Bronchodilator Pretreatment Improves Aerosol Deposition Uniformity in HIV-Positive Patients Who Cough While Inhaling Aerosolized Pentamidine*

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Objective: To determine the effect of bronchodilator pretreatment on deposition uniformity of aerosolized pentamidine (AP) in HIV-positive patients who coughed while inhaling AP.

Design: Nonrandomized control trial.

Setting: A university hospital.

Patients: Ten HIV-positive patients who were using AP prophylactically.

Intervention: Four patients who coughed during AP administration were pretreated with 10 mg metaproterenol aerosol prior to a second inhalation of AP.

Measurements: Skew, a measure of overall deposition symmetry, deposition in the apex vs the base of the right lung (A:B ratio), and percentage of change from baseline in peak expiratory flow rate (PEFR).

Results: At baseline, the average (± SD) skew value for four subjects who coughed (0.89 ± 0.19) was significantly higher than for six control subjects (0.58 ± 0.07) (p<0.01), indicating enhanced nonuniformity of AP distribution. After bronchodilator, no one coughed and the average skew value was normalized to 0.57 ± 0.13. The A:B ratios at baseline and after metaproterenol were not significantly different, suggesting that deposition of AP in the apex, relative to basal deposition, was not enhanced by bronchodilator treatment. When no bronchodilator was administered, average PEFR decreased to 330 ± 162 from baseline (410 ± 84). Average PEFR increased to 429 ± 85 from baseline (395 ± 110) after bronchodilator pretreatment.

Conclusions: These results suggest that in addition to relieving cough in patients receiving AP prophylactically, pretreatment with metaproterenol enhances uniformity of distribution of AP and improves PEFR.

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AP=aerosolized pentamidine; MMAD=mass median aerodynamic diameter; PCP=Pneumocystis carinii pneumonia; PEFR=peak expiratory flow rate

Clinical trials have demonstrated that oral administration of trimethoprim-sulfamethoxazole is the preferred method of prophylactic treatment against Pneumocystis carinii pneumonia (PCP) in HIV-infected patients. Nevertheless, up to 50 percent of patients using this drug have adverse reactions that may necessitate changing to an alternate treatment. Inhalation of aerosolized pentamidine isethionate (AP) has also been shown to provide effective prophylaxis. In previous clinical trials, however, a subset of patients treated with AP developed cough (38 percent) while receiving a 300-mg monthly dose of AP. It has been recommended that these patients inspire a bronchodilator before pentamidine treatment to eliminate cough. Although bronchodilators may relieve this symptom, to our knowledge, there are no studies to demonstrate that this pretreatment improves the deposition of AP in the lungs. Since the P. carinii protozoan is found predominantly in alveolar spaces near the epithelial cell surface, it is essential that the dose of pentamidine penetrate beyond the conducting airways and deposit uniformly in the lung periphery so that potential or existing sites of infection are targeted. In this study, AP deposition uniformity was quantified before and after bronchodilator (metaproterenol) pretreatment in HIV-positive patients who coughed while inhaling pentamidine.

METHODS

Subjects

Ten outpatients were recruited from the HIV Clinic of the Johns Hopkins Hospital. They were HIV positive, had CD4 cell counts <200/mm³, and were using AP prophylactically. Written informed consent was obtained from each patient and the protocol was approved by the Institutional Review Board. The study subject characteristics are shown in Table 1. Four patients reported cough during AP treatment. Their average age was 39 years. Two were men; two were women. Two had a history of PCP. One patient had a history of asthma as a child. All four had a smoking history. The FEV₁/FVC ratio (an indicator of bronchial obstruction), measured on a screening day, was abnormal in patient 2. Three patients were injecting drug users. Six subjects who did not report any cough with AP served as control subjects. Their average age was 33 years. All were men. Four had a history of PCP, one had a history of asthma while in college, and one (patient 7) had an abnormal FEV₁/FVC ratio on...
the screening day. Two patients had a smoking history. None had a history of injecting drug use.

Aerosol Generation and Delivery

Aerosol was generated from a 50-μg/ml solution of pentamidine isethionate (Lyphomed, Inc, Rosemont, Ill) by a nebulizer (Respigard II, Marquest, Englewood, Colo) as recommended by the FDA. Compressed air flowed continuously into the nebulizer from a wall-attached hospital flowmeter set at 6 to 7 L/min. Aerosol was inspired by the patient while sitting upright. During exhalation, aerosol was directed through a one-way valve and collected on a particle filter.

Aerosol Particle Sizing

To determine the aerodynamic particle size characteristics of AP, the pentamidine solution was radiolabeled with 99mTc sulfur colloid (Syncor International Corp, Chatsworth, Calif). Radioaerosol was generated by the nebulizer (Respigard II) and sampled for 1 min by an impactor (Andersen Mark II, Andersen Samplers, Inc, Atlanta). Flow through the impactor was 28 L/min, which simulated the inspiratory flow rate of patients during actual inhalation of AP. Radioactivity collected at each aerodynamic diameter interval was determined by counting each impactation stage and filter using a single-probe scintillation detector. Each particle was assumed to contain radioactivity proportional to its mass. Particle size characteristics were expressed in terms of the mass median aerodynamic diameter (MMAD).

Because 99mTc sulfur colloid did not appear to be completely soluble in the pentamidine solution, aerosols made from a mixture of pentamidine and technetium pertechnetate (a more soluble form of the isotope) or 99mTc sulfur colloid alone were also sized as described above. These sizing experiments were carried out to determine if mixing the colloid tracer with pentamidine significantly altered the particle size distribution of AP.

Quantification of AP Deposition Uniformity

Gamma Camera Imaging Procedure: The AP deposition uniformity was quantified from gamma camera images of the lungs following inhalation of radiolabeled pentamidine aerosol, made by admixing 99mTc sulfur colloid with the pentamidine solution. Posterior images were obtained in the sitting position. The control group (n=6) inhaled radiolabeled AP during a single visit to the Johns Hopkins Hospital Nuclear Medicine Clinic. They inhaled AP, while breathing slowly and continuously from functional residual capacity, for an average (±SD) of 4.8±2.2 min. No one coughed. After inhaling the aerosol, a lung image was acquired in a 256×256 picture element (pixel) matrix using a large field of view gamma camera (Technicare Gemini; Solon, Ohio). Imaging proceeded until 50 to 100,000 counts were recorded. Images were analyzed in a 64X64 matrix using a computer (Star II, GE, St. Albans, Herfordshire, England).

The four subjects who had a history of coughing during AP administration were studied on two different occasions, 3 to 8 days apart. On a baseline day, they inhaled a radiolabeled gas (xenon 133) to equilibrium first and then they underwent an imaging procedure while sitting upright. The resulting scan represented a ventilation image of the lungs. In analyses of aerosol deposition (see below), the ventilation scan was used to define the right lung region. Following the ventilation scan, subjects inhaled AP without radiolabel until they began to cough (4.9±3.6 min). Then, they inhaled radiolabeled AP for an average of 1.9±1.2 min. After inhaling the radioaerosol, a lung image was recorded. On day 2, the four subjects inhaled 10 mg of the bronchodilator, metaproterenol, generated by the nebulizer (Respigard II), approximately 5 to 10 min prior to inhalation of AP. Then, they inhaled unlabeled pentamidine for the same amount of time that produced coughing on the baseline day. At that time point, subjects inhaled radiolabeled AP, as described above, and underwent another gamma camera imaging procedure.

Gamma Camera Image Analyses: The gamma camera images of the lungs were analyzed in two ways. First, frequency distribution histograms were generated from each of the radioaerosol deposition images of the entire lung field (right and left lungs combined) for all ten subjects. This method of image analysis has been used by us previously to distinguish normal subjects from patients with asthma. The number of pixels with a given count value (expressed as a percentage of total lung pixels) was represented on the Y axis of the histogram and the count values were represented on the X axis. Histograms were scaled to a maximum pixel value for comparability. The histograms were analyzed for skew (a measure of histogram symmetry). For this analysis, low values of skew indicated a more even distribution of aerosol per pixel, whereas high skew values indicated nonuniform aerosol deposition. Skew values, therefore, represented a measure of distribution uniformity on a local level.

Right lung images of the four subjects who coughed during AP treatment were analyzed a second way. First, the right lung region in the xenon scan was divided into apical, intermediate, and basal zones by dividing the vertical distance between the highest and lowest points of the lung into three equal segments as described previously by Agnew et al. Mean counts per pixel in

### Table 1—Study Subject Characteristics

<table>
<thead>
<tr>
<th>Subject No./Age, yr/Sex</th>
<th>HIV Positive</th>
<th>History of Pneumocystis Pneumonia</th>
<th>History of Asthma</th>
<th>Smoker</th>
<th>FEV₁/FVC, %</th>
<th>Aerosolized Pentamidine</th>
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<tbody>
<tr>
<td>1/26/M</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>82</td>
<td>Yes</td>
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<tr>
<td>2/50/M</td>
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<td>No</td>
<td>No</td>
<td>Yes*</td>
<td>58</td>
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<tr>
<td>3/42/F</td>
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<td>Yes</td>
<td>No</td>
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<td>81</td>
<td>Yes</td>
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<tr>
<td>4/39/F</td>
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<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>80</td>
<td>Yes</td>
</tr>
<tr>
<td>5/33/M</td>
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<td>No</td>
<td>Yes</td>
<td>71</td>
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<tr>
<td>6/31/M</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Yes*</td>
<td>78</td>
<td>No</td>
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<tr>
<td>7/31/M</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes†</td>
<td>No</td>
<td>83</td>
<td>No</td>
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<tr>
<td>8/42/M</td>
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<td>Yes</td>
<td>No</td>
<td>No</td>
<td>83</td>
<td>No</td>
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<td>Yes</td>
<td>No</td>
<td>No</td>
<td>81</td>
<td>No</td>
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<tr>
<td>10/33/M</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>81</td>
<td>No</td>
</tr>
</tbody>
</table>

*As a child.
†As teenager.

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The apical and basal zones were calculated and expressed as an A:B ratio. These same zones were superimposed on the right lung regions of the two aerosol scans. Mean counts per pixel in the apical and basal zones of the aerosol scans were calculated and expressed as an A:B ratio. Aerosol A:B ratios were divided by the xenon A:B ratio to correct for lung volume differences in the two regions. Ratios of 1.00 indicated similar deposition on a per pixel basis in the two regions. Lower ratio values indicated enhanced deposition in the basal zone relative to the apical zone.

**Pulmonary Function Measurements**

A peak flow meter (Mini-Wright, Armstrong Medical Industries Inc, Lincolnshire, Ill) was used to measure peak expiratory flow rate (PEFR) at baseline and after pentamidine inhalation on both study days in the four subjects who coughed.

**Statistics**

Skew values for the patients who coughed (n=4) on the baseline day were compared with skew values for the control subjects (n=6) using a Mann-Whitney U test (two-tail). Skew values post-bronchodilator (n=4) were compared with skew values for the control subjects using a Mann-Whitney U test (two-tail). Skew values for the four subjects on the baseline day and after bronchodilator pretreatment were compared using a paired t test (two-tail). A paired t test (two-tail) was also used to compare A:B ratios. Paired t tests were used for the last two analyses instead of a Wilcoxon signed rank test because groups of n=4 cannot be compared with the nonparametric test. For the paired t tests, individual data points were transformed to logarithmic values to increase normality in data distribution. P values ≤0.05 were considered to be significant.

**Results**

**Aerosol Particle Size Characteristics**

In six trials, the average MMAD for particles generated from the pentamidine/sulfur colloid mixture was 1.5 μm (geometric standard deviation (SD) =1.6). The average MMAD for particles generated from the pertechnetate/pentamidine mixture was 1.1 μm in two trials (geometric SD=1.6). In one trial, MMAD for particles produced from the sulfur colloid solution alone was 1.1 μm (geometric SD=1.6). No statistical analysis was applied to these data because of the small n.

**Gamma Camera Images**

Figure 1 is the gamma camera image of the lungs of one of the control subjects after inhalation of pentamidine on the baseline day. Qualitatively, aerosol deposition in this subject appears to be uniform. There are no “hot spots” that would indicate higher concentrations of radioactivity or drug. Lung margins are well defined, indicating aerosol penetrated into the lung periphery.

Figure 2 is a gamma camera image of the lungs of one of the four subjects who coughed during pentamidine inhalation on the baseline day. The numerous “hot spots” indicate that aerosol deposited unevenly throughout both lungs. Lung margins are not well defined, indicating that less aerosol penetrated to the lung periphery.

Figure 3 is a gamma camera image of the lungs of the subject shown in Figure 2 after pretreatment with metaproterenol on the second study day. This image appears to be uniform.
value for the subjects who coughed (0.89 ± 0.19) was significantly higher than for the control subjects (0.58 ± 0.07, p < 0.01), indicating enhanced nonuniformity of AP distribution. The average skew value for these patients was significantly reduced to 0.57 ± 0.13 after bronchodilator (p < 0.01) and fell within the range of patients who did not cough.

A:B Ratio Measurements

On day 1, A:B ratios for the four subjects averaged 0.75 ± 0.30 (range 0.42 to 1.15). On day 2, A:B ratios averaged 0.50 ± 0.13 (range 0.35 to 0.63) and were not significantly different from the baseline day, suggesting that deposition of AP in the apex, relative to basal deposition, was not enhanced by bronchodilator treatment.

Peak Flow Measurements

Average PEFR for the four subjects at baseline and after AP, on study days with and without bronchodilator pretreatment, are shown in Table 2. On the day with no bronchodilator pretreatment, the average PEFR at baseline was 410 ± 84. After pentamidine, PEFR values decreased to an average of 330 ± 162. Before bronchodilator pretreatment on day 2, baseline PEFR values averaged 395 ± 116. After bronchodilator pretreatment and pentamidine inhalation, PEFR values increased to an average of 429 ± 85. After bronchodilator pretreatment, some of the subjects coughed during pentamidine administration.

Discussion

To our knowledge, this study is the first to demonstrate an improvement in the distribution of AP as a result of bronchodilator pretreatment in HIV-positive patients. To quantify the distribution of AP, we admixed pentamidine isethionate (50 mg/ml) with the radiotracer 99mTc sulfur colloid before aerosolization and assumed that mixing the two did not substantially alter the particle size characteristics of AP. We used 99mTc sulfur colloid as the radiotracer rather than other commonly used technetium iso-

![Figure 3](http://journal.publications.chestnet.org/pdfaccess.ashx?url=/data/journals/chest/21698/) Gamma camera image of the lungs of the subject shown in Figure 2 after pretreatment with metaproterenol followed by inhalation of radiolabeled pentamidine aerosol. AP distribution appears to be more uniform compared with Figure 2. shows a decrease in “hot spots” and an increase in overall distribution uniformity.

**Skew Measurements**

Measurements of skew are shown in Figure 4 for all subjects. On the baseline day, the average skew

![Figure 4](http://journal.publications.chestnet.org/pdfaccess.ashx?url=/data/journals/chest/21698/) Skew values for the six control subjects (open circles) who inhaled aerosolized pentamidine on the baseline day and did not cough compared with skew values for the four subjects (closed circles) who inhaled aerosolized pentamidine on the baseline day and coughed and after metaproterenol treatment on day 2.

**Table 2—PEFR Measurements at Baseline and After Aerosolized Pentamidine (AP) With and Without Bronchodilator Pretreatment**

<table>
<thead>
<tr>
<th>Subject No.</th>
<th>No Bronchodilator</th>
<th>Bronchodilator</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>( \bar{X} ) PEFR (Baseline)</td>
<td>( \bar{X} ) PEFR (AP)</td>
</tr>
<tr>
<td>1</td>
<td>530</td>
<td>510</td>
</tr>
<tr>
<td>2</td>
<td>390</td>
<td>120</td>
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<tr>
<td>3</td>
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<td>313</td>
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<tr>
<td>4</td>
<td>385</td>
<td>377</td>
</tr>
<tr>
<td>( \bar{X} )</td>
<td>410</td>
<td>330</td>
</tr>
<tr>
<td>SD</td>
<td>84</td>
<td>162</td>
</tr>
</tbody>
</table>

Bronchodilator Pretreatment in HIV+ Patients (Harrison and Laube)
topes, such as diethylenetriaminepentaacetate (DTPA) or technetium pertechnetate, because $^{99m}$Tc sulfur colloid is a nondiffusible agent that remains in the lung during the decay process or is cleared by mucociliary clearance to the gastrointestinal tract where it is excreted. DTPA and pertechnetate, on the other hand, are highly diffusible and are cleared in minutes from the lungs into the pulmonary circulation. Such rapid clearance necessitates higher radioactivity doses to accomplish rapid (1 to 3 min) image acquisitions and increased exposure to radioactivity by other organs due to uptake from the systemic circulation. We tested our assumption that the aerosol particle size characteristics of AP were unchanged by admixing pentamidine with sulfur colloid by comparing the MMAD of the pentamidine/sulfur colloid aerosol with that of a pertechnetate/pentamidine aerosol and an aerosol of sulfur colloid alone. The MMAD for the sulfur colloid/pentamidine aerosol (1.5 μm) tended to be slightly larger than for the pertechnetate/pentamidine aerosol (1.1 μm) or sulfur colloid alone (1.1 μm). However, this small increase in particle size (0.4 μm) probably did not significantly alter the pattern of deposition for the sulfur colloid/pentamidine aerosol compared with pentamidine alone.

Results from this study suggest that in addition to relieving cough in patients receiving AP prophylactically, bronchodilator pretreatment enhances uniformity of distribution of AP on a per pixel basis and improves PEFR. Enhanced uniformity in AP distribution may have been related to an improvement in pulmonary function, as measured by changes in PEFR before and after bronchodilator, since it is well documented that the distribution of aerosol within the lungs can be altered by changes in airway obstruction. Others have demonstrated that airway narrowing exists at flow-limiting segments in normal persons during cough and Smaldone et al have shown that aerosol particle deposition is significantly increased at these flow-limiting segments in dogs during cough. This effect may explain the nonuniform pattern of aerosol deposition observed in HIV-infected patients who coughed during AP administration in the present study. It is unknown if patients who do not cough or have only minimal symptoms would show improvement in the distribution of AP in the lungs with bronchodilator pretreatment.

Another group of investigators has found that aerosol distribution homogeneity of AP is also affected by body position at the time of aerosol inhalation. They generated frequency distribution histograms from gamma camera images of the lungs of HIV-positive patients, following inhalation of a technetium/pentamidine aerosol, in the supine or sitting position. In that study, inhalation of the aerosol in the supine position improved overall distribution uniformity compared with the sitting position. Although this method of administration appears to optimize distribution uniformity, the administration of AP in this manner is not common, probably due to the difficulty of inhaling aerosol while in a supine position.

Any intervention that enhances distribution of AP to the lung apices is probably warranted because previous studies of patients receiving AP for prophylaxis have noted relapses of PCP occurring in the upper lung fields. In the present study, deposition of AP in the apex did not appear to be enhanced by metaproterenol pretreatment, since the A:B ratios on the two treatment days were not significantly different. O'Riordan and Smaldone have reported similar findings following pretreatment with albuterol. They found that the upper to lower deposition pattern was unaffected by the use of albuterol in ten patients undergoing prophylaxis with AP. Fractional deposition within the apex, as a percentage of the inhaled fraction, was not quantified on either study day in the present study. Thus, it is unknown if percentage of deposition to the right apex was enhanced after bronchodilator despite the finding that the A:B ratio was unchanged.

In summary, this study demonstrates the usefulness of bronchodilator pretreatment with inhaled metaproterenol in patients with moderate to severe symptoms of cough while undergoing AP prophylaxis. Distribution of AP within the lungs became more uniform, which may improve the efficacy of prophylaxis against PCP in this subset of patients.

REFERENCES


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