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Total desmosines in plasma and urine correlate with lung function

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ABSTRACT: The aim of the present study was to evaluate the relationship between the matrix degradation biomarkers, desmosine and isodesmosine (desmosines), and lung function.

Plasma and creatinine-corrected urinary total desmosines (P- and U-desmosines, respectively), lung function and diffusing capacity of the lung for carbon monoxide ($D_{L,CO}$) were measured in a cohort of subjects from the Swedish Twin Registry.

Concentrations of U- and P-desmosines were measured in 349 and 318 subjects, respectively; approximately one-third of subjects had chronic obstructive pulmonary disease (COPD). Age, female sex, body mass index (BMI) and smoking were significantly associated with U-desmosines in a multiple linear regression analysis. In the overall population, after adjustments for age, sex, height, BMI and smoking, concentrations of U-desmosines were significantly correlated with all lung function measures, and P-desmosines with forced expiratory volume in 1 s and $D_{L,CO}$ ($p<0.05$). With the exception of residual volume *versus* P-desmosines, relationships between concentrations of desmosines and lung function measures were markedly stronger in subjects with COPD compared with those without COPD.

These cross-sectional data showing associations between desmosines and several lung function variables suggest that desmosines, particularly U-desmosines, could be a useful biomarker of COPD status.

KEYWORDS: Biomarkers, chronic obstructive pulmonary disease, cigarette smoking, elastin, emphysema, lung function tests

Chronic obstructive pulmonary disease (COPD) fundamentally affects daily living [1] and in particular, exacerbations of COPD reduce patients' quality of life [2, 3]. The disease is characterised by progressive airflow limitation [4], and clinical symptoms of dyspnoea, cough and sputum production [4, 5]. Classification of COPD is usually based on the severity of airflow obstruction, as assessed using forced expiratory volume in 1 s (FEV_1) [5]. However, there is a lack of biomarkers to measure disease activity and disease progression in COPD [6]. Matrix degradation is a key feature of COPD, leading to lung destruction, emphysema and impaired pulmonary function. The elasticity and resilience of the lungs are mainly provided by elastin, which is synthesised as a soluble precursor, tropoelastin. The elastin fibres are formed by crosslinking through post-translational modification of lysine residues by lysyl oxidase, to form inter- and intramolecular covalent crosslinks [7]. The crosslinking structures, desmosine and isodesmosine (collectively referred to as desmosines), are unique to mature elastin. They are released, either in free form or conjugated to peptides, as a result of elastin degradation, and can be found in sputum, blood and urine [8–10]. Desmosine and isodesmosine have, therefore, been proposed as biomarkers of lung matrix degradation in COPD [11, 12].

Most published studies have analysed urinary desmosines (U-desmosines), rather than plasma desmosines (P-desmosines), probably because of the higher concentrations found in the urine. However, recent studies have shown that P-desmosines might be the preferred analyte, in so far as P-desmosines have demonstrated a clearer distinction between COPD patients and healthy subjects [9, 10].

There is accumulated evidence that the urinary excretion of desmosines is significantly higher in

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smokers than in nonsmokers, and also higher in COPD patients than in subjects with normal lung function (see review by LUISETTI *et al*. [13] and references cited therein). However, different studies have shown slightly different results; the lack of consistency may be due to the small size of the studies, the use of carefully selected subjects or the use of many different analytical methods. The hypothesis tested in the present study was that there would be correlations between desmosines in urine and plasma, and lung function. Desmosines were measured using a highly specific and sensitive analytical method based on liquid chromatography (LC) combined with tandem mass spectrometry (MS/MS).

MATERIALS AND METHODS

Study population

The current study was approved by the Ethics Committee at the Karolinska Institute (Stockholm, Sweden; number 03-461). All participants gave informed consent. The study was based on the Swedish Twin Registry (STR), which contains information on >80,000 pairs of twins (160,000 individuals) born from 1886 to 2000. Between 1998 and 2002, all living twins in the STR born in 1958 or earlier were contacted using a computer-assisted telephone interview developed for the SALT (Screening across the Lifespan Twin) study [14]. The interview included a checklist of common diseases and respiratory symptoms, as well as smoking habits. From a population of 26,516 twin pairs (n=53,032), where both twins participated in the telephone interview, a subgroup was selected (515 pairs; n=1,030). These twins were invited to participate in more in-depth measures of lung function. The selection of subjects was initially conducted for a heritability study, as described previously, in a manner such that disease-concordant and -discordant twins were prioritised over symptom-free twin pairs [15]. The selection was also based on feasibility aspects, such as geography, budget and personnel. In total, 392 (38%) out of 1,030 twins accepted the invitation to participate.

Urine samples were obtained from 349 subjects for measurement of desmosines, and data on spirometry, body plethysmography and diffusing capacity of the lung for carbon monoxide ($D_{L,CO}$) were also collected in these subjects. In addition, blood samples for P-desmosine determination were obtained from 318 of these subjects.

Pack-yrs were quantified from information in the SALT telephone interview and subsequently confirmed using a questionnaire at the clinic; smoking status was defined as current smoker, ex-smoker, never-smoker or occasional (social) smoker. The question was ''Do you smoke or have you previously smoked?'' Respondents chose from four alternative responses: 1) ''no''; 2) ''current smoker''; 3) ''have previously smoked''; or 4) ''only smoke at parties/have only smoked at parties''. Subjects giving the fourth response were defined as occasional smokers. The distribution, based on smoking habits and disease status, of subjects with urine samples available for determination of desmosines is presented in table 1.

Determination of U- and P-desmosines

The concentration of the sum of total (free plus peptide-bound) desmosine and isodesmosine in urine (U-desmosines) and plasma (P-desmosines) was measured by LC-MS/MS (further details are available in the online supplementary information).

TABLE 1 Smoking status and lung function variables according to the Global Initiative for Chronic Obstructive Lung Disease (GOLD) stage[#]

	Lung function			
	Normal	GOLD I	GOLD II	GOLD III
Subjects n	234	67	43	5
Never-smokers	96 (41)	12 (18)	6 (14)	0
Ex-smokers	95 (41)	29 (43)	19 (44)	3 (60)
Current smokers	32 (14)	18 (27)	17 (40)	2 (40)
Occasional smokers	11 (5)	8 (12)	1 (2.3)	0
FEV_1 L	2.9±0.68	2.6±0.60	2.0±0.46	1.3±0.24
FEV_1 % pred	103±12.7	98±11.3	70±6.5	43±5.2
FVC L	3.8±0.94	4.0±0.93	3.3±0.75	3.2±1.16
FVC % pred	113±14.6	121±14.4	98±13.9	87±22.0
RV L	2.1±0.52	2.4±0.51	2.8±0.75	3.5±1.08
RV/TLC	0.36±0.06	0.38±0.06	0.46±0.07	0.52±0.04
$D_{L,CO}$ mL·min^{-1}·mmHg^{-1}	24±6.6	22±5.8	19±6.7	18±3.4

Data are presented as n (%) or mean±SD, unless otherwise stated. No subjects fulfilled criteria for GOLD IV. FEV_1: forced expiratory volume in 1 s; % pred: % predicted; FVC: forced vital capacity; RV: residual volume; TLC: total lung capacity; $D_{L,CO}$: diffusing capacity of the lung for carbon monoxide. #: defined in [16] as GOLD I (mild), FEV_1 ⩾80% pred; GOLD II (moderate), FEV_1 <80 and ⩾50% pred; GOLD III (severe) FEV_1 <50 and ⩾30% pred; GOLD IV (very severe), FEV_1 <30% pred.

Creatinine concentration in urine was measured by LC with ultraviolet detection [16]. Urinary levels of desmosines were normalised by creatinine concentration and are reported as nmol desmosines per mmol creatinine.

Statistical methods

The median and range for levels of U- and P-desmosines were calculated and presented for descriptive purposes. The correlation between the two was assessed using the Spearman rank correlation.

The relationship between U- and P-desmosines and patients' baseline characteristics (sex, age, smoking status, body mass index (BMI) and Global Initiative for Chronic Obstructive Lung Disease (GOLD) disease severity) were also assessed, using the Spearman rank correlation for continuous variables and the Mann–Whitney U-test or Kruskal–Wallis test for categorical variables.

A general linear model was used to compare log U-desmosines and log P-desmosines in GOLD stage categories, with adjustments for age and sex. Subjects without COPD were the reference category. The anti-log values (*i.e.* geometric means) were calculated to facilitate the interpretation of the log-transformed measures.

Multiple linear regression analysis was used to explore whether the following factors were associated with desmosine concentrations: age, sex, height, smoking status and BMI. Sex and smoking (current and occasional smokers *versus* never-smokers and former smokers) were modelled as dichotomous variables; age, height and BMI were modelled as continuous

variables; and levels of U- and P-desmosines were included as dependent variables.

Multiple linear regression was also used to assess the relationship between U- and P-desmosines and lung function measures (FEV_1, FEV_1% predicted, forced vital capacity (FVC), FVC % pred, residual volume (RV), RV/total lung capacity (TLC) and $D_{L,CO}$), with lung function measures as the dependent variables. Predicted values for FEV_1 and FVC were calculated using published reference equations [17]. A separate analysis of the relationship between desmosines and lung function was conducted to compare those with and without COPD (defined as $FEV_1/FVC < 0.70$ according to GOLD [18]). In both analyses, the data were adjusted for confounding factors, *i.e.* factors found to be independently associated with desmosines in the first part of the analysis. In the first step, age, sex and height were assessed, and in the second step, smoking and BMI were added.

Due to positively skewed distributions, log-transformed (natural logarithm) U- and P-desmosine levels were used in all multivariate analyses. The relationships between lung function measures and desmosines are presented as standardised β-coefficients, in order to increase the comparability between the lung function measures.

The relationship between concentrations of desmosines and lung function was also studied within twin pairs of the same sex. The desmosines and lung function values of the first twin were subtracted from the corresponding values in the second twin. Relationships between the differences were assessed using Spearman rank correlations.

PASW statistics software (version 18) was used for all calculations (IBM, Stockholm, Sweden).

RESULTS

Factors associated with concentrations of desmosines

U-desmosines were measured in 349 subjects, while P-desmosines were measured in 318 of these subjects. The Spearman correlation between U- and P-desmosines was $r=0.70$ ($p<0.001$). The median levels of U- and P-desmosines according to patients' baseline characteristics are presented in table 2. Age was significantly correlated with both U-desmosines (Spearman's $r=0.53$, $p<0.001$) and with P-desmosines ($r=0.55$, $p<0.001$). The correlations with age were observed in both males and females, in smokers and nonsmokers, and in subjects with and without COPD ($p<0.005$ for all groups).

Results of the multiple linear regression analyses to explore factors independently associated with concentrations of desmosines are shown in table 3. Log U-desmosines were associated with age, sex, height (females only), smoking (males only) and BMI (females only). Log P-desmosines were associated with age, sex and BMI. The relationship between

TABLE 2 Concentrations of desmosines in relation to baseline characteristics of the study cohort

	U-desmosines nmol·mmol^{-1} creatinine		P-desmosines nmol·L^{-1}	
	Subjects n	Median (range)	Subjects n	Median (range)
Sex				
Males	128	2.2 (1.3–6.0)	122	0.47 (0.16–1.3)
Females	221	2.8 (1.5–5.7)	196	0.48 (0.28–1.4)
Age yrs				
46–55	104	2.2 (1.4–4.1)	98	0.41 (0.16–0.76)
55–59	83	2.4 (1.3–4.2)	75	0.43 (0.28–0.87)
60–64	77	2.8 (1.8–5.2)	72	0.51 (0.32–1.3)
65–69	41	3.0 (1.7–6.0)	34	0.58 (0.37–1.1)
70–87	44	3.7 (2.3–5.7)	39	0.71 (0.41–1.4)
Smoking habit				
Current smokers	69	2.7 (1.5–6.0)	61	0.51 (0.16–1.3)
Occasional smokers	20	2.5 (1.5–4.0)	20	0.47 (0.31–0.68)
Ex-smokers	146	2.6 (1.4–5.2)	138	0.48 (0.28–1.2)
Never-smokers	114	2.5 (1.3–5.7)	99	0.46 (0.28–1.4)
BMI kg·m^{-2}				
<25	187	2.5 (1.4–6.0)	170	0.46 (0.16–1.3)
25–30	131	2.7 (1.3–5.7)	120	0.48 (0.28–1.4)
⩾30	31	2.9 (1.9–5.0)	28	0.53 (0.37–1.3)
Disease severity#				
No disease	234	2.5 (1.3–5.7)	212	0.46 (0.16–1.4)
GOLD I	67	2.6 (1.5–5.0)	61	0.49 (0.30–1.3)
GOLD II	43	2.9 (1.7–6.0)	40	0.55 (0.33–1.2)
GOLD III	5	2.8 (2.0–4.1)	5	0.64 (0.47–1.1)

BMI: body mass index; GOLD: Global Initiative for Chronic Obstructive Lung Disease. #: defined in [16] as GOLD I (mild), forced expiratory volume in 1 s (FEV_1) ⩾80% predicted; GOLD II (moderate), FEV_1 <80 and ⩾50% pred; GOLD III (severe) FEV_1 <50 and ⩾30% pred; GOLD IV (very severe), FEV_1 <30% pred.

TABLE 3 Multivariate analysis of factors associated with desmosines measures

Dependent variable	Males				Females				All			
	Ln U-desmosines[#]		Ln P-desmosines[¶]		Ln U-desmosines[+]		Ln P-desmosines[§]		Ln U-desmosines[f]		Ln P-desmosines[##]	
	β	p-value	β	p-value	β	p-value	β	p-value	β	p-value	β	p-value
Age (per 1 yr)	0.023	<0.001	0.023	<0.001	0.018	<0.001	0.022	<0.001	0.021	<0.001	0.022	<0.001
Sex (*versus* males)									0.21	<0.001	0.067	0.014
Height (per 1 cm)	-0.001	0.83	0.003	0.44	-0.006	0.02	0.00	0.92				
Current smoking (*versus* nonsmokers)	0.210	<0.001	0.089	0.09	0.002	0.94	0.017	0.64	0.092	<0.001	0.054	0.08
BMI (per 1 kg·m^{-2})	-0.003	0.72	0.008	0.039	0.014	<0.001	0.020	<0.001	0.011	<0.001	0.017	<0.001

All independent variables were simultaneously entered into the multiple linear regression model. 349 individuals were available for the analysis of U-desmosines and 318 individuals for the analysis of P-desmosines. BMI: body mass index. [#]: n=128; [¶]: n=122; [+]: n=221; [§]: n=196; [f]: n=359; [##]: n=318.

log P-desmosines and smoking was not significant in the multivariate analysis (p=0.08).

Desmosines and lung function

Mean±SD FEV1 was 93.4±17.3% pred in males and 99.8±6.5% pred in females. The proportion of subjects with COPD was 36% in males and 31% in females. The median levels of U-desmosines and P-desmosines were higher in subjects with GOLD stage I–III disease, compared with those without COPD (table 2). After adjustments for age and sex in a general linear model, the log U-desmosines values were significantly higher in GOLD stage II patients, compared with those without COPD (p=0.003). There was also a significant difference in log P-desmosine levels between GOLD stage III patients and those without COPD, after adjustments for age and sex (p=0.032). In terms of geometric mean values, the age- and sex-adjusted U-desmosines values were 2.63, 2.49, 2.94 and 2.73 nmol·mmol^{-1} creatinine, respectively, for no COPD, GOLD I, GOLD II and GOLD III. The corresponding values of P-desmosines were 0.48, 0.48, 0.52 and 0.61 nmol·L^{-1}.

The relationships between FEV1 % pred, RV/TLC and $D_{L,CO}$ and log U-desmosines are presented in figure 1. After adjustments for age, sex, height, BMI and smoking, log U-desmosines were significantly and inversely associated with FEV1, FVC and $D_{L,CO}$, and positively associated with RV and RV/TLC (table 4). The relationships between U-desmosines and lung function variables were generally stronger for males than for females. Log P-desmosines were significantly associated with FEV1 and $D_{L,CO}$.

Results of the analysis to compare subjects with and without COPD are shown in table 5. With the exception of RV and P-desmosines, relationships were markedly stronger in subjects with COPD after adjustment for age, sex, height, BMI and smoking. All measures of lung function were significantly associated with U-desmosines in this group. When the analysis was restricted to females with COPD, all lung function measures, except for RV, were significantly associated with U-desmosines, after adjustment for age, height, BMI and smoking (data not shown). Except for a significant inverse relationship between $D_{L,CO}$ and P-desmosines, there were no significant relationships in subjects without COPD.

Comparisons within twin pairs of the same sex are provided in the online supplementary material.

DISCUSSION

In this study, we investigated the association between U- and P-desmosines and different lung function measures (FEV1, FVC, RV, RV/TLC and $D_{L,CO}$). The correlations with lung function parameters were much more pronounced in COPD subjects than in those without COPD.

Previous studies on the correlation between desmosines and lung function have not demonstrated consistent results. For example, GOTTLIEB *et al*. [19] studied apparently healthy, smoking, adult males and found significantly higher U-desmosine excretion in rapid FEV1 decliners than in slow decliners, and a significant correlation between desmosine excretion and the rate of FEV1 decline over 6.3 yrs. In contrast, BOUTIN *et al*. [20] found significantly lower levels of U-desmosine in COPD patients with rapid FEV1 decline compared with slow decliners over 15 yrs.

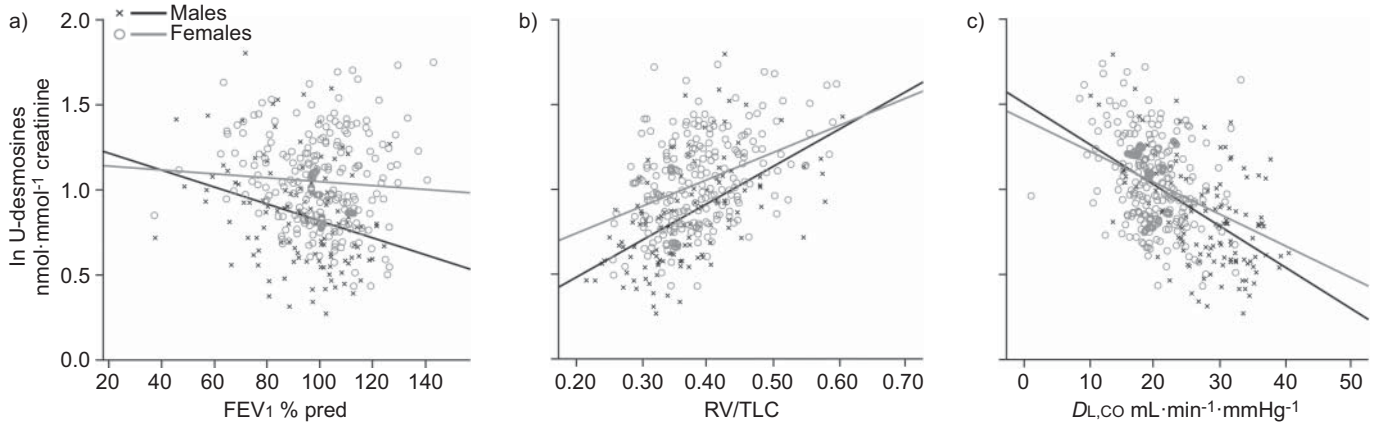

FIGURE 1. Scatterplot with regression slopes of log U-desmosines in relation to a) forced expiratory volume in 1 s (FEV1) % predicted (% pred) (r^2=0.08 for males and r^2=0.005 for females), b) residual volume (RV)/total lung capacity (TLC) (r^2=0.29 for males and r^2=0.16 for females), and c) diffusing capacity of the lung for carbon monoxide ($D_{L,CO}$) (r^2=0.26 for males and r^2=0.11 for females).

Another study by VIGLIO *et al*. [21] showed a correlation between U-desmosine and FEV1 % pred in a group of patients with destructive lung disease (cystic fibrosis, bronchiectasis, COPD and α_1-antitrypsin deficiency), but MA *et al*. [9] could not demonstrate any correlation between desmosines and various lung function parameters. Explanations for the different results may be the different methods used, the small sample sizes (usually <20 subjects) and varying correction for other, potentially confounding, factors. Another explanation for inconsistent results between studies could be that the relationship between desmosines and lung function may be different in different populations depending on disease status and severity. This is supported by our finding that the correlation with FEV1 was only seen in the subjects fulfilling the COPD criteria.

Adjusted for age and sex, 1 standard deviation of U-desmosine (0.86 nmol·mmol^{-1} creatinine) corresponded to ~3% units lower FEV1 % pred and about 1 mL·min^{-1}·mmHg^{-1} lower $D_{L,CO}$; the relationships were even stronger among males. There was a significant association between U-desmosines and several lung function parameters (FEV1, FVC, $D_{L,CO}$, RV and RV/TLC) after adjustment for age, sex, height, BMI and smoking status, while P-desmosines were significantly associated only with FEV1 and $D_{L,CO}$. The finding that the correlations with lung function are stronger for U-desmosines compared with P-desmosines are not completely consistent with the data from MA *et al*. [9], which indicate that P-desmosine gives a better separation between COPD patients and healthy subjects. In the current study, the relationships with lung function were mainly seen in subjects with COPD. In this context, it is interesting to note that the pathophysiology of COPD involves neutrophil-driven inflammation and lung matrix degradation [22]. Desmosines in healthy subjects may be more strongly determined by the normal, age-related elastin degradation from all elastin-containing organs. Elastin is not only a major component of the lungs but also serves an important function in arteries, as a medium for pressure wave propagation to help blood flow, and is particularly abundant in large elastic blood vessels, such as the aorta. Elastin is also important in other tissues, such as elastic ligaments, the skin

TABLE 4 Standardised β-coefficients for correlations between concentrations of desmosines and lung function measures

	Males				Females				All			
	Ln U-desmosines[#]		Ln P-desmosines[¶]		Ln U-desmosines[+]		Ln P-desmosines[§]		Ln U-desmosines[ƒ]		Ln P-desmosines[##]	
	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2
FEV1	-0.29**	-0.20*	-0.17	-0.12	-0.12	-0.11	-0.11	-0.11	-0.17***	-0.14**	-0.11*	-0.10*
FEV1 % pred	-0.37***	-0.26*	-0.22*	-0.16	-0.13	-0.12	-0.10	-0.10	-0.23***	-0.18**	-0.14*	-0.13*
FVC	-0.27**	-0.22*	-0.06	-0.03	-0.19**	-0.14*	-0.20**	-0.16*	-0.17***	-0.15***	-0.09*	-0.07
FVC % pred	-0.34**	-0.28*	-0.08	-0.03	-0.24**	-0.18*	-0.24**	-0.19[¶¶]	-0.27***	-0.23***	-0.16*	-0.12
RV	0.26*	0.16	0.06	0.03	0.03	0.08	-0.02	-0.002	0.14*	0.14*	0.02	0.03
RV/TLC	0.34***	0.24*	0.09	0.05	0.15*	0.15*	0.11	0.10	0.24***	0.22***	0.10	0.09
$D_{L,CO}$	-0.33***	-0.14	-0.35***	-0.30***	-0.01	-0.06	-0.09	-0.17*	-0.14**	-0.13*	-0.18***	-0.21***

Multiple linear regression with the lung function variable as dependent variable. Model 1: adjusted for age, sex (all subjects only) and height. Model 2: + body mass index and current smoking. FEV1: forced expiratory volume in 1 s; % pred: % predicted; FVC: forced vital capacity; RV: residual volume; TLC: total lung capacity; $D_{L,CO}$: diffusing capacity of the lung for carbon monoxide. [#]: n=128; [¶]: n=122; [+]: n=221; [§]: n=196; [ƒ]: n=349; [##]: n=318; [¶¶]: p=0.052; *: p<0.05; **: p<0.01; ***: p<0.001.

TABLE 5 Standardised β-coefficients for correlations between concentrations of desmosines and lung function measures in subjects with and without chronic obstructive pulmonary disease (COPD)

	No COPD[#]		COPD[¶]	
	U-desmosines	P-desmosines	U-desmosines	P-desmosines
M/F n	82/152	78/134	46/69	44/62
FEV_1	-0.02	0.02	-0.32***	-0.22*
FEV_1 % pred	-0.03	0.06	-0.42***	-0.29*
FVC	-0.05	0.03	-0.29***	-0.21*
FVC % pred	-0.08	0.03	-0.41***	-0.29*
RV	0.07	0.01	0.23*	0.01
RV/TLC	0.10	0.00	0.40***	0.18
$D_{L,CO}$	-0.02	-0.12*	-0.25**	-0.29**

Data adjusted for age, sex, height, body mass index and current smoking. Multiple linear regression was used with the lung function variable as the dependent variable. M: males; F: females; FEV_1: forced expiratory volume in 1 s; % pred: % predicted; FVC: forced vital capacity; RV: residual volume; TLC: total lung capacity; $D_{L,CO}$: diffusing capacity of the lung for carbon monoxide. [#]: $FEV_1/FVC \geqslant 0.70$; [¶]: $FEV_1/FVC < 0.70$. *: $p<0.05$; **: $p<0.01$; ***: $p<0.001$.

and the bladder. Therefore, it is possible that degradation from tissues other than the lung may have contributed to the associations seen in this study. However, since a number of different tissues are affected in COPD, this may also be relevant to the pathophysiology of the disease. It is difficult to assess how much lung elastin constitutes the total amount in the body; estimates vary from 1.3% to 24% [23–25]. It has also been estimated that the normal rate of lung elastin turnover accounts for only ~19% of the desmosines excreted in urine [12].

The present study also demonstrated a strong correlation between age and either U- or P-desmosines. This is consistent with the finding that increased elastolysis and degradation of elastin are features of normal ageing [26] and the concept of ''senile emphysema'', normal physiological ageing of the lung associated with dilatation of alveoli, enlargement of airspaces and loss of supporting tissue for peripheral airways [27]. It should be noted, however, that previous studies have only shown a correlation between age and U-desmosine in the presence of another risk factor. STONE *et al.* [12] reported a positive correlation between age and U-desmosine excretion (creatinine normalised) in a small group of current smokers (n=13) but they found no significant correlation in a group of never-smokers (n=22). A strong correlation was also found between U-desmosines and age in patients with pseudoxanthoma elasticum but not in healthy subjects [28].

Smoking was significantly associated with levels of U-desmosines ($p<0.001$) and nonsignificantly ($p=0.08$) with P-desmosines, after adjustment for age, sex and BMI. Interestingly, no significant association with smoking was observed in the unadjusted analysis; however, smokers were somewhat younger than nonsmokers and age was a very strong determinant of desmosine levels. The results illustrate the importance of adequately correcting for other risk factors, especially age, in studies of desmosines. U- and P-desmosines were also significantly associated with sex, with higher concentrations in females than in males. Furthermore, females showed weaker correlations between desmosines and lung function than males. However, it should be noted that FEV_1 was lower in males than in females (mean ± SD FEV_1 93.4 ± 17.3 *versus* 99.8 ± 6.5% pred), and many females had relatively high FEV_1. As the relationship between desmosines and lung function was observed mainly in subjects with COPD, the stronger correlation in males seems to be related to degree of lung function impairment rather than sex differences. BMI also showed association with desmosines in this study. Potential explanations for this association may be either increased mass of elastic tissues such as skin and blood vessels, or increased systemic inflammation in subjects with high BMI.

One of the limitations of this study was the generally mild level of COPD in study subjects (according to the current GOLD classification) [18]. Relationships between lung function and desmosines may be stronger in a more severe cohort. The use of twins for the study may also have influenced the outcome, as the selection procedure may have resulted in an over-representation of certain groups or because genetic factors may influence the relationships studied. The results therefore need to be confirmed in additional COPD cohorts with varying degrees of disease severity. Furthermore, longitudinal studies are needed to address whether an increase in desmosine is associated with a decrease in lung function and other measures of disease progression, as has been suggested [13]. In addition, although it is one of the most accurate analytical techniques available, MS/MS requires relatively expensive instrumentation and skilled personnel, which may limit a wider distribution of the method.

In summary, this study, which, to our knowledge, is the largest study on desmosines and lung function, demonstrates that both U- and P-desmosines are correlated with a number of lung function measurements in subjects with COPD. The finding that desmosines can be independently influenced by a number of factors other than lung function (age, sex, height, smoking and BMI) emphasises the need to correct for these factors to avoid confounding results.

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STATEMENT OF INTEREST

Statements of interest for all authors and for the study itself can be found at www.erj.ersjournals.com/site/misc/statements.xhtml

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