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A twin study of genetic and environmental influences on tobacco initiation, regular tobacco use and nicotine dependence

HERMINE H. MAES*, PATRICK F. SULLIVAN, CYNTHIA M. BULIK, MICHAEL C. NEALE, CAROL A. PRESCOTT, LINDON J. EAVES AND KENNETH S. KENDLER

Departments of Human Genetics and Psychiatry, Virginia Institute for Psychiatric and Behavioral Genetics, Virginia Commonwealth University, Richmond, VA, USA; Massey Cancer Center, Virginia Commonwealth University, Richmond, VA, USA; Departments of Genetics and Psychiatry, University of North Carolina, Chapel Hill, NC, USA

ABSTRACT

Background. Numerous twin studies have reported significant genetic contributions to the variability of tobacco initiation (TI), while fewer studies have shown similar results for the persistence of smoking behavior, or nicotine dependence (ND). As the development of ND requires regular tobacco use (RTU) which in turn requires TI, a conditional approach is necessary.

Method. We used structural equation modeling of multi-step conditional processes to examine the relationship between genetic and environmental risk factors for TI, RTU and ND. The tobacco variables were assessed by personal interview in female, male and opposite-sex twin pairs from the population-based Virginia Twin Registry.

Results. The results suggested that the liabilities to TI, RTU and ND were correlated. Over 80% of the variance in liability to TI and RTU were shared, and a smaller proportion was shared between RTU and ND. The heritabilities were estimated at 75%, 80% and 60% respectively for TI, RTU and ND. The variance specific to liability to RTU was entirely accounted for by additive genetic factors. Only a modest part of the heritability in liability of ND was due to genetic factors specific to ND. Shared environmental factors were not significant. No sex differences were found for the sources of variation or causal paths, but prevalences were significantly greater in males *versus* females.

Conclusions. This study showed significant overlap in the contribution of genetic factors to individual differences in TI, RTU and ND. Furthermore, there was evidence for significant additional genetic factors specific to RTU and ND.

INTRODUCTION

Smoking is a serious public health problem. Briefly, tobacco smoking is associated with increased morbidity, mortality, and personal and public cost (US Department of Health and Human Services, 1989; WHO, 1997). In the USA, cigarettes are responsible for 30% of all cancer deaths and 21% of deaths from cardiovascular disease (US Department of Health and Human Services, 1989). Half of those beginning to smoke in adolescence will die from a cigarette-related cause (WHO, 1997). Costs of medical care attributable to smoking in the USA were estimated to be $50 billion in 1993 and the true total may have approached $100 billion (Centers for Disease Control, 1994).

* Address for correspondence: Dr H. H. Maes, Virginia Institute for Psychiatric and Behavioral Genetics, PO Box 980003, Richmond, VA 23298-0003, USA.
(Email: hmaes@hsc.vcu.edu)

Although many individuals try smoking, the determinants of the trajectory from tobacco initiation (TI) to regular tobacco use (RTU), and then to nicotine dependence (ND) are not entirely clear. To better understand the determinants of ND, partitioning the variance into genetic and environmental factors is potentially useful. Given that ND is contingent upon TI, a conditional approach is necessary. The present analysis explores the correlations across the liabilities of TI, RTU and ND.

Twin studies have consistently shown a significant genetic component to the liability to smoking initiation. A recent review summarized the sample sizes, correlations and genetic and environmental variance components of these studies (Sullivan & Kendler, 1999). About 60% of the variance in liability to initiate smoking is accounted for by additive genetic factors. Shared environmental factors also account for a small but replicable proportion of the variation in smoking initiation (see also Sullivan & Kendler, 1999). Whether gender differences exist in the magnitude or nature of individual differences in smoking-related variables is not entirely clear. Studies that include both male and female participants mostly show a greater [Heath *et al.* 1993 (for the Virginia and AARP sample); Madden *et al.* 1999] or equal (Pedersen, 1981; Kaprio *et al.* 1984; Boomsma *et al.* 1994; Koopmans *et al.* 1999; Maes *et al.* 1999) contribution of the shared environment in females compared to males. One exception is an Australian study, described by Heath *et al.* (1993), in which about 40% of the variance of the liability to smoking initiation in males was explained by shared environmental factors, compared to 10% in females. However, estimates from a recent meta-analysis (Li *et al.* 2003) suggested a greater shared environmental component for male adults ($c^2 = 0{\cdot}49$ for males and 0·24 for females) and greater heritability for female adults ($h^2 = 0{\cdot}37$ for males and 0·55 for females).

The genetic epidemiology of ND has received relatively little attention. To our knowledge only two studies examined ND in a genetically informative population (Kendler *et al.* 1999; True *et al.* 1999). The sample for the Kendler *et al.* study was a subset of that of the current report. The results suggested an overlap between the etiological factors that influence smoking initiation and ND. True *et al.* (1999) estimated a heritability of 60% for ND without taking into account smoking initiation. Other studies have used rough proxy measures for ND, such as heavy smoking (Kaprio *et al.* 1982; Swan *et al.* 1997), cigarette consumption (Carmelli *et al.* 1990) or quantity smoked (Meyer *et al.* 1992; Swan *et al.* 1996; Koopmans *et al.* 1999). Meyer *et al.* (1992) used non-metric multidimensional scaling and found that the onset of the smoking habit and the quantity smoked represent a unidimensional process. In contrast, Koopmans *et al.* (1997) concluded that the dimensions of smoking initiation and quantity smoked were partly independent.

At least seven studies have investigated the relationship between smoking initiation and smoking persistence (Eaves & Eysenck, 1980; Hannah *et al.* 1985; Heath & Martin, 1993; Edwards *et al.* 1995; True *et al.* 1997; Madden *et al.* 1999; Heath *et al.* 2002) which may be viewed as a proxy for ND. Eaves & Eysenck (1980) were first to point out that although a unidimensional multi-factorial model was adequate to distinguish those who have never smoked from those who persist in smoking, a distinct dimension is required to account for both age of onset and average consumption. Most studies provide evidence for correlated liabilities to smoking initiation and smoking persistence, using structural equation models proposed by Heath (1990) and Neale (unpublished observations). However, Hannah *et al.* (1985) and Edwards *et al.* (1995) found evidence for independent genetic effects on smoking initiation and persistence using logistic regression.

In this report, we extend previous studies by examining the relationship among TI, RTU and ND simultaneously. This approach allows us not only to quantify the contributions of genetic and environmental factors specific to RTU and ND, but also those effects in common with TI. In addition, we test whether these sources of variation differ by gender in a large sample of adult twins from the Virginia Twin Registry.

METHOD

Subjects

Participants in the present investigation were drawn from two longitudinal studies conducted in a similar manner by the same research group.

Both investigations were reviewed by the ethical review boards and all participants provided written informed consent (or verbal consent for telephone interviews) prior to participation. Each sample was ascertained from the population-based Virginia Twin Registry. The first study was of female–female twin pairs (FF) and the second of male–male and male–female twin pairs (MMMF). These studies are described at length elsewhere (Kendler & Prescott, 1999). Zygosity determination was based on questionnaire responses and DNA polymorphisms where required (Spence *et al*. 1988).

FF study

The third interview wave of the FF study (1992–95) was the source of the majority of data in this report. Wave three data collection included telephone interviews with 1846 individuals (88% of the wave one sample) whose mean age was 35·1 years (S.D. = 7·5). As the TI item was queried in the fourth FF interview wave but not the third, it was necessary to 'bring backward' this item.

MMMF study

The data for this report are from the second interview wave of the MMMF study (1994–98). Of 9417 eligible individuals for the first wave, 6814 (72·4%) completed an interview. At least 1 year after the completion of the first-wave interview, we successfully completed a wave-two interview with 4959 individuals (82·6%) whose mean age was 37·0 (S.D. = 9·1) years.

Measures

The data analyzed here were collected as part of a 1–3-hour personal interview. The interviews for both the FF and MMMF studies were highly homologous. All interviews were conducted by individuals with Master's degrees in social work or psychology or a Bachelor's degree plus at least 2 years of clinical experience. All interviewers underwent rigorous training and all interviews were reviewed by a senior editor for consistency and accuracy. Interviewers were blinded to all prior data from the twin they interviewed as well as to data on the co-twin.

In the MMMF study, we assessed all common forms of tobacco self-administration (cigarettes, cigars, pipe tobacco, chewing tobacco, and snuff) whereas FF study participants were only asked about cigarettes. To equate these data, we were forced to assume that the 1846 FF study participants had an extremely low prevalence of non-cigarette forms of tobacco use. This assumption was strongly supported by data from the MMMF study in which none of 1195 women from opposite-sex twin pairs reported the use of non-cigarette forms of tobacco use.

For the purposes of this report, we focused on five tobacco-related variables. Tobacco initiation was defined according to the responses to the questions, 'Have you ever smoked cigarettes?' and the follow-up query 'Not even once?' Regular tobacco use was defined as the use of an average of at least seven cigarettes per week for a minimum of 4 weeks. Individuals who met criteria for RTU were given a modified version of the Fagerström Tolerance Questionnaire (FTQ). Nicotine dependence was considered present if an individual's score on the FTQ was seven or more during the period of lifetime maximal cigarette use (Fagerström, 1978; Fagerström & Schneider, 1989). In addition to the dichotomized ND variable, we performed the analyses on the FTQ score which ranges from zero to eleven, and the revised Fagerström Test for Nicotine Dependence (FTND) score ranging from zero to ten (Heatherton *et al*. 1991). As the FTND score is a revision of the FTQ score, has slightly better reliability and internal consistency (Pomerleau *et al*. 1994), and because the results were similar for the FTQ and FTND score, we limit the presentation of the results to the dichotomous ND diagnosis and symptoms of ND as defined by the FTND.

Scoring of non-cigarette tobacco use

Most of the FTQ items adapt readily to all forms of tobacco use. Two items required modification. First, it was necessary to translate use of cigars, pipes, smokeless tobacco into the equivalent number of cigarettes. One 'chaw' was considered equal to 1·5 cigarettes, one cigar to 2·3 cigarettes, one 'dip' to 2·0 cigarettes, and one pipeful to 2·1 cigarettes (Benowitz *et al*. 1988). Secondly, we assumed that smokeless tobacco users always 'inhale'.

Statistical analyses

Structural modeling of the data was undertaken which assesses the contributions of additive genetic effects in the presence of effects of shared

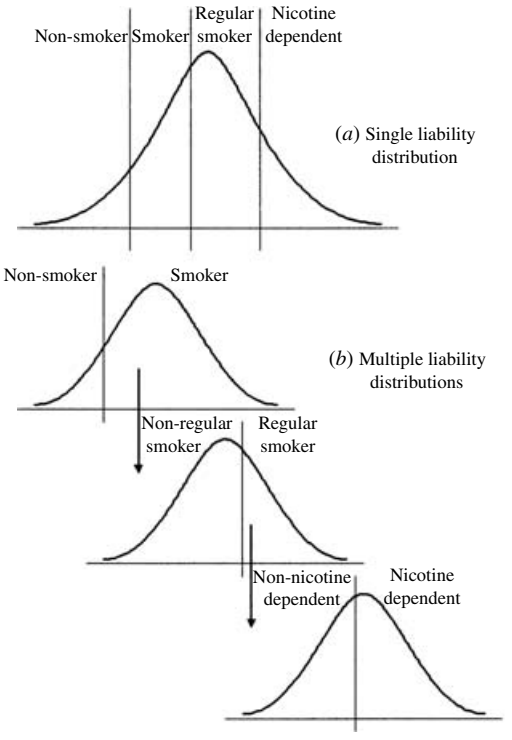

FIG. 1. (*a*) Single-liability distribution model and (*b*) multiple-liability distribution model for tobacco initiation, regular tobacco use and nicotine dependence.

and within-family environment. Between-family (or shared) environmental effects make family members relatively more similar, whereas within-family (or specific) environmental factors are unique to individuals within a family and contribute to differences between family members. The contribution of genetic and environmental factors may be dependent upon sex, both in their magnitude and nature. To partition the variance of the liability to RTU and ND, it is necessary to do a multivariate genetic analysis including TI, as RTU and ND can be assessed only in individuals who have initiated tobacco use.

We tested whether the liabilities to TI, RTU and ND were consistent with a single or multiple liability distributions (see Fig. 1). If the single-liability distribution (SLD) model is rejected, the liabilities to TI, RTU and ND could be independent (independent liability model, ILD) or correlated (correlated liability model, CLD). Given that ND is contingent on RTU and RTU is contingent on TI, it is necessary to fit a causal,

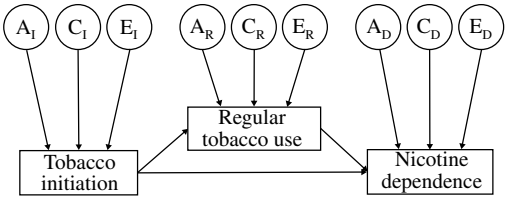

FIG. 2. Causal contingent common pathway (CCC) model for tobacco initiation, regular tobacco use and nicotine dependence.

contingent, common-pathway (CCC) model (see for example Kendler *et al*. 1999). However, if the first dimension can be divided into more than two categories, including at least two categories for which data are available on the second dimension, it is possible to fit a traditional Cholesky decomposition model to test the degree of overlap of genetic and environmental factors to TI and ND (Heath *et al*. 2002). [See Neale & Cardon (1992) and Neale *et al*. (unpublished observations) for a detailed description of these models and extensions to allow for sex limitation.]

Fig. 2 presents a path diagram of the CCC model for three variables, which is an extension of the two-variable model used in Kendler *et al*. (1999). In addition to being contingent, the model is also causal in that it assumes a direct path from the liability to TI to the liability to RTU and ND, which also implies that the genetic and environmental contributions to TI can only affect RTU and ND through the observed phenotype of TI (common pathway). The full model includes a causal path between TI and RTU, between RTU and ND and between TI and ND, as well as genetic, shared and specific environmental factors on each of the phenotypes. With the recent availability of ordinal raw data analysis in Mx (Neale *et al*. 2002), the analyses were performed on the original ordinal data, including individual twins whose co-twin did not cooperate.

RESULTS

Descriptive statistics

The twin sample contained 6805 individuals of whom 55·3% were male and 44·7% were female. The mean age of the sample was 36·2 years (S.D. = 8·6) with a range of 20·4–59·5 years. Overall, individuals in the sample reported: lifetime

TI 78·2%, lifetime RTU 54·2%, and lifetime ND 19·8%. Only individuals who were regular smokers were given the Fagerström Questionnaire, from which the FTQ score and FTND score were calculated. From a lifetime perspective, the cross-tabulation of TI, RTU and ND resulted in four possible outcomes: (1) never smokers, (2) non-regular smokers, (3) non-nicotine-dependent regular smokers (non-ND), and (4) nicotine-dependent regular smokers (ND). Gender clearly had a strong influence ($\chi^2 = 353$, $p < 0{\cdot}0001$). The distribution of the ND symptom scores (FTND) somewhat approached normality but also shows gender differences ($\chi^2 = 70$, $p < 0{\cdot}0001$ for FTND).

Univariate analysis of tobacco initiation

As previous genetic analyses of the smoking data were limited to females (Kendler *et al.* 1999), we first extended them to include male and opposite-sex twins and estimated the correlations by gender and zygosity for TI. Thresholds could not be equated across zygosity or gender without significant loss of fit. Univariate genetic analyses suggested a heritability of 73% for TI with 2% of the variance accounted for by shared environmental factors for the combined male and female sample. Although the contribution of shared environmental factors appeared larger in females than in males, the estimated variance components were not significantly different across gender and could be dropped without significant loss of fit.

Bivariate analysis of tobacco initiation and nicotine dependence

We then estimated correlations under three alternative models to data of TI and ND: (1) a single-liability model, (2) an independent liability model, and (3) a CCC model. For the latter model, we tested whether the causal paths could be equated across zygosity and gender, and whether the correlations for TI and for ND could be equated. This provided a one-degree-of-freedom test of whether the causal path between TI and ND was significantly different from one, as for the single-liability model. The results favored the CCC model with equal causal paths for all zygosity by gender groups over the single-liability model ($\chi^2_6 = 1058{\cdot}32$). The correlations for TI and ND could not be equated ($\chi^2_1 = 15{\cdot}74$). The causal path was estimated at 0·36, suggesting that the liability to TI and ND were only moderately correlated.

Bivariate analysis of tobacco initiation and regular tobacco use

As RTU was a prerequisite to be evaluated for ND, it should be included in the analysis of the progression from TI to ND. Thus, we fitted the same set of models to TI and RTU to test whether RTU lies on the same liability distribution as TI (single-liability model with two thresholds) or whether its distributions are correlated (CCC model with one threshold per variable). The results favored the CCC model over the single-liability model ($\chi^2_6 = 42{\cdot}75$). This implies that at least partially different genetic and environmental factors contribute to liability to TI and RTU, with an estimate of 0·88 for the causal path. Therefore the final analyses are made including the three variables – TI, RTU and ND – simultaneously in one analysis.

Trivariate analysis of tobacco initiation, regular tobacco use and nicotine dependence

Results of the trivariate analysis paralleled those of the previous analysis, with causal paths of 0·88 between TI and RTU, of 0·70 between RTU and ND and $-0{\cdot}12$ between TI and ND, indicating three highly correlated rather than one underlying liability distribution (correlation between TI and RTU, 0·88, between RTU and ND, 0·59, and between TI and ND, 0·50). Results for the 10-category FTND score indicated that the liabilities to TI, RTU and ND are correlated with causal paths of 0·88 for TI to RTU, 0·91 for RTU to ND, and $-0{\cdot}29$ for TI to ND, which could be equated across zygosity and sex.

Twin correlations and thresholds from the best-fitting model are shown in Table 1. The higher MZ *versus* DZ correlations are consistent with the presence of genetic factors in the liability to TI, RTU and ND, in addition to specific environmental factors. The thresholds were lower for DZ than for MZ twins and for male *versus* female twins, indicating a greater prevalence of TI, RTU and ND in DZ and male twins compared to MZ and female twins.

Extending the models to estimate the contribution of genetic and environmental factors to the liability of TI, RTU and ND also resulted in the selection of the CCC model over the single-liability model (see Table 2). Allowing the nature

Table 1. *Estimates of correlations, causal paths and thresholds for tobacco initiation, regular tobacco use and nicotine dependence in the Virginia Twin Registry sample*

Zygosity-gender groups	Tetrachoric correlations				Causal paths	Zygosity-gender groups	Thresholds		
	TI	RTU	ND				TI	RTU	ND
MZM	0·75	0·80	0·54	TI→RTU	0·88	MZM	−0·93	−0·21	0·58
DZM	0·31	0·40	0·36	RTU→ND	0·57	DZM	−1·12	−0·40	0·41
MZF	0·79	0·83	0·60	TI→ND	0·49	MZF	−0·37	0·30	1·22
DZF	0·49	0·48	0·38			DZF	−0·56	0·12	0·98
DZO	0·32	0·33	0·30			DZOM	−1·13	−0·41	0·54
						DZOF	−0·56	0·06	1·00
	TI	RTU	FTND				TI	RTU	FTND*
MZM	0·73	0·79	0·68	TI→RTU	0·88	MZM	−0·926	−0·204	
DZM	0·32	0·39	0·35	RTU→FTND	0·65	DZM	−1·117	−0·403	
MZF	0·80	0·82	0·60	TI→FTND	0·51	MZF	−0·375	0·301	
DZF	0·49	0·51	0·40			DZF	−0·563	0·116	
DZO	0·31	0·34	0·35			DZOM	−1·126	−0·412	
						DZOF	−0·559	0·057	

* Thresholds for FTND available upon request.

TI, tobacco initiation; RTU, regular tobacco use; ND, nicotine dependence dichotomized; MZM, monozygotic males; DZM, dizygotic males; MZF, monozygotic females; DZF, dizygotic females; DZO, dizygotic opposite-sex twins; DZOM, dizygotic opposite-sex males; DZOF, dizygotic opposite-sex females; FTND, Fagerström Test for Nicotine Dependence score.

Table 2. *Model-fitting results for tobacco initiation, regular tobacco use and nicotine dependence in the Virginia Twin Registry sample*

	EP	−2 log-likelihood	MCN	Likelihood ratio	df	*p*
Tobacco Initiation & Regular Tobacco Use & Nicotine Dependence						
1. Full CCC model	42	17 136·48				
2. 1 + causal paths equated across gender	39	17 140·19	1	3·79	3	0·34
3. 2 + variance components equated across gender	30	17 146·45	2	6·26	9	0·71
4. 3 + no TI→ND	29	17 146·59	3	0·14	1	0·71
5. 4 + no specific c^2 for RTU & ND	27	17 148·04	4	1·59	1	0·21
6. 5 + no c^2 for TI	26	17 148·04	5	1·59	1	0·21
7. 6 + no specific a^2 for RTU	25	17 193·81	6	45·77	1	0·00
8. 6 + no specific a^2 for ND	25	17 168·76	6	20·72	1	0·00
Tobacco Initiation & Regular Tobacco Use & Fagerström Test for Nicotine Dependence score						
1. Full CCC model	96	28 970·85				
2. 1 + causal paths equated across gender	93	28 972·42	1	1·57	3	0·67
3. 2 + variance components equated across gender	84	28 982·55	2	10·13	9	0·34
4. 3 + no TI→FTND	83	28 987·05	3	4·50	1	0·03
5. 3 + no specific c^2 for RTU & FTND	82	28 985·89	3	3·34	1	0·07
6. 5 + no c^2 for TI	81	28 985·83	5	3·28	1	0·07
7. 6 + no specific a^2 for RTU	80	29 038·01	6	52·18	1	0·00
8. 6 + no specific a^2 for FTND	80	29 064·56	6	78·73	1	0·00

EP, number of estimated parameters; MCN, model comparison number; CCC, causal, contingent, common-pathway; TI, tobacco initiation; ND, nicotine dependence dichotomized; RTU, regular tobacco use; FTND, Fagerström Test for Nicotine Dependence score.

of the genetic and shared environmental effects to be different for males and females did not improve the fit of the models. Furthermore, the non-significant likelihood ratio tests between the full model and sub-models constraining the causal paths and the estimates of the genetic and environmental parameters across gender indicated no significant gender differences in the correlation between the liabilities to TI, RTU and ND and in the contribution of genetic and environmental factors to these liabilities. In addition, the causal path from TI to ND could be dropped from the model without a significant loss of fit, suggesting that there was no additional correlation between the liabilities of TI and ND after allowing for correlations between the liabilities of TI and RTU and between RTU and ND. Finally, the shared environmental components specific to the liability to RTU and to the liability to ND were non-significant. In

Table 3. *Estimates of standardized genetic and environmental parameters for tobacco initiation, regular tobacco use and nicotine dependence in the Virginia Twin Registry sample Full model with sex differences*

Variance components	TI	RTU	ND	RTUs	NDs	TI	RTU	FTND	RTUs	FTNDs
a^2m	0·72	0·76	0·50	0·25	0·07	0·72	0·81	0·56	0·32	0·13
c^2m	0·00	0·04	0·12	0·04	0·10	0·00	0·00	0·12	0·00	0·12
e^2m	0·28	0·20	0·38	0·00	0·26	0·28	0·19	0·32	0·00	0·25
a^2f	0·63	0·67	0·40	0·14	0·19	0·58	0·56	0·34	0·08	0·11
c^2f	0·17	0·15	0·18	0·01	0·14	0·21	0·25	0·25	0·07	0·12
e^2f	0·21	0·15	0·46	0·00	0·37	0·21	0·16	0·41	0·02	0·34
Causal paths	TI→RTU	RTU→ND	TI→ND			TI→RTU	RTU→FTND	TI→FTND		
m	0·84	0·69	0·08			0·82	0·87	−0·20		
f	0·92	0·64	−0·09			0·91	0·99	−0·39		
Best-fitting model										
Variance components	TI	RTU	ND	RTUs	NDs	TI	RTU	FTND	RTUs	FTNDs
a^2	0·75	0·80	0·62	0·21	0·24	0·75	0·80	0·67	0·23	0·26
e^2	0·25	0·20	0·38	0·00	0·28	0·25	0·20	0·33	0·02	0·25
Causal paths	TI→RTU	RTU→ND	TI→ND			TI→RTU	RTU→FTND	TI→FTND		
	0·89	0·69	—			0·87	0·93	−0·29		

TI, tobacco initiation; RTU, regular tobacco use; s, specific variance; ND, nicotine dependence dichotomized; FTND, Fagerström Test for Nicotine Dependence score; a^2, additive genetic variance; m, male; c^2, shared environmental variance; e^2, specific environmental variance; f, female.

fact, shared environmental factors did not contribute significantly to the liability to TI. On the contrary, the additive genetic contribution specific to the liability to RTU and to the liability to ND were significant.

The same series of models was fitted to TI, RTU and the ND symptoms. The best-fitting model was the CCC model with equal causal paths and variance components for males and females. Furthermore, additive genetic factors specific to ND symptoms were significant. However, the shared environmental factors specific to ND symptoms, as well as for TI and RTU, could be dropped from the model. In the final AE model (model with additive genetic and specific environmental factors) the third causal path between the liability to TI and ND was estimated to be negative but only borderline significant. If dropped from the model, the effect on the estimated proportions of variance was negligible (see also Table 2).

The estimates of the genetic and environmental contributions to the liability of TI, RTU and ND are presented in Table 3. We opted to present both the estimates under the full model, allowing sex differences in variance components and causal paths, as well as the best-fitting model. Under the best-fitting model, genetic factors accounted for 75% of the variance in TI with the remainder explained by specific environmental factors. Eighty percent of the variance in liability to RTU was attributed to genetic factors, of which 20% were specific to RTU. The heritability for the liability to ND was estimated at 62%, of which one-third was due to factors specific to ND (see also Fig. 3). Specific environmental factors accounted for 38% of the overall variance. The variance specific to ND was almost equally partitioned into genetic and specific environmental factors, accounting respectively for 24% and 28% over the total variance in liability to ND. The corresponding percentages for the variance in liability of ND symptoms are 26% and 25%, with a total genetic contribution of 67%. The shared environmental variance, when included in the model, explained between zero and 25% of the variance in liability to ND but did not reach statistical significance.

DISCUSSION

Heritability of tobacco initiation, regular tobacco use and nicotine dependence

Using a population-based sample of male and female adult twins, the heritability for liability

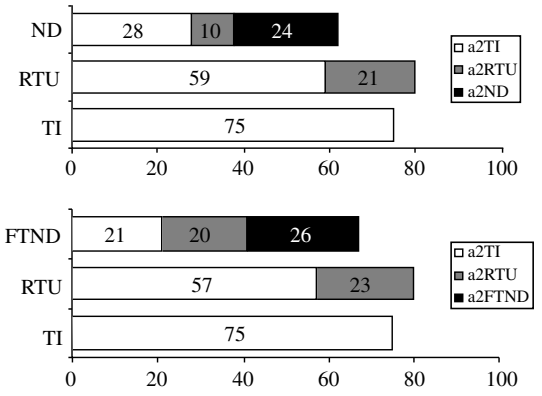

FIG. 3. Proportions of genetic variance from the best-fitting model for tobacco initiation, regular tobacco use and nicotine dependence in the Virginia Twin Registry sample. TI, Tobacco initiation; RTU, regular tobacco use; ND, nicotine dependence dichotomized; FTND, Fagerström Test for Nicotine Dependence score; a2TI, heritability in common with initiation; a2RTU, heritability in common with regular tobacco use; a2ND, heritability in common with nicotine dependence; a2FTND, heritability in common with nicotine dependence symptoms.

to TI was estimated at 75%. Although within the range of estimates obtained in previous studies (Sullivan & Kendler, 1999), these results suggest a considerable genetic influence to initiating tobacco use. In a previous report limited to the female sample, Kendler *et al.* (1999) reported a heritability of 78% for liability to TI. We found no evidence for either gender-specific effects of genes or environments, nor for gender differences in the magnitude of genetic and environmental effects. However, we allowed for prevalence differences in males and females. Results from previous studies on sex differences in the genetic and environmental influences on TI are inconsistent. Greater genetic variance for females than males was found for Australian twins in two earlier reports (Heath & Martin, 1993; Heath *et al.* 1993). A later report by Madden *et al.* (1999) on combined data from Australian and Scandinavian twins showed a higher heritability for females only in the older cohort but not in the younger two cohorts. Higher heritability for Australian females aged 30 and older was confirmed in a recent report (Heath *et al.* 2002). Heath *et al.* (1993) also reported USA data for TI which suggest a higher heritability for males. Their Virginia sample overlaps in part with the data reported here. No sex differences in genetic and environmental influences on TI were found for a sample of Dutch adolescents (Koopmans *et al.* 1999) or Virginia adolescents (Maes *et al.* 1999).

Contrary to most previous studies, shared environmental factors did not contribute significantly to the variation in liability of TI in adult Virginia twins. However, allowing for sex differences in the sources of variation, shared environmental factors accounted for approximately 20% of the variance in females but not males. While USA samples are suggestive of a greater shared environmental influence on TI in females than in males (Heath *et al.* 1993), results from Australian samples (Heath & Martin, 1993; Heath *et al.* 1993; Heath *et al.* 2002) mostly show the opposite, except for the younger age groups (Madden *et al.* 1999). The contribution of shared environmental factors was highest in the adolescent Dutch sample (Koopmans *et al.* 1999). The Virginia adolescent sample (Maes *et al.* 1999), however, did not find shared environmental factors to be significant.

To our knowledge, this was the first study to include RTU, in a conditional approach with TI. Our data suggested that the liabilities to TI and RTU were highly correlated but a single-liability model was rejected. Eighty percent of the variance in liability to RTU was in common with the variance in liability for TI. The variance specific to the liability for RTU was entirely genetic. This resulted in a total heritability for liability of RTU of 80%.

The heritability of ND has also received relatively little attention. Only the previous report by Kendler *et al.* (1999) used an approach similar to the one used here, but was limited to the female sample and did not include a three-stage approach. Results from fitting the correlated liability model to TI and ND in females showed an estimated heritability for ND of 59%, with 12% of the variance accounted for by shared environmental factors. Of the 40% of the variance in liability to ND that was not shared with TI, half was due to familial factors which could be either additive genetic or shared environmental. The results of the present study in the combined male and female sample are consistent with the earlier results, although both the heritability and the shared environmental component to the liability to ND and the causal path are somewhat attenuated. We extended the model to include RTU, as the FTQ questionnaire was only administered to those who

reported RTU of tobacco and applied it to measures of ND: a dichotomous variable and an ND symptom score. Results from all analyses suggested that the liabilities to TI, RTU and ND are correlated. The heritability for liability to ND was estimated around 60–67%, of which the majority is due to factors in common with TI and/or RTU. Although the best-fitting model was an AE model without sex differences in causal paths or variance components, the full model hinted at a greater importance of shared environmental factors, especially in females. Genetic factors specific to the liability to ND accounted for about 25% of the total variance or more than one-third of the genetic variance.

At least six studies have investigated whether the underlying factors for TI are correlated with those for smoking persistence. Early work by Eaves & Eysenck (1980) suggested a single-liability dimension for TI and smoking persistence. Contrary, Hannah *et al.* (1985) observed complete independence of the genetic and environmental factors for initiation and persistence. Heath & Martin (1993) concluded that the combined model (combination of the single-liability model and the independent liability model) fitted best for a young cohort of Australian twins, but could not determine whether the familial resemblance accounting for 49–58% of the variance for smoking persistence was due to genetic or shared environmental factors. The heritability of liability to smoking persistence was estimated to be 70% in male twins from the Vietnam Era Twin Study (True *et al.* 1997) using a combined model. In an extensive study by Madden *et al.* (1999) on three large twin samples in Australia, Sweden and Finland, results from fitting a correlated liability model to smoking initiation and persistence were presented for three age groups. The genetic variance ranged from 39 to 49% and was primarily specific to smoking persistence. The contribution of shared environmental factors to liability of smoking persistence was stronger in females than in males, decreased from the younger to the older age groups, and was mostly in common with smoking initiation. Similar results were found in a recent report on the Australian sample (Heath *et al.* 2002). Although not significant overall, evidence for shared environmental contributions was also greater in females than in males in the current analyses, but largely specific to ND. In fact, the shared environmental factors for TI was estimated at zero in males. In contrast with the Australian and Scandinavian data, the heritability of smoking persistence was mostly accounted for by factors also present for TI and/or RTU. The inclusion of RTU and the use of different measures might explain some of the differences between the results of current and previous analyses.

Relationship between tobacco initiation, regular tobacco use and nicotine dependence

Our results indicated a strong correlation between the liability to TI and the liability to RTU, as well as a strong correlation between the liability to RTU and ND, resulting in a moderate correlation between the liability to TI and ND, with around 50% of the variance for ND shared with variance at TI or RTU. This is consistent with the earliest results on females only (Kendler *et al.* 1999) showing an estimate of 0·77 for the causal path between TI and ND, even though a different definition was used for TI. In both studies, a model assuming a single liability or independent liabilities for TI and ND could be clearly rejected.

Of the studies of TI and persistence, only two fit the same correlated liability model (Madden *et al.* 1999; Heath *et al.* 2002). Both previous studies rejected the independent liability model, as did the current study. However, only a small percentage of the variance was shared between smoking initiation and persistence (7–28%) in the Australian and Scandinavian samples and estimates could be constrained across sex in the younger and older age groups. Only for 26- to 35-year-old Australian and Scandinavian women was a sizable proportion of the variance in smoking persistence shared (46%) with the variance in smoking initiation. Limiting the current analysis to TI and ND, we also found that almost 80% of the variance in liability to ND was not shared with TI.

As far as we know, this is the first study to include TI, RTU and ND in one analysis, with RTU conditional on TI and ND conditional on RTU. It therefore has the added advantage of investigating whether RTU and ND share any additional variation after taking into account TI or whether there are genetic and environmental influences specific to RTU and/or ND. Our data

suggest that there are not only additional primarily genetic factors for RTU but also independent familial and specific environmental factors for ND. The sizable proportion of unique environmental factors specific to the liability of ND could reflect differences in the reliability of the measurement of ND relative to the measurement of TI or RTU. This may also contribute to the reduced power to disentangle genetic from shared environmental factors specific to the liability of ND.

Limitations

This study should be interpreted in the context of potential methodological limitations. First, our sample was entirely Caucasian and we do not know whether a similar pattern would hold in other ethnic groups.

Secondly, despite the large initial sample, power to detect genetic and environmental factors specific to ND is limited, because of the relatively low prevalence and the high correlation between the liability of TI, RTU and ND. However, the current results are consistent with previous studies in suggesting genetic factors specific to ND.

Thirdly, twin resemblance for ND was predicted by frequency of adult contact, which could be a violation of the equal environment assumption. However, that would assume that frequent contact 'causes' resemblance for ND which may be less likely than twins with similar smoking habits choosing to be in closer contact. The validity of the equal environment assumption was supported for most psychiatric disorders including substance use (Kendler & Gardner, 1998).

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DECLARATION OF INTEREST

None.

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