Correspondence

ERRATUM: EXTRACELLULAR SUPEROXIDE DISMUTASE ATTENUATES LUNG INJURY AFTER HEMORRHAGE

To the Editor:

Several errors were noted in our article (1). The F2 isoprostanes values were reversed; when correctly rounded, they should read 2.0 ± 0.2 µg/lung for extracellular superoxide dismutase (EC-SOD) transgenic mice and 3.4 ± 0.3 µg/lung for wild-type mice. The relative absorbance for the nuclear factor kappa B should read 1.5 ± 0.1 in the EC-SOD transgenic mice and 2.5 ± 0.3 for the wild-type mice. In the sixth paragraph of the results section, we wish to change the sentence, “In the EC-SOD transgenic positive mice, there was a smaller increase in isoprostanes after hemorrhage . . .” to read “In the EC-SOD transgenic mice, lung isoprostanes were not statistically different from unhemorrhaged controls.” We do not feel that these changes substantially alter the conclusions of this manuscript.

We regret any inconvenience that this may have caused.

RUSSELL BOWLER
National Jewish Medical and Research Center
Denver, Colorado


PC20 ADENOSINE 5’-MONOPHOSPHATE IS MORE CLOSELY ASSOCIATED WITH AIRWAY INFLAMMATION IN ASTHMA THAN PC20 METHACHOLINE

To the Editor:

Van Den Berge and colleagues compared the PC20 methacholine and the PC20 adenosine 5’-monophosphate (AMP) in two subsequent papers (1, 2). The conclusions of the papers were that the PC20 AMP better reflects (1) the airway inflammation and (2) the steroid-induced improvement. Their conclusions are based on the differences in the correlation coefficients between the percentage of eosinophils in induced sputum and the PC20 AMP (r = −0.49) or PC20 methacholine (r = −0.28) (1) or the change in these percentages (r = −0.43 and r = −0.28) (2).

A correlation between two parameters is always a weak basis to claim a true relation between the two. The association needs to be strong, specific, and repeatable. In this case, the correlation cannot be considered as strong: a r value of −0.49 means that only 24% of the variance is explained by the independent parameter.

In both papers, the noted differences remain untested: the authors only compare the point estimates of the correlation coefficients. They seem not to realize that every point estimate has an uncertainty reflected by the sampling variance, and that a conclusion as to whether or not two estimates truly differ must be based on that variance. The authors state that the PC20 methacholine and the PC20 AMP describe different inflammatory mechanisms and are therefore independent: in several statistical text books, comparative tests are therefore independent: in several statistical text books, comparative tests are

We agree with Dr. Zanen that a monovariate correlation between two parameters is a weak basis to claim a relation between two variables. For this reason, we clearly stated in both papers that the conclusions were based on the outcomes of a multivariate regression analysis, which is the appropriate method to determine the dependency of a variable on a series of independent variables.

In our first paper, we investigated the association between the severity of hyperresponsiveness to AMP or methacholine with FEV1 and inflammatory markers in sputum, blood, and exhaled air in a large group of 120 subjects with asthma (1). We found a dichotomy in the factors explaining the level of PC20 methacholine and PC20 AMP. The FEV1 was the most important explanatory variable for the variation in PC20 methacholine (explained variance [ev] = 18%) with the number of peripheral blood monocytes being a weak additional predictor (total ev = 23%). In contrast, the level of PC20 AMP was predominantly predicted by the percentage of sputum eosinophils (ev = 25%), whereas FEV1 was only an additional independent predictor.

In our second paper, we assessed the relationship between changes in both PC20 methacholine and PC20 AMP after two weeks of treatment with corticosteroids and the concomitant change in FEV1 and airway inflammation (2). Improvement in PC20 AMP was solely associated with reduction in airway inflammation in the multivariate analysis, whereas improvement in PC20 methacholine was associated with both reduction in airway inflammation and increase in FEV1. Moreover, the total explained variance of the model accounting for improvement in hyperresponsiveness was larger for AMP than for methacholine (36% versus 22%). On the basis of these analyses, we concluded that (1) the PC20 AMP better reflects airway inflammation and (2) the corticosteroid-induced improvement in PC20 AMP better reflects the concomitant reduction in airway inflammation (1, 2).

Another point of criticism of Zanen was that the correlation coefficients presented in the first paper in Figure 2 could have been influenced by outlying data of patients who were unresponsive to AMP (2) (in our study those patients were assigned a value of twice the highest concentration of AMP applied). However, the presented correlation coefficients were derived from a nonparametric analysis, and therefore, the presented regression coefficients would not change from any other handling of the truncated data except leaving them out all together. In summary, we feel that the conclusions in both our papers were properly founded.

Maarten Van Den Berge
Huib A. M. Kerstjens
Dirkje S. Postma
University Hospital Groningen
Groningen, The Netherlands


MORE INFLAMMATION THAN LUNG IN EMPHYSEMA

To the Editor:

I believe there may be an error in the computation of quantitative measures of inflammation in emphysema, as described by Retamales and colleagues (1). In their paper, Table 3 reports inflammatory cell numbers, presumably per lung,
with macrophages, lymphocytes, CD4 lymphocytes, CD8 lymphocytes, and CD20 lymphocytes all having a multiplier of $10^3$. The numbers of macrophages and CD8 lymphocytes in severe emphysema were $4,000 \times 10^2$ and $1,400 \times 10^2$, respectively. Simple arithmetic shows that these values are virtually impossible.

Using round numbers to make the point, a typical lymphocyte approximates $10 \mu m$ ($0.01 \text{ nm}$, or $0.001 \text{ cm}$, $10^{-3} \text{ cm}$) in diameter. Assuming cubic geometry for computational simplicity, each cell occupies about $10^{-6}$ cm$^3$. Thus, one billion ($10^9$) cells would occupy about $1 \text{ cm}^3$. The volume of 1 L would be occupied by $10^{12}$ cells. Retamales and colleagues report 4,000 times that volume ($4,000 \text{ L}$) of macrophages and 1,400 ($1,400 \text{ L}$) of CD8 lymphocytes in Table 3.

Is it possible that the $10^9$ exponent is incorrect, and that it should be $10^9$ or something smaller? Even at a $10^9$ multiplier, 4 L of macrophages would be physically impossible.

This arithmetic question aside, the authors are to be commended for their careful quantitative histology and morphometry, the intriguing link between adeno viral infection and emphysema severity, and the important and notable (2) insights for the pathogenesis of severe obstructive lung disease.

WILLIAM J. CALHOUN

Asthma, Allergy, and Airway Research Center
Pittsburgh, Pennsylvania


From the Authors:

We write in reply to Dr. Calhoun’s inquiry concerning our estimates of the number of inflammatory cells in the lungs of patients with severe COPD (1). The quantitative approach that we used is designed to calculate the fraction of the tissue and airspace taken up by a particular cell type. This volume fraction was converted to a volume of cells by multiplying it by the total volume of tissue and air measured from the preoperative CT scan. The number of cells was then calculated by dividing the calculated total volume of each cell type by previously reported values of the volume of single fixed cells (2). In our initial analysis, we expressed the number of cells per surface area in cm$^2$ and then corrected this to number per m$^2$ by multiplying by $10^3$. Unfortunately, when we expressed the cells as the total number in the lung to account for the fact that the surface area was markedly reduced in severe emphysema, we made a calculation error. When we re-examined the spreadsheet after receiving Dr. Calhoun’s letter, we discovered (to our chagrin) that the conversion factor was inadvertently carried over, resulting in numbers that were too large by a factor of $10^3$. Unfortu nately, when we corrected this to number per m$^2$ by multiplying by $10^3$, the numbers of macrophages and CD8 lymphocytes in Table 3 from our paper should be the 8th power rather than the 9th power.

We regret any inconvenience this may cause and thank Dr. Calhoun for pointing out that we needed to check our arithmetic. However, our main point that the inflammatory process present in the lungs of patients with severe COPD is amplified compared with persons with similar smoking histories that do not develop airway obstruction is unchanged.

IVAN RETAMALES

Hospital San Borja-Arriraran
Santiago, Chile

W. MARK ELLIOTT

BERNARDO MESSI

HARVEY O. COXXON

SHIZU HAYASHI

PETER D. PATE

JAMES C. HOGG

St. Paul’s Hospital
Vancouver, Canada


THE DISCRIMINATORY CAPACITY OF THE BRONCHODILATOR RESPONSE

To the Editor:

Nielsen and Bisgaard concluded that in children, the specific airway resistance ($R_{sp}$) discriminates best between asthmatic and healthy subjects, because the sensitivity and specificity of that parameter was highest (1). However, that conclusion could be challenged on statistical grounds. Table 3 in the Nielsen and Bisgaard paper lists the sensitivity and specificity of all the parameters examined, together with the positive/negative predictive value. Based on the combination of the highest sensitivity/specificity, indeed the order as is listed by Nielsen and Bisgaard. Based on the combination of the highest positive/negative predictive value, the ranking would be different. The latter serves to indicate the instability of the conclusions, which is caused by the fact that the authors apparently used the point estimates of the sensitivity/specificity to rank the tests. These estimates are random variables and are always imprecise to a certain degree: in other samples these estimates will be (slightly) different and so the ranking of the tests may change. Point estimates will vary, and one has to decide whether differences are due to chance or truly exist.

The authors seem not to test whether the discriminatory power of the parameters under question differs significantly and base their conclusions on the point estimates only. They generated receiver operating characteristics curves, but they do not list an important characteristic, i.e., the area under the curve (AUC). The latter is a widely accepted measure of the discriminatory power of a test (an AUC of 0.5 indicates no power at all, whereas an AUC of 1 is equivalent to a perfect separation). The differences between AUCs can be tested for significance: Hanley designed methods to do so (both for the parallel and cross-over situation (2)). Such an evaluation gives a much better insight into the ranking of the tests than a comparison of point estimates.

P. ZANEN

Universitair Medisch Centrum
Utrecht, The Netherlands

