ON THE RELATIONSHIP BETWEEN THE SINGLE BREATH AND MULTIPLE BREATH NITROGEN WASHOUT TESTS

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SUMMARY

A comparison of methods for measuring the nonuniformity of distribution of ventilation (nitrogen single breath test versus multiple breath nitrogen washout test) in subjects with pulmonary disease was performed. dN_2 (increase in per cent N_2 from 750 to 1250 ml expired volume) was found to be poorly correlated ($r = -0.44$ for 78 subjects when O_2 volume = VC; $r = -0.34$ for 32 subjects when O_2 volume = IC) with IAV (index of alveolar ventilation). dN_2 when O_2 volume = VC correlated somewhat better with total ventilation to 2% N_2 dilution ($r = +0.57$) and with end-alveolar N_2 at 7 minutes ($r = +0.57$). Probable causes for these variations are discussed.

The N_2 single breath test is recommended only as a supplement to the N_2 multiple breath washout test, when abnormal values have been obtained, to determine the influence of nonuniform sequential ventilation.

SINGLE VS. MULTIPLE BREATH WASHOUT TESTS

INTRODUCTION

For clinical usage, an estimation of the extent of nonuniformity of intrapulmonary distribution of ventilation can be obtained with the nitrogen single breath test of Fowler[1]. A quantitative measure of distribution abnormality requires the more extensive multiple breath nitrogen washout test (Bouhuys)[2] first established by Cournand and associates [3,4]. The latter test also gives much more information about the ventilatory efficiency as affected by both volume and distribution of inspired gas.

Each test has been discussed by several authors, from both analytical and diagnostic standpoints. The only reported comparative study of the results obtained when both the single and multiple breath tests are used, however, is that by Fowler in 1952[5]. He stated that the observed correlation ($r = +0.76$) between the values obtained by the two methods in a group of 27 subjects "supports the general validity of the 'single breath' test". Recognition of the different major measures involved (temporal inequality - N_2 Single; spatial inequality - N_2 Multiple: International Labor Office report, 1965)[6] (Bouhuys)[7] has prompted a re-examination of the relationship between the results of these two tests.

The present study was intended to more clearly define the interpretability of the test values and to compare results from tests on a larger number of subjects than was used in Fowler's study, a need explicitly stated by Bouhuys in 1959[2].

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METHODS

Nitrogen multiple breath and single breath tests were performed as a part of the U.S. Public Health Service pneumoconiosis laboratory study of southern West Virginia coal miners. From approximately 140 cases with two or more technically acceptable multiple breath tracings for each subject, 78 cases were chosen with single breath tracings which adhered to the criteria given previously for this test[8]. A comparison of dN_2 (difference in percent nitrogen from 750 ml to 1250 ml of expired gas following a single inhalation of oxygen) and the IAV_a (index of alveolar ventilation - by algebraic computation)[9] was made by a regression analysis performed by digital computer. Computer regression analyses were also made of (a) dN_2 and the end-alveolar N_2 at 7 minutes and (b) dN_2 and the total ventilation to the 2.0% dilution level.

A regression analysis of the IAV_a versus the IAV_p (semi-graphical method)[10] was also performed. This was done to insure that the IAV_a was a true measure of deviation from a straight line semi-log N_2 washout ventilation, as was evident for the IAV_p. A low dead space valve, designed by the author, was used in the multiple breath test to insure the minimal effect of dead space.*

RESULTS

A direct data plot of dN_2 versus index of alveolar ventilation (IAV) is given in Figure I. A separate plot of these same data, with identification of the degree of obstructive and restrictive ventilatory impairment in each case, showed no clear relationship of the differences between these values and gross spirometric flow values.

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* Measured dead space = 16 ml

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Computer regression analyses provided the following comparisons.

x	vs. y	$\bar{x}$	$\bar{y}$	S.D. x	S.D. y	n	S.E.E. y	S.E.E. x	Correlation Coefficient
IAV_p	IAV_a	49.7	52.5	15.7	15.9	291	2.9	2.9	+0.98
dN_2 (MEEP)*	IAV_a	2.5	51	1.5	14.8	78	13	1.4	-0.444***
dN_2 (REEP)**	IAV_a	3.1	51	2.1	17.6	32	16.5	1.9	-0.344****
dN_2 (MEEP)	Total vent to 2% N_2	2.5	45	1.5	.17	78	14	1.2	+0.57***
dN_2 (MEEP)	End Alveolar N_2 at 7 min.	2.5	2.0	1.5		78			+0.57***

* Maximum End-Expiratory Position
** Resting End-Expiratory Position
*** Significant at $<$1% level
**** Significant at 5.1% level (Snedecor, G.W. Statistical Methods, fifth edition, 1961, p.174)

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DISCUSSION

The very poor correlation observed in this study between dN_2 in the single breath test and the IAV as a chosen measure of uneven ventilation in the multiple breath test is in conflict with Fowler's 1952 test results and would not support his conclusion concerning the comparability of the two tests[5]. This difference is, in part, due to the different indexes used (IAV versus pulmonary N_2 clearance delay percentage). Consideration of subsequent discussions by other authors of the causes of distribution abnormality, however, provides an adequate explanation for the observed lack of agreement between these tests. A different clinical interpretation of abnormality, based solely on one of these tests, would, therefore, be anticipated.

The pulmonary N_2 clearance delay percentage has been used by Bouhuys[11,12,13] and Lundman[14]. Lundman provided an equipment modification to homogenize the expired gas before measurement, giving the approximate necessary mean N_2 concentration. Directly obtaining these N_2 values eliminated one portion of the extensive calculation of this index. The index "is designed primarily to assess the degree of nonuniformity" of alveolar ventilation (Bouhuys)[13]. It would be expected to compare favorably with any other measures which are largely dependent upon variation from uniform distribution.

The IAV was chosen because it does not require measurement of the functional residual capacity (FRC) and is a direct composite measure of the deviation from a straight-line dilution course (uniform ventilation) in a semilog plot of percentage nitrogen versus breath number. It "is independent of volume measurements" (Lichtneckert)[9] and the value obtained can be related to an ideal condition, an attribute which is also found in the pulmonary N_2 clearance delay percentage. Improvement of uniformity of distribution is indicated by a larger IAV value, whereas a smaller clearance delay percentage corresponds to an increased distribution uniformity.

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In the IAV, the respiratory dead space is included in the volume to be freed of its nitrogen[9], whereas in the pulmonary N_2 clearance delay percentage, the dead space is excluded in the calculation[5]. This dead space difference, due to the wide variation of Vd/Vt values obtained (20% to 73%) in the subjects investigated in the PHS pneumoconiosis laboratory study, provides a partial explanation as to why the correlation between the N_2 single and multiple breath tests found here was poorer than that found in Fowler's 1952 study.

The major source of variation between the single breath and multiple breath values seems to lie in the basic conclusions previously drawn by several authors regarding sequential (series) versus parallel ventilation. Bouhuys[2,7] summarized his findings and those of other investigators. The result in the single breath test is, apparently, dependent upon several factors, but is principally influenced by asynchronous ventilation which is much more time dependent than volume dependent. The "sloping third phase of a single breath record can only be explained if lung regions with different ventilation rates do not empty in phase"[7]. Lobar spirometry[15] has shown that differences in volume changes occur between lobes during inspiration and the pattern of inspiratory filling usually differs from the pattern of expiratory emptying, supporting the first-in, last-out (sequential ventilation) hypothesis of Fowler. An alteration in filling distribution as a result of a difference in preinspiratory volume has been demonstrated in the single breath test by Milic-Emili[16]. The reciprocal of preinspiratory volume has been shown by Jones[17] to be linearly related to dN_2 in normal subjects using 750 ml O_2 inspired volume and 30 L/min expiratory flow. The entry of dead space gas also influences the outcome, giving a higher nitrogen concentration in early filling and late emptying regions.

A major basis for the alternative, first-in, first-out hypothesis is the theoretical analysis of Otis et al[18], in which they proposed a number of parallel units with separate time constants caused by a resistance and a compliance element in series. They suggest that normally unit compliance is more important than resistance in determining distribution. Flow rates utilized in the single breath test markedly complicate the result with low rates favoring emptying of units with low dV/V ratios (Young, et al)[19].

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Bouhuys[7,21] summarized the reported effects of postural changes on producing detectable distribution changes in the Multiple Breath washout process, but not in the single breath dilution test. He stated that these results "suggest that the gas distribution changes are caused by alterations of the relative volume expansion in different lung regions, and not by changes in the timing of lung emptying"[7].

Consideration for the accuracy of the first-in, first-out (mechanical) hypothesis, at least under some circumstances (Bouhuys)[2,7], lends support for the conclusion that dN_2 in the single breath test is largely, but not completely, a measure of distribution in time. Bouhuys' studies on the effect of posture accentuate the importance of different regional volume changes over solely time-dependent phenomena. The time-dependent phenomena are involved, however, as would be expected, from different bronchial pathway lengths[20] and the contribution of single breaths to the over-all distribution, including the unequal distribution of dead space (mixed) gas.

The dead space volume, acting as a mixing chamber, has been shown to significantly influence the results of multiple breath tests not only by delaying dilution but by producing more uniform distribution[13]. In the single breath test, the dead space effect would be limited to the N_2 inequality resulting from its unequal contribution to different regions.

The somewhat better correlation noted when dN_2 (MEEP) is compared to the total ventilation to 2% N_2 and to the end-alveolar N_2 at 7 minutes may be due, in part, to the differences in effect of hyperventilation on these values. Increased ventilation will produce a general moderate increase in the IAV[22], a marked reduction in the end-alveolar N_2 at 7 minutes, and only a slight reduction in the total volume required to dilute the N_2 to the 2.0% level[23]. The correlation of 0.57, however, may still be considered a distinct lack of close agreement between the results of these tests.

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CONCLUSIONS

The marked differences found in this study between the interpretability of the multiple breath and single breath tests can be attributed to the following:

1. A larger dead space volume was required in the apparatus for the single breath test. The contribution and relative effect of dead space gas would, thus, be different in each test.

2. The single breath test was measured with full inhalation from both volume levels. The distribution pattern would not be expected to be directly comparable to that under tidal volume conditions, as large tidal volumes increase the IAV[22].

3. Anxiety and/or physiological hyperventilation was observed in many subjects. This prevented direct comparisons of dN_2, on the basis of the effects of both preinspiratory volume and tidal volume changes.

4. More complete mixing of gas in the dead space volume would occur in the multiple breath test.

5. Incomplete penetration of inspired gas into poorly ventilated areas in the single breath test has been expected (West)[24].

The higher correlation noted by Fowler[1] may have been due to possible technical errors influencing his single breath measurements[8] and to differences in the nitrogen clearance delay percentage as compared to the IAV, particularly as produced by the effect of dead space on the IAV.

A complete differential assessment of nonuniformity of distribution would seem to require use of both the multiple breath and single breath tests. But, the single breath test should be used only as a supplement to determine what influence the time-dependent factor has on the distribution and only when abnormal uniformity is demonstrated with the multiple breath test. Some error in the presently reported correlation between these tests, due to the effect on distribution of the difference in lung volume and tidal volume used in each case, is recognized. Compliance with previous recommendations (James)[8] will enable a more accurate estimate of the actual difference. This magnitude of error is not expected, however, to be sufficiently important to produce a considerably closer agreement to allow for usage of either test as a substitute for the other. This opinion is supported by a recognition of the different aspects of distribution (relative contribution to the total pattern of distribution) being measured in each test.

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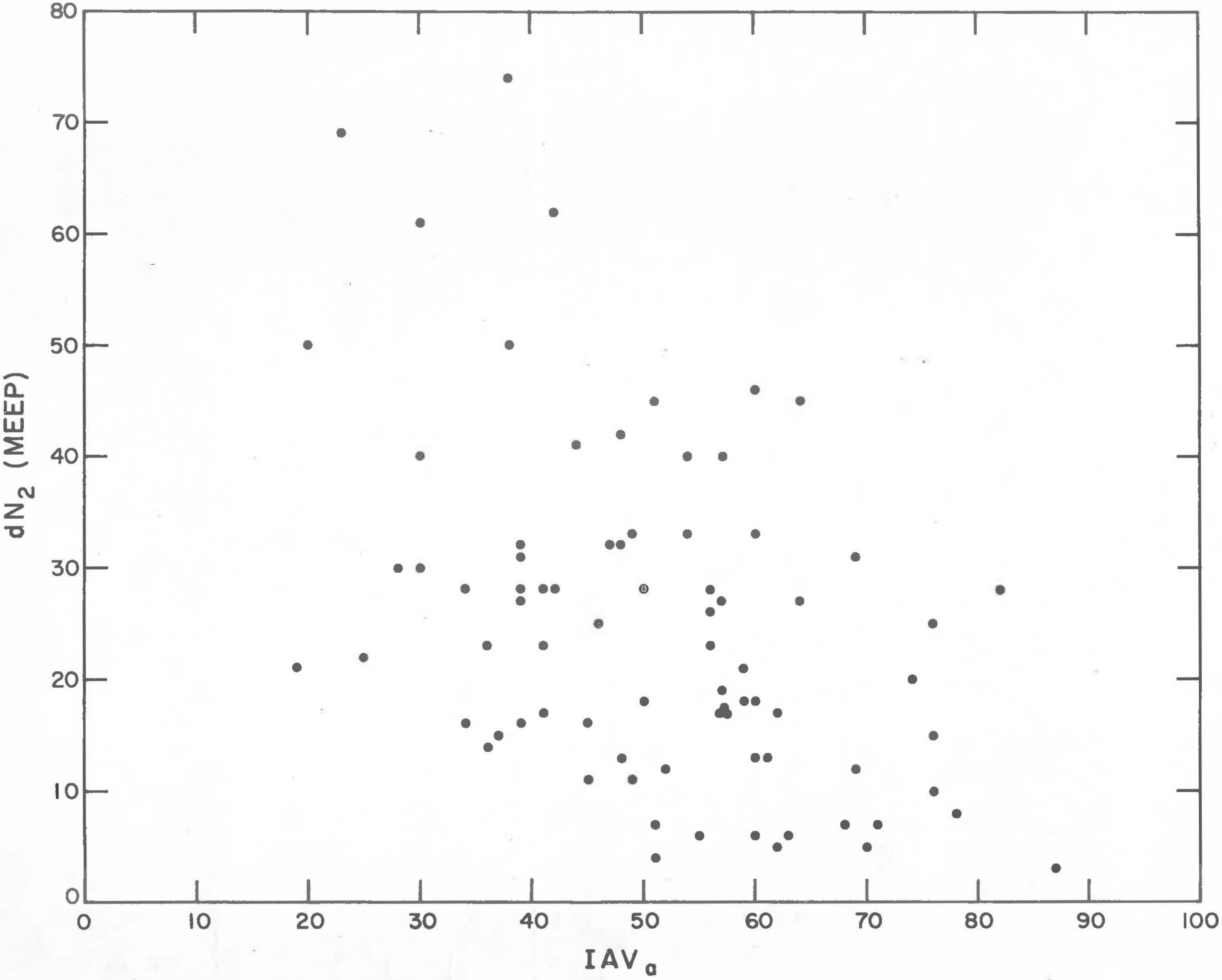

Figure 1. dN_2 in the Nitrogen Single Breath test versus the IAV in the Nitrogen Multiple Breath Washout test. n = 78