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Carboxyhaemoglobin level, smoking habit and mortality in 25 years in the Renfrew/Paisley prospective cohort study

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Abstract

Objective To investigate how carboxyhaemoglobin level was related to smoking habit and to assess whether carboxyhaemoglobin level was related to mortality.

Design Prospective cohort study.

Setting Residents of the towns of Renfrew and Paisley in Scotland.

Participants The whole Renfrew/Paisley study, conducted between 1972 and 1976, consisted of 7048 men and 8354 women aged 45-64 years. This study was based on 3372 men and 4192 women who were screened after the measurement of carboxyhaemoglobin level was introduced about half way through the study.

Main outcome measures Deaths from coronary heart disease (CHD), stroke, chronic obstructive pulmonary disease (COPD), lung cancer and all causes in 25 years after screening.

Results Carboxyhaemoglobin level was related to self-reported smoking and was higher in participants who reported inhaling compared with those who reported not inhaling, for each smoking category. There were positive relationships between carboxyhaemoglobin level and all causes of mortality analysed (relative rate associated with 1 SD (2.93) increase in carboxyhaemoglobin for all causes, CHD, stroke, COPD and lung cancer were 1.26 (1.19-1.34), 1.19 (1.13-1.26), 1.19 (1.13-1.26), 1.64 (1.47-1.84) and 1.69 (1.60-1.79) respectively). Adjustment for self-reported cigarette smoking attenuated the associations but they remained relatively strong.

Conclusions The self-reported smoking data was validated by the objective measure of carboxyhaemoglobin level. Since carboxyhaemoglobin level remained associated with mortality after adjusting for smoking, carboxyhaemoglobin seems to capture more of the "risk" associated with smoking tobacco than self-reported tobacco consumption alone. Analysing mortality by self-reported cigarette smoking will underestimate the strength of association between smoking and mortality.

Abstract word count : 250 words

Introduction

Epidemiological studies often suffer from under-reporting of habits known to be unhealthy or undesirable. In particular, the validity of self-reported smoking has been questioned. Studies have used physical measures such as cotinine[1][2] (in blood, urine or saliva) or carboxyhaemoglobin[3][4] to validate self-reported smoking habit. The Second National Health and Nutrition Examination Survey (NHANES II) used carboxyhaemoglobin measurements to investigate bias towards reporting multiples of 10 cigarettes per day.[4] The BUPA study found positive relationships between carboxyhaemoglobin and amount smoked and amount inhaled.[5] In this study, carboxyhaemoglobin is used to investigate the relationship with smoking habits, and to see how carboxyhaemoglobin level is related to mortality from various diseases in the 25 years following screening.

Methods

The Renfrew/Paisley study was a general population prospective cohort study, conducted between 1972 and 1976 on residents of the towns of Renfrew and Paisley in Scotland.[6] These towns are located near to the city of Glasgow in Scotland and are areas of high socioeconomic deprivation with life expectancy figures amongst the worst in the whole of the United Kingdom (http://www.statistics.gov.uk/statbase/Product.asp?vlnk=8841). Men and women aged between 45 and 64 years were invited to take part and the response rate of 78% resulted in 7048 male and 8354 female participants.

Participants completed a questionnaire and attended a clinical examination at specially set up screening centres. The questionnaire included questions on smoking habit, from which smoking categories were derived. These were: never smoked, smoked only cigars or a pipe, smoked cigarettes (1-5 per day, 6-14, 15-24 or 25 or more) or ex-smokers. Ex-smokers were defined as not having smoked for at least 12 months before screening, otherwise they were categorised as current smokers. Current smokers were also asked if they inhaled. At the screening examination, height was measured without shoes and the forced expiratory volume in one second (FEV1) was measured with a vitalograph spirometer with the subject standing.[7] Predicted FEV1 was calculated from equations derived from a healthy subset of the population for age, height and sex.[7] Adjusted FEV1 was calculated as a percentage of actual FEV1 to predicted FEV1. In 1975, about half way through the Renfrew/Paisley study, co-oximeter measurements of carboxyhaemoglobin level in the participants' blood were introduced.

Mortality details are received routinely via the flagging system at the National Health Service Central Register in Edinburgh, the normal method for UK cohort studies. Additionally deaths were obtained from a computerised linkage with deaths in Scotland. For this study, deaths occurring in the 25 year period after screening were defined as coronary heart disease (CHD) (ICD9 codes 410-414), stroke (codes 430-438), chronic obstructive pulmonary disease (COPD) (codes 490-494 and 496) and lung cancer (code 162). Deaths from all causes are also reported.

There were 7809 participants screened after the carboxyhaemoglobin measurement was introduced. Of these, 229 had missing values of carboxyhaemoglobin and there was one obvious outlier. Also excluded from this study were 9 participants with missing data on adjusted FEV1 and 6 participants who had embarked from the UK during the follow up period. The analysis was therefore performed on 3372 men and 4192 women (total 7564) with complete data. Trends of carboxyhaemoglobin level by amount smoked were calculated by regression analysis for never and current cigarette smokers using number of cigarettes smoked per day. Differences between mean carboxyhaemoglobin level by inhalation were

calculated using t tests. Means of adjusted FEV1 by quintile of carboxyhaemoglobin were additionally adjusted for smoking using PROC GLM in SAS.[8] The smoking adjustment was entered as the number of cigarettes smoked daily by current cigarette smokers, with zero allocated to non and former cigarette smokers. Trends for adjusted FEV1 with carboxyhaemoglobin were calculated by linear regression analysis. Cox's proportional hazards models were used to estimate relative rates of mortality by quintile of carboxyhaemoglobin and to calculate the relative rate associated with one standard deviation increase in carboxyhaemoglobin level with carboxyhaemoglobin as a continuous variable. Adjustments were made for smoking by adding the number of cigarettes per day for current and ex-smokers, with an additional variable for ex-smokers (1 if ex-smoker, 0 otherwise). Cox's models were also used for estimating the relative rates of mortality by smoking category, with adjustments made for carboxyhaemoglobin level.

Results

Table 1 presents the mean carboxyhaemoglobin level for each smoking category. Never smokers had the lowest carboxyhaemoglobin level followed by ex-smokers. There was a dose response relationship seen with number of cigarettes smoked per day, with the pipe or cigar smokers having a mean carboxyhaemoglobin level between the 1-5 and the 6-14 cigarettes per day categories. Results were consistent for men and women.

Table 1 Mean carboxyhaemoglobin percent by smoking habit in Renfrew/Paisley men and women

	All			Men			Women		
Smoking	N	Mean	SD	N	Mean	SD	N	Mean	SD
Never	2448	1.59	1.72	547	1.77	1.72	1901	1.53	1.71
Ex smoker	1206	1.96	1.87	876	2.09	1.94	330	1.62	1.64
Pipe / cigar	91	2.63	2.62	86	2.52	2.60	5	4.42	2.53
1-5	162	2.31	1.94	36	2.19	1.58	126	2.34	2.03
6-14	1002	4.39	2.48	368	4.13	2.30	634	4.54	2.57
15-24	1945	5.68	2.64	921	5.48	2.63	1024	5.86	2.64
25+	710	6.02	2.86	538	5.95	2.84	172	6.23	2.90
All	7564	3.52*	2.93	3372	3.81*	2.90	4192	3.27*	2.93

*$p<0.0001$ for never and current cigarette smokers

The majority of cigarette smokers reported inhaling (93% of men and 82% of women). For each smoking category and for each sex, inhalers had higher carboxyhaemoglobin levels than non-inhalers (table 2). Adjusting for number of cigarettes smoked did not affect the difference in carboxyhaemoglobin level between inhalers and non-inhalers (not shown). Overall, in current smokers, inhalers had a mean adjusted carboxyhaemoglobin level of 5.63 compared with 4.40 in non-inhalers ($p<0.0001$).

Table 2 Mean carboxyhaemoglobin percent for current cigarette smokers by inhalation with P value for difference in means (1709 men and 1863 women)

	All			Men			Women		
Smoking	N	Mean	SD	N	Mean	SD	N	Mean	SD
1-5									
Inhale	87	2.46	2.10	26	2.43	1.69	61	2.48	2.26
Not inhale	67	2.07	1.65	9	1.64	1.15	58	2.13	1.71
P value		0.20			0.21			0.35	
6-14									
Inhale	767	4.67	2.42	303	4.40	2.28	464	4.84	2.49
Not inhale	180	3.96	2.46	38	3.30	1.77	142	4.13	2.59
P value		<0.0001			0.004			0.004	
15-24									
Inhale	1670	5.98	2.53	815	5.75	2.53	855	6.21	2.51
Not inhale	174	4.56	2.39	48	4.38	2.34	126	4.63	2.41
P value		<0.0001			<0.0001			<0.0001	
25+									
Inhale	583	6.63	2.58	445	6.59	2.53	138	6.77	2.74
Not inhale	44	5.25	2.20	25	5.0	2.30	19	5.56	2.08
P value		0.001			0.002			0.07	

Carboxyhaemoglobin was strongly related to adjusted FEV1 in men and women (table 3). Participants with lower carboxyhaemoglobin levels had higher adjusted FEV1 and there were significant trends even after adjustment for cigarette smoking.

Table 3 Mean adjusted FEV1 percent by quintile of carboxyhaemoglobin in Renfrew/Paisley men and women, unadjusted and adjusted for smoking

Quintile	All			Men			Women		
COHb	N	Mean*	Mean†	N	Mean*	Mean†	N	Mean*	Mean†
1 (0.1-0.8)	1512	94.3	92.2	533	92.8	90.3	979	95.2	93.6
2 (0.9-2.0)	1622	93.8	92.1	630	91.8	89.9	992	95.1	93.7
3 (2.1-3.7)	1454	90.0	89.5	729	89.0	88.1	725	91.1	90.8
4 (3.8-6.1)	1466	86.7	88.5	738	84.6	86.2	728	88.8	90.6
5 (6.2-24.2)	1510	84.4	87.1	742	84.2	86.8	768	84.6	87.0
Trend		p<0.0001	p<0.0001		p<0.0001	p=0.007		p<0.0001	p<0.0001

COHb is carboxyhaemoglobin
* unadjusted
† adjusted for cigarettes smoked per day

Carboxyhaemoglobin level was positively related to all cause mortality, adjusting for age and sex (table 4). Adjustment for smoking considerably attenuated the associations but they remained relatively strong. There were similar relationships with CHD mortality. A less clear relationship was seen between carboxyhaemoglobin and stroke and this was attenuated after adjustment for smoking. Strong relationships were seen with COPD mortality, including after adjustment for smoking, although smaller numbers produced large confidence intervals. Carboxyhaemoglobin level was strongly related to lung cancer mortality, even after adjustment for smoking. Tests for interactions between sex and carboxyhaemoglobin level were not significant for any of the causes of death analysed (all cause p=0.10, CHD p=0.08, stroke p=0.57, COPD p=0.09 and lung cancer p=0.30). When analyses were performed by smoking category, the positive relationships with mortality were similarly attenuated when adjusted for carboxyhaemoglobin level (not shown).

Table 4 Relative rates (and 95% confidence intervals) of all cause, CHD, stroke, COPD and lung cancer mortality over 25 years by quintile of carboxyhaemoglobin and per standard deviation increase in carboxyhaemoglobin in Renfrew/Paisley men and women

	COHb Quintile (range)	No of deaths	Model 1*	Model 2†
All causes	1 (0.1-0.8)	686	1	1
	2 (0.9-2.0)	727	0.97 (0.88 – 1.08)	0.95 (0.85 – 1.05)
	3 (2.1-3.7)	764	1.22 (1.10 – 1.35)	1.13 (1.01 – 1.25)
	4 (3.8-6.1)	863	1.54 (1.39 – 1.71)	1.28 (1.15 – 1.43)
	5 (6.2-24.2)	942	1.77 (1.60 – 1.95)	1.41 (1.26 – 1.57)
RR associated with 1 SD increase in carboxyhaemoglobin			1.26 (1.19 – 1.34)	1.16 (1.09 – 1.23)
CHD	1 (0.1-0.8)	213	1	1
	2 (0.9-2.0)	247	1.05 (0.87 – 1.26)	1.03 (0.86 – 1.24)
	3 (2.1-3.7)	276	1.35 (1.13 – 1.62)	1.28 (1.06 – 1.53)
	4 (3.8-6.1)	253	1.37 (1.14 – 1.65)	1.20 (0.98 – 1.46)
	5 (6.2-24.2)	280	1.59 (1.33 – 1.90)	1.34 (1.09 – 1.64)
RR associated with 1 SD increase in carboxyhaemoglobin			1.19 (1.13 – 1.26)	1.12 (1.06 – 1.19)
Stroke	1 (0.1-0.8)	106	1	1
	2 (0.9-2.0)	86	0.76 (0.57 – 1.01)	0.74 (0.56 – 0.99)
	3 (2.1-3.7)	100	1.13 (0.86 – 1.49)	1.05 (0.79 – 1.38)
	4 (3.8-6.1)	107	1.41 (1.08 – 1.85)	1.18 (0.88 – 1.59)
	5 (6.2-24.2)	89	1.28 (0.96 – 1.70)	1.03 (0.74 – 1.42)
RR associated with 1 SD increase in carboxyhaemoglobin			1.19 (1.13 – 1.26)	1.09 (0.97 – 1.22)
COPD	1 (0.1-0.8)	12	1	1
	2 (0.9-2.0)	16	1.22 (0.58 – 2.57)	1.15 (0.54 – 2.43)
	3 (2.1-3.7)	24	2.16 (1.08 – 4.33)	1.90 (0.94 – 3.84)
	4 (3.8-6.1)	50	5.10 (2.71 – 9.60)	3.94 (2.03 – 7.66)
	5 (6.2-24.2)	57	6.17 (3.30 –11.54)	4.42 (2.23 – 8.67)
RR associated with 1 SD increase in carboxyhaemoglobin			1.64 (1.47 – 1.84)	1.46 (1.23 – 1.74)
Lung cancer	1 (0.1-0.8)	34	1	1
	2 (0.9-2.0)	26	0.69 (0.41 – 1.15)	0.64 (0.38 – 1.06)
	3 (2.1-3.7)	61	1.82 (1.20 – 2.77)	1.43 (0.93 – 2.19)
	4 (3.8-6.1)	109	3.57 (2.43 – 5.26)	2.14 (1.43 – 3.23)
	5 (6.2-24.2)	159	5.41 (3.73 – 7.85)	2.89 (1.92 – 4.34)
RR associated with 1 SD increase in carboxyhaemoglobin			1.69 (1.60 – 1.79)	1.42 (1.27 – 1.59)

COHb is carboxyhaemoglobin
1 SD=2.93%
* Model 1 adjusts for age and sex
† Model 2 adjusts for age, sex and smoking

Discussion

This analysis has validated the self-reported smoking habits in the Renfrew/Paisley cohort by an objective measure. Carboxyhaemoglobin levels were strongly related to self-reported smoking levels, self-reported inhalation and respiratory function, and in addition were related to various causes of death, even after adjustment for smoking.

Since carboxyhaemoglobin level is a physical measurement, it can be considered a proxy for risk of disease caused by smoking. In a study of about 1000 Swedish middle-aged men, morning carboxyhaemoglobin levels were measured.[9] Levels were low for both non-smokers and ex-smokers. Higher levels were reported in current smokers and there was a dose response relationship as normal daily consumption increased. A large variation in carboxyhaemoglobin level was seen in people who reported smoking the same amount daily. It was suggested that prospective studies were required to see if carboxyhaemoglobin measurement were a better measurement than tobacco consumption by questionnaire.

NHANES II found bias towards reporting daily amount smoked in multiples of 10 cigarettes per day which could have been systematic (ie rounding down) or random.[4] There was no similar pattern seen for carboxyhaemoglobin level. It was suggested that biochemical verification and self-reported smoking level could be combined when examining smoking-disease relationships. It was also suggested that, since there were errors in self-reported smoking, reported relationships between smoking and disease may not have been accurate.

In addition to finding positive relationships between carboxyhaemoglobin and amount smoked, the BUPA study found carboxyhaemoglobin was related to the risk of mortality from CHD, lung cancer or chronic obstructive lung disease (defined as ICD9 codes 416, 491, 492, 496 and 519) independently of smoking category or amount smoked.[5] Analysis by smoking after adjustment for carboxyhaemoglobin removed the smoking-disease relationship. In the current study, carboxyhaemoglobin level remained associated with mortality after adjusting for smoking, suggesting that carboxyhaemoglobin is capturing more smoking-related “risk” than just self-reported smoking. Examples could be unmeasured differences in the way the cigarettes were smoked, such as number of puffs per cigarette, smoking right down to the butt, or depth of inhalation. The carboxyhaemoglobin level could also have included any passive smoking which could have had an increased effect on mortality risk.

Experiments in smokers showed that carboxyhaemoglobin level did not increase during the day as more cigarettes were smoked, but remained around each individual’s mean value.[10] Implications for the current study are that time of day of screening or time since the last cigarette was smoked would not affect the carboxyhaemoglobin level measured. The strong relationship between carboxyhaemoglobin and lung function was to be expected, since carbon monoxide is removed from the body by expiration and expiration is less efficient in participants with poorer lung function.[11]

It is known that cigarette smokers who report not inhaling still have raised carboxyhaemoglobin levels.[3] However these levels are lower than for reported inhalers. In the current study, from a validation viewpoint, carboxyhaemoglobin levels were consistently lower in non-inhalers than in inhalers. Further studies of passive smoking in this cohort will be able to use carboxyhaemoglobin level to exclude participants who may have misreported their smoking habits.[12]

This research suggests that the smoking-mortality risk may have previously been underestimated due to misreporting of cigarette consumption. Carboxyhaemoglobin level could be picking up unmeasured risk in several ways. Firstly participants could have under-reported their tobacco consumption, through rounding their response to a pack size or giving

an incorrect reply because they know that smoking is harmful. Secondly, carboxyhaemoglobin level could be a better measure of the harmful effects of cigarettes than just number smoked, since it is capturing additional unmeasured effects such as depth of inhalation and how much of each cigarette is smoked. In consequence the proportion of mortality in a population attributed to smoking will be underestimated through applying the associations observed between reported smoking in mortality in prospective epidemiological studies.

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Competing interest statement
None.

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