in lung function and bronchial responsiveness during a prolonged period of time have not been evaluated. The purpose of this article is to describe, in a population with asthma, the circadian stability or lack thereof during a 1-year

Abbreviations used

PEFR: Peak expiratory flow rate
MC: Methacholine challenge
PC₂₀: Provocative concentration of methacholine that produced a 20% fall in the FEV₁
STC: Serum theophylline concentration

period for (1) lung function, (2) bronchial responsiveness, and (3) the relationship between these variables.

METHODS

Subjects

Nine of the 20 adult patients with asthma who participated in our previous study¹ and one additional patient agreed to participate in the present year-long study (Table I). All were nonsmokers. Each patient had at least a 15% improvement in FEV₁ after an inhaled bronchodilator and evidence of moderate to severe bronchial reactivity⁵ in previous challenge studies. All patients had seasonal allergy symptoms and a family history of asthma and/or allergies. There were no other pulmonary problems or occupational-exposure history. Patients were receiving only twice-daily theophylline and/or two to four times daily inhaled β_2 -adrenergic agents. For those subjects receiving theophylline (N = 8), STCs were obtained at each laboratory testing time. All subjects signed an institutional review board approved informed consent.

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TABLE I. Patient's characteristics

Subject	Age (yr)	Sex	Height (cm)	Weight (kg)	Inhaled BD*	Theophylline preparation†
1	36	F	173	120	+	—
2	38	F	177	68	+	+
3	45	M	173	86	+	+
4	46	M	182	81	+	+
5	52	F	162	67	+	+
6	29	M	170	68	+	—
7	27	F	170	63	+	+
8	24	M	180	59	+	+
9	27	M	183	77	+	+
10	26	F	163	60	+	+
$\bar{X}$	35.0	5:5	173.4	75.0	10/10	8/10
SEM	3.1		2.4	5.8		

* β_2 -Agonist, four to eight puffs per day.

†Twice daily preparations.

PEFR

The best of three PEFRs recorded at home, in each case before inhaled β -agonists, at bedtime (10 PM [2200 hour] to 11 PM [2300 hour]) and on awakening (6 to 8 AM), everyday for the week before each quarterly laboratory testing, was used to determine the severity of the change in PEFR overnight in each patient. The difference in the mean PEFR at bedtime and difference on awakening divided by the bedtime value was expressed as the percentage overnight change in PEFR.

Methacholine testing

Bronchial responsiveness to methacholine was assessed at four time intervals during the year, in the months of May, August, November, and February, and performed at 4 PM and 4 AM each time. The time of the initial assessment was randomized, and the second challenge was performed 12 hours later. For the 4 PM MC, the subjects rested in a chair for 30 minutes before the testing. For the 4 AM MC, subjects slept uninterrupted in the laboratory from their usual bedtime (between 10 PM and 11 PM) until 3:45 AM.

MC was performed after obtaining two reproducible FVC determinations within 8% of each other. The equipment and techniques were previously described in detail.¹ The testing was concluded when the PC_{20} of methacholine was attained. In the laboratory, identical intervals between medicine administration before each testing was maintained, and no medication was taken less than 6 hours before testing. Since subjects 7 to 10 had >20% decrease in FEV_1 after inhaled normal saline at 4 AM, the PC_{20} was calculated with the lowest methacholine concentration of 0.0175 mg/ml and the baseline spirometry. This method has validity at 4 AM in that those individuals who have >20% fall to inhaled normal saline fall to the same degree to 0.0175 mg/ml of methacholine.¹ Additionally, the consistency of this saline re-

sponse at 4 AM was observed in this study since subjects 7 to 10 all reacted to saline at each point throughout the year.

Statistics⁶

For each variable measured, a univariate repeated-measures analysis of variance was fit with period (month) and time as the main effects. When the time effect was significant, linear contrasts were constructed, within the context of the repeated-measures analysis of variance models, to test for time differences at each period separately. The interaction between period and time was also included. The same model was fit for the overnight change in PEFR excluding the time effect. When the interaction was not significant, it was dropped from the model, and the model was refit. Correlations were performed with Pearson's correlation test. Statistical analysis of the PC_{20} was performed on the logarithmic transformation of these data since the raw values had a skewed distribution.

To test for individual variability in FEV_1 measurements during the year, the coefficient of variation was calculated and expressed as a mean. A >10% coefficient of variation was considered to be indicative of clinically significant variation in the FEV_1 . For individual variability in bronchial responsiveness, the standard deviation of the logarithmic value was calculated for each individual with a value >1 (a doubling dose), indicative of clinically significant variation in the PC_{20} . The McNemar's test was used to determine if a difference existed in the variability of FEV_1 and PC_{20} measurements.

A p value of <0.05 was considered significant.

RESULTS

Nine of the 10 subjects completed all testing. Subject No. 10 was not tested during the last quarter of

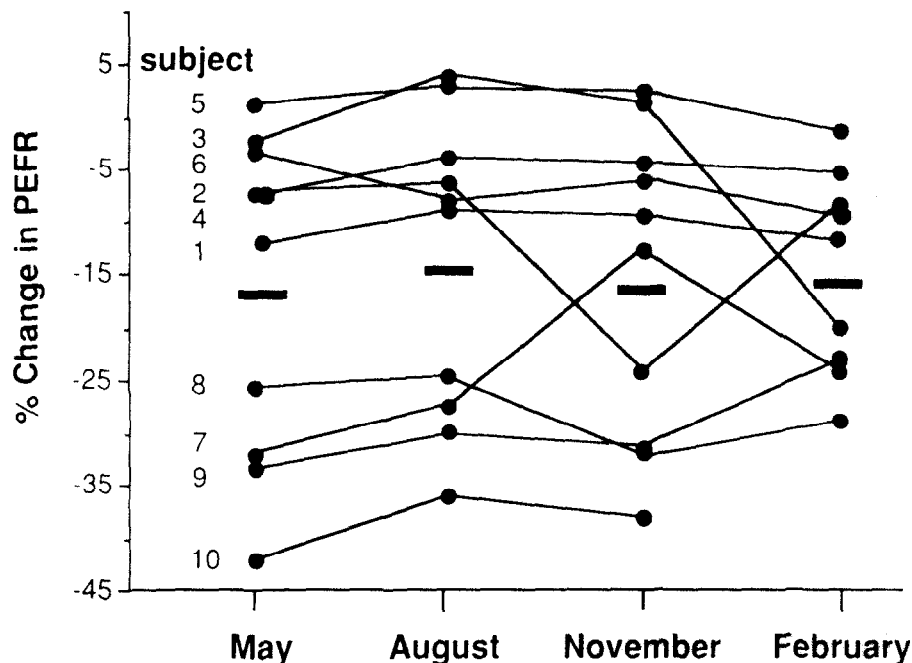


FIG. 1. Individual overnight change in PEFR during a 1-year period. Solid bars represent mean for each period. Subjects 3, 6, and 7 demonstrated variation in PEFR, each for only one period.

the study because of a move from Colorado. For each repeated-measures model described below, the interaction between time and period was not significant and was therefore dropped from the model.

PEFR

The individual results of the overnight changes in PEFR for the four testing periods during the year of study are illustrated in Fig. 1. The overnight decrement in PEFR stayed relatively constant ($p = 0.61$) with the means ranging from a fall of 13.8% to 16.4%. Individually, only three subjects (Nos. 3, 4, and 7) varied by >10 percentage points during the year, and this change occurred for each subject at only one period. This finding was not related to symptoms of a respiratory infection, changes in rhinitis, or sinusitis.

FEV₁

The individual data for the FEV₁ measurements at 4 PM and 4 AM during the year are presented in Table II. The consistency of lung function during periods ($p = 0.38$) is also reflected in the means of FEV₁ at 4 PM (range, 2.65 to 2.88 L) and at 4 AM (range, 2.21 to 2.50 L). Across periods, the mean FEV₁ at 4 AM was constantly lower than at 4 PM ($p < 0.001$). During the year, at both 4 PM and 4 AM, the coefficient of variation $>10\%$ for the FEV₁ occurred in four of the 10 subjects.

Bronchial responsiveness

During the entire year, the 4 PM PC₂₀ values were not statistically different (geometric mean range, 0.214 to 0.359 mg/ml), nor were the 4 AM geometric means (range, 0.051 to 0.152 mg/ml) (Table II). Across periods there was a consistent increase in mean responsiveness (lower PC₂₀) at 4 AM compared to the 4 PM values ($p < 0.001$). If a more than twofold change in PC₂₀ is taken as a clinically important variable, then analysis of each subject independently demonstrated variability at 4 PM for at least one of the different months tested. Similarly, this holds for subjects 1 to 6 tested at 4 AM. This individual variability could not be related to symptoms of a respiratory infection, changes in rhinitis, or sinusitis.

As previously reported,¹ the response to inhaled normal saline is significantly greater at 4 AM compared to 4 PM (Fig. 2); that is, at 4 AM, certain individuals' airways are so reactive that saline alone can cause a decrement in the FEV₁. For the four periods, the change from the baseline FEV₁ value to inhaled normal saline at 4 PM was -0.7% , -4.2% , -3.4% , and -3.9% , respectively ($p > 0.05$). At 4 AM, the FEV₁ change to saline was -15.1% , -15.0% , -12.5% , and -15.6% ($p > 0.05$). At 4 AM, the decrement in FEV₁ caused by saline inhalation was significantly greater ($p < 0.01$) for each period compared to the 4 PM value.

TABLE II. Longitudinal and circadian data

Subject	4 PM FEV ₁ (L)				4 AM FEV ₁ (L)			
	May	Aug	Nov	Feb	May	Aug	Nov	Feb
1	3.25	2.99	3.02	2.96	2.99	2.70	3.12	3.24
2	2.22	2.26	2.24	2.19	2.18	2.47	2.13	2.22
3	2.63	2.70	3.11	2.78	2.42	2.11	2.35	1.97
4	2.32	2.59	1.71	2.61	2.18	2.16	1.41	2.50
5	2.35	2.61	2.42	2.40	2.53	2.56	2.30	2.22
6	4.44	4.44	4.31	4.14	4.23	4.23	4.34	3.84
7	2.10	2.67	3.67	3.18	1.25	0.95	3.34	1.59
8	2.40	3.04	3.63	3.13	2.45	2.02	2.60	2.63
9	2.78	1.81	2.12	2.55	1.52	1.48	2.15	1.73
10	2.0	2.18	2.13	—	1.17	1.37	1.27	—
$\bar{X}$	2.65	2.73*	2.83	2.88*	2.29	2.21*	2.50	2.43*
SEM	0.23	0.22	0.27	0.19	0.29	0.29	0.29	0.24

Subject	4 PM PC ₂₀ (mg/ml)				4 AM PC ₂₀ (mg/ml)			
	May	Aug	Nov	Feb	May	Aug	Nov	Feb
1	4.83	0.02	0.40	0.26	1.5	0.03	0.54	0.018
2	3.24	2.63	1.25	1.37	2.38	0.096	2.42	1.00
3	10.00	4.38	1.25	2.97	1.93	3.60	1.17	0.018
4	0.358	0.79	0.118	2.19	0.079	0.257	0.018	0.022
5	0.90	2.00	2.05	2.70	1.00	0.90	1.95	1.05
6	0.50	0.093	0.701	0.547	0.018	0.128	0.257	0.171
7	0.018	0.018	0.096	0.036	0.013	0.014	0.013	0.012
8	0.036	0.240	0.177	0.045	0.018	0.012	0.013	0.014
9	0.023	0.036	0.064	0.018	0.012	0.017	0.015	0.011
10	0.020	0.038	0.055	—	0.011	0.012	0.012	—
$\bar{X}$	1.993	1.025	0.616	1.126*	0.696	0.507	0.649	0.257*
SEM	1.029	0.475	0.217	0.404	0.294	0.354	0.283	0.146
Geometric mean	0.308	0.214	0.302	0.359*	0.121	0.080	0.141	0.051*

*Significant difference between 4 PM and 4 AM during same month.

Theophylline concentrations

The STCs were similar between periods at the same testing hours and within the periods at the two different testing hours. For the four testing periods at 4 PM and 4 AM, the STCs were 11.4 ± 1.6 and 11.4 ± 1.9 ; 14.0 ± 1.7 and 12.1 ± 1.3 $\mu\text{g/ml}$; 13.5 ± 1.4 and 12.5 ± 1.4 ; and 12.5 ± 2.0 and 11.5 ± 1.2 $\mu\text{g/ml}$, respectively.

Correlation between overnight change in PEFR with 4 PM and 4 AM FEV₁

At 4 PM for each individual's testing period, the correlation between the overnight change in PEFR and the percent predicted FEV₁ did not reach significance. However, for periods 1 and 3, the correlations did approach significance ($r = 0.59$; $p = 0.07$; $r = 0.58$; $p = 0.08$, respectively). At 4 AM, the percent predicted FEV₁ was significantly correlated with the overnight fall in PEFR for each individual period. These correlations for the months of May, August, November, and February were $r = 0.82$ and

$p = 0.004$, $r = 0.80$ and $p = 0.006$, $r = 0.70$ and $p = 0.02$, and $r = 0.68$ and $p = 0.05$.

Bronchial responsiveness correlation with lung function

There was no significant correlation between the PC₂₀ for any period at 4 PM and the starting percentage predicted FEV₁ at those intervals. For each period, respectively, the correlation was $r = 0.54$, $r = 0.07$, $r = 0.45$, and $r = 0.21$ (all $p > 0.05$). There was a significant correlation between the 4 AM PC₂₀ and the starting percentage predicted FEV₁ for period 2, $r = 0.65$ and $p = 0.04$. However, for periods 1, 3, and 4, the correlation did not reach significance, $r = 0.53$, $r = 0.59$, and $r = 0.53$, respectively.

Variability comparison between FEV₁ and PC₂₀

Since subjects 7 to 10 had little variation in their PC₂₀ values at 4 AM testing because of the response to inhaled saline (see above), variability comparisons

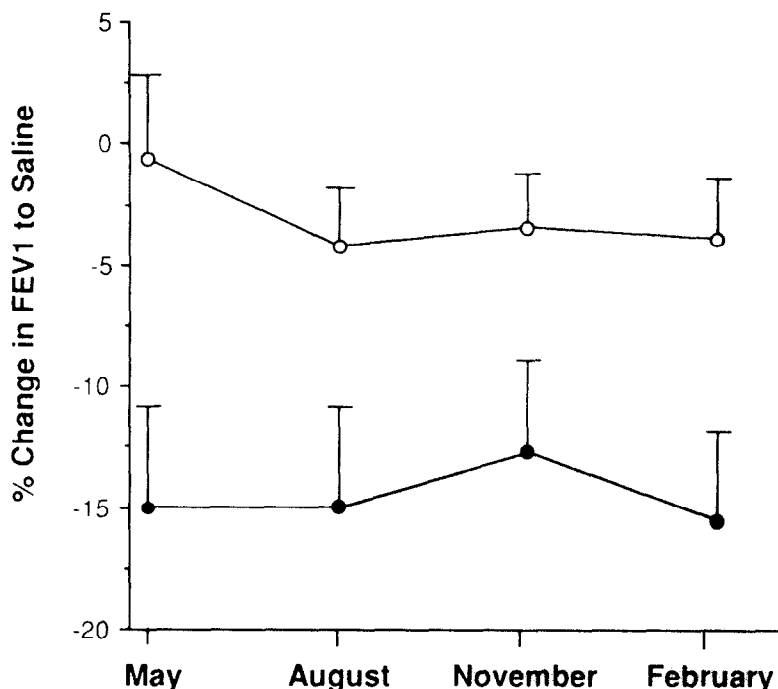


FIG. 2. Response to inhaled normal saline at 4 PM (○) and 4 AM (●) during year; $p < 0.01$, comparing time intervals at each testing period.

were performed on subjects 1 to 6. At 4 AM, there was more variability in the PC_{20} during the year than in the FEV_1 ($p < 0.05$). At 4 PM, this finding was also present but only approached significance in this small sample size ($p = 0.08$).

DISCUSSION

This study demonstrates the relative stability of circadian changes in lung function and bronchial responsiveness during a year-long testing interval in a group of patients with asthma. The mean overnight decrement in PEFR measured quarterly for 1-week intervals only changed substantially in three of 39 possible measurements. Each of these three changes was in different individuals. Of the subjects with asthma who had the greater overnight falls in PEFR ($>20\%$), one (subject 7) improved this decrement to 12.5% during one testing period. Of the subjects with asthma with less of an overnight decrement in lung function, two subjects (Nos. 3 and 4) worsened in the fall on separate occasions to 20% to 22.5%. Otherwise, all 10 individuals remained consistent in their overnight alterations in lung function. This consistency in lung function was also reflected in the 4 PM and 4 AM FEV_1 and PC_{20} measurements during the entire year for the group as a whole. At the 4 AM testing, these values were lower than values at 4 PM. The marked response to inhaled normal saline at 4 AM was also noted in our previous article.¹ Again, this

suggests a marked increase in bronchial responsiveness at night. This response does not imply that saline and methacholine act by the same mechanism but rather that the airways at 4 AM can be exquisitely sensitive.

Our group data are in agreement with data of other investigators who have demonstrated reproducibility of bronchial responsiveness during the daytime during a week interval^{2,3} and up to 30 months⁴ when exacerbating factors, such as upper respiratory tract infections, are not present. However, our study demonstrated more individual variation in the PC_{20} values. This variation was random in nature, and thus, the group consistency remained intact. The difference between our study and that of the other studies^{2,4} most probably lies in the selection of the study population. Each of the other studies selected subjects with relatively mild asthma and with a percentage of predicted $FEV_1 > 70\%$. Additionally, one study used normal volunteers.² In our study, the mean initial daytime percentage of predicted FEV_1 was 73.5% (range, 54% to 103%), and during the night, it was 64.7% (range, 36% to 102%). Thus, our patient population had more severe asthma, which probably accounts for the increased intrasubject variability.

Another possible explanation for the individual variability in the PC_{20} values in our study would be the development of a respiratory infection or allergen exposures. We could not symptomatically relate these

possibilities to changes in bronchial responsiveness. Josephs et al.,⁷ in a longitudinal study, found results similar to our results in that the daytime bronchial responsiveness was significantly correlated to the morning PEFr and circadian variation in PEFr. Additionally, within-subjects variation in the PC₂₀ measurements occurred and were not consistently related to concurrent asthma severity, that is, changes in symptoms or PEFr.

In regard to possible seasonal alterations in bronchial responsiveness, our study design did not allow for an assessment of this potential effect, and the literature is not clear if seasons alter the PC₂₀. Sotomayor et al.⁸ did demonstrate seasonal increases in responsiveness to carbachol. However, findings by Rosenthal et al.⁹ did not reveal any change in bronchial responsiveness to methacholine before and during ragweed-pollen season in patients with asthma or hay fever.

It is recognized that some inherent problems exist with the interpretation of changes in bronchial responsiveness to methacholine or histamine and that differences in responsiveness may reflect differences in baseline FEV₁.^{10, 11} Other investigators, however, have demonstrated that responsiveness does not appear to be related to the FEV₁.^{1, 12, 13} For the present study, there was no correlation at 4 PM between starting percentage predicted FEV₁ and PC₂₀ at any testing period. At 4 AM, as lung function worsened, only periods 1 and 2 revealed a significant correlation between starting percentage predicted FEV₁ and PC₂₀. Thus, overall, our data suggest that there was a true increase in bronchial responsiveness during the night.

Our data demonstrate that the overnight decrement in PEFr is a consistent finding throughout a year period of time in otherwise nonacutely ill patients with asthma. Additionally, the diurnal and nocturnal bronchial responsiveness is also constant for the group, and both times correlate well to the nocturnal alterations in PEFr. Individual differences, particularly in PC₂₀ values, occur, but group values remain relatively constant. These are important observations for long-

term studies attempting to alter the circadian changes in lung function and/or bronchial responsiveness by pharmacologic or other types of intervention.

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