

The Washington University Twin Study of Alcoholism

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Genetic contributions to the liability to develop alcoholism in males of Northern and Western European ancestry are well-established. However, questions remain concerning the role of genetic variation in the etiology of alcoholism among non-white populations, among women, and the possibility of etiological heterogeneity in subtypes of alcoholism. The answers to these questions are needed to help define phenotypes for molecular genetic studies searching for QTLs for alcoholism. Twins from 295 pairs were consecutively ascertained at inpatient and outpatient psychiatric and alcohol treatment facilities in St. Louis, MO in 1981–1986. Probands and willing cotwins were evaluated by structured psychiatric interviews, psychometric assessment, and lifetime treatment records. One hundred fifty-four probands met criteria for alcohol abuse/dependence (AAD), including twins from 45 MZ, 50 same-sex DZ, and 59 opposite-sex pairs. Twin-pair resemblance was evaluated for AAD and alcohol dependence (AD), as well as for subsets defined by gender, patterns of comorbidity, ethnic background, and clinical features. Among males, heritability of AAD and AD was substantial, with little evidence for common environmental contributions to family resemblance. Pair resemblance among females was also substantial, but similar for MZ and DZ pairs, yielding near-zero heritability estimates. However, based on these sample sizes, the sex differences were not statistically significant. The results confirm prior studies of strong genetic influences on alcoholism in males, but suggest lower genetic influence in females. Power to test other sources of heterogeneity was limited, but the results suggest no

evidence for higher heritability for male early onset alcoholism or for alcoholism with comorbid antisocial personality. © 2005 Wiley-Liss, Inc.

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INTRODUCTION

The importance of genetic contributions to the development of alcoholism in males of Northern and Western European ancestry is well-established [e.g., Ball and Collier, 2002; Prescott, 2003; McGue and Iacono, 2004]. It is clear that alcoholism in women from these groups is familial, but there is a lack of consensus as to the relative importance of genetic and environmental sources of vulnerability. Questions also remain concerning genetic contributions to the etiology of alcoholism among non-white populations and the possibility of etiological heterogeneity in alcoholism subtypes.

Three explanations have been advanced for the lack of agreement among studies regarding apparent sex differences in heritability of alcoholism: inadequate statistical power, ascertainment differences among studies, and etiologic heterogeneity.

There is lower statistical power to obtain precise estimates of genetic and environmental effects in women because relatively few genetically-informative studies have included women. Even studies based on large population-based twin registries have included relatively few female cases of alcohol dependence because of the lower prevalence of alcoholism in women. Small samples of twins have low power to distinguish whether familial effects are due to genetic or environmental transmission. Furthermore, heritability estimates may vary widely under different diagnostic definitions or samples. For example, Pickens et al. [1991] reported an heritability for alcohol dependence of 0.42 in a subset of women from the sample found by McGue et al. [1992] to have a heritability of 0.00 for alcohol abuse/dependence (AAD). Although the estimates from various female twin studies are quite different, a re-analysis by Heath et al. [1998] suggests these values do not differ significantly from one another.

Adoption studies, despite their greater power to distinguish genetic from family environmental effects, have included even fewer alcoholic women. Five independent studies which followed up female offspring of alcoholics and controls included only 22 alcoholic women (of 499 studied) born to alcoholic parents and 27 alcoholic women (of 1,259 studied) born to control parents [Roe and Burks, 1945; Goodwin et al., 1977; Cadoret et al., 1980; Bohman et al., 1981; Sigvardsson et al., 1996].

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The finding of lower heritability among women could also arise from ascertainment differences among studies. This is suggested by the observation that studies of female twins which have identified cases through treatment facilities have found the least evidence for genetic contributions, whereas those based on general population samples obtain substantial heritabilities, not different from the value found for men [Prescott, 2003]. Two studies based on US twin samples suggest that treatment seeking is not a simple function of liability for alcoholism [True et al., 1996; Prescott and Kendler, 2000] and that the use of treatment samples could result in underestimates of heritability.

Another possible explanation for variation among study results is etiologic heterogeneity. Some research suggests there are subtypes of alcoholism which differ in degree of genetic etiology [Cloninger et al., 1981; McGue et al., 1992]. If the samples or ascertainment method used in different studies differed in the proportions of these subtypes, this could produce the observed heterogeneity of heritability estimates observed among studies.

One approach used to test for etiologic heterogeneity is to sub-divide samples on the basis of a clinical feature and examine whether heritability differs in these subgroups. Two studies using age at onset found evidence for greater heritability among males with early onset alcoholism. Based on the Swedish adoption studies of alcoholism, Cloninger et al. [1981] developed a typology in which later-onset alcoholism in males was less familial and theorized to be clinically and etiologically more similar to alcoholism in women. In a test of this hypothesis, McGue et al. [1992] divided twin pairs by age of symptom onset in the proband. They found that among pairs in which the proband's symptoms began by age 20, the estimated heritability for AAD was 0.73 compared to 0.30 among pairs with later-onset. The results among female twins were in the same direction, with greater similarity among pairs with early onset than pairs with later-onset (although in the females of this sample, familiarity was due to environmental not genetic sources). These typologies are similar to those described by Babor [1996] and Jellinek [1960], in which female alcoholics typically have later-onsets, and comorbidity characterized by affective and anxiety symptoms, whereas males are more likely to have early onsets and externalizing symptomatology.

Another explanation for sex differences in heritability is sex-specific etiology. Males and females may differ in levels of genetic expression or in processes that interact with sex-dependent characteristics such as hormonal variation or cultural pressures. The existence of sex-specific etiology is indicated when (after adjusting for sex differences in prevalences) the risk to a relative is greater when the relative and proband are the same sex than when they are of different sexes. Studies which have addressed this issue have yielded somewhat contradictory findings [e.g., Cloninger et al., 1978; Heath et al., 1997; Prescott and Kendler, 1999], but suffer from limited statistical power.

Another sex-related etiological process proposed for alcoholism is higher genetic liability in affected females. The sex-dependent threshold model proposed by Cloninger et al. [1978] would explain the sex differences in alcoholism prevalences and predicts a greater risk of alcoholism among relatives of affected women than among relatives of affected men.

As noted previously, these hypotheses are difficult to evaluate due to the absence of large studies of females and the low base rate of alcoholism among women in the general population. Furthermore, higher liability and sex-dependent transmission make partially overlapping predictions. Both predict greater risk to female relatives of female versus male alcoholics. They differ only in their predictions about the relative risks to male relatives of alcoholics. Sex-specificity predicts higher risk among male relatives of male versus female

alcoholics, whereas greater liability among females would predict higher risk among male relatives of female versus male alcoholics.

A further limitation to our understanding of the genetic etiology of alcoholism is the lack of evidence concerning individuals whose ancestry is other than Northern and Western European. Published twin and adoption studies have been based on samples from Scandinavia, the United States, and Australia and have included few participants not of European origin. The existence of ethnic variation for ethanol metabolism suggests there may also be ethnic variation for genetic factors contributing to alcohol use and abuse [e.g., Asaka, 1992]. New US studies of adolescent twins in Missouri [Heath et al., 2002] and the Mid-Atlantic states [Maes et al., 1997] include substantial samples of minority twin pairs and should help to address these deficiencies in the literature.

In summary, prior genetic epidemiological studies have established the importance of genetic factors in the etiology of alcoholism in Caucasian men. However, there remain other questions concerning etiology in women, in non-Caucasian groups, and the possibility of differential heritability among subgroups of alcoholics. In this article, we address these issues using previously unpublished data from the Washington University Twin Study.

METHODS

Subjects

Between 1981 and 1986, Gottesman, Carey, and colleagues at Washington University School of Medicine in St. Louis, MO conducted a prospective twin study in five psychiatric treatment facilities in the St. Louis metropolitan area. These included the inpatient and outpatient services of two private urban teaching hospitals, and the emergency room of a large, inner-city public mental health center. In addition, 21 pairs were ascertained through alcoholism treatment centers. On admission, patients were asked if they had a twin. Those who responded yes were followed up to verify cotwin status. Three hundred and four probands were identified who had a cotwin who was alive or had lived until age 15. Nine of the pairs were doubly ascertained (both twins were independently ascertained through their admission to one of the sites). Thus, the final sample includes 295 twin pairs, 304 probands, and 590 individuals. All participants provided informed consent.

Zygoty determinations were based on detailed questionnaire information about physical similarity and color photographs of heads taken from front and in profile. For a few pairs whose zygoty was problematic, blood groupings were used to make assignments. Four pairs could not be typed based on these sources of information. The remaining 291 pairs included 89 MZ (41 MZM, 48 MZF), 101 same-sex DZ (43 DZM, 58 DZF), and 101 opposite-sex DZ pairs (OS). The proportions of MZ and DZ pairs are comparable to those in the general population.

This sample has been described previously in publications on twin resemblance for personality characteristics [DiLalla et al., 1993; DiLalla and Gottesman, 1995] and illicit substance use disorders [Gynther et al., 1995], as well as in dissertations and abstracts of presentations of twin resemblance for schizophrenia [Prescott and Gottesman, 1990; Prescott, 1991]. The results for alcoholism have not been published but were presented previously [Caldwell and Gottesman, 1991; Caldwell, 1992].

Alcohol diagnoses comprised the most common category of diagnoses for the sample, occurring among 71% of male and 44% of female probands. There were 148 pairs (43 MZ and 105 DZ) in which the proband received a diagnosis of alcohol abuse (AA) or alcohol dependence (AD) and there was adequate clinical information to assign a cotwin diagnosis. Six of these pairs (including 1 MZM, 1 MZF, and 4 OS) were doubly

ascertained, yielding a proband-wise count of 154 pairs (28 MZM, 17 MZF, 26 DZM, 24 DZF, and 59 OS). This group (henceforth called the "alcohol sample") forms the basis for the analyses reported here. The opposite-sex pairs included 40 with a male proband and female cotwin (which we abbreviate OSMf) and 19 pairs with a female proband (OSFm). Thus, the "alcohol proband" group consists of 94 males and 60 females, and the "alcohol cotwin" group consists of their 73 male and 81 female cotwins.

The alcohol sample was 31.8% non-white (all African American). Subjects ranged in age from 15 to 76, with a mean of 34.7 (SD = 12.1) years. Among the alcohol probands, 38% had not completed high school and only 33% were married or cohabiting at the time of interview. Demographic characteristics of the alcohol sample did not differ from those of the sample as a whole.

Measures

All ascertained pairs were invited to participate in a detailed clinical evaluation. This included a structured clinical interview by an experienced clinician using the Diagnostic Interview Schedule [DIS, Robins et al., 1981] and the Home Environment and Lifetime Psychiatric Evaluation Record, and a battery of psychological tests. Twins within a pair were interviewed by different clinicians who were kept blind to the zygosity and clinical status of the cotwin. Assessments were conducted in the hospital, outpatient clinics, or twins' homes. Hospital records from the current admission and discharge summaries from prior inpatient and outpatient admissions were also obtained and reviewed. Interviewers prepared written summaries of the interview, which omitted identifying information. The interviews included family history of heavy drinking and treatment for alcohol problems.

Clinical diagnoses were made by a panel of project clinicians who reviewed interview and admissions information, but were blind to the zygosity, cotwin status, and proband's psychometric data. The panel included a senior clinical psychologist (I.I.G.), a senior psychiatrist (J.W. Knesevich), and several other clinical researchers. Members of the panel made independent decisions as to primary and secondary diagnoses, as well as a level of confidence to each diagnosis after reviewing diagnostic materials and attending a case conference in which the interviewer presented details of the case. For possible diagnoses, which did not reach clinical were rated on a 1–7 scale for "caseness," with 7 representing a probable case just missing diagnostic criteria. Ratings of different judges were combined using a computer algorithm [Vogler et al., 1988], which included diagnoses and caseness ratings of 5 or greater. Ratings were weighted by the number of judges making the ratings. For the purposes of the analyses presented here, subjects whose probability for being in a class exceeded 50% were considered to have the diagnosis. This procedure resulted in 27 diagnostic categories.

Alcoholism definitions employed in the current analyses include DSM-III [American Psychiatric Association, 1980] alcohol dependence with or without abuse (AD), alcohol abuse without dependence (AA), and a broad definition of alcohol abuse and/or dependence (AAD). AD and AA are exclusive categories. Other diagnostic categories included in analyses of comorbidity include antisocial personality disorder and affective/anxiety disorders (including generalized anxiety, panic, phobias, obsessive-compulsive, major depression, and bipolar). These classifications were also based on the algorithm diagnoses.

Following diagnostic reviews, trained research assistants followed a structured procedure to review all sources of materials and complete checklists of demographic information, current and prior alcohol consumption, symptoms and treat-

ment, and a family history checklist. A total symptom score was tallied based on 23 items from the alcohol checklist and included DSM symptoms as well as other signs of drinking problems (e.g., morning drinking, binge drinking, blackouts).

Statistical Analyses

We summarize twin pair similarity using proband-wise concordances and tetrachoric correlations. Tetrachorics can be calculated for clinically ascertained samples of relatives, as long as the degree of selection is known. We calculated twin pair correlations and standard errors using the method described by Smith [1974], (Equation 2b) which permits use of sex-specific thresholds for cotwins and probands. This method assumes a normal distribution of liability and employs an estimate of the population prevalence. We used age- and sex-adjusted lifetime prevalences of AAD and AD from the St. Louis site of the Epidemiological Catchment Area Study [ECA; Robins et al., 1984; Regier et al., 1990]. Pooled over age, the lifetime prevalences were AAD = 0.249, AD = 0.143 for males and AAD = 0.057, AD = 0.035 for females. For comorbidity analyses, we used the estimates of the prevalence of AAD plus the comorbid disorder. The ECA is an excellent source of prevalence estimates for this sample because the data were also collected in the late 1970s using the DIS.

For the purposes of model fitting, we used the NLP procedure in SAS version 8 [SAS Institute, 1999] to calculate the number of pairs in the "unaffected" cell (which is unobserved in a proband design), which were expected given the observed tetrachoric. The resulting 2×2 tables for each group were then used as input into the structural modeling program Mx [Neale et al., 1999]. Although some imprecision may have resulted from rounding, these are expected to be small relative to the standard errors obtained from our modeling analyses. We report estimated proportions of variance and 95% confidence intervals from full models obtained using maximum likelihood estimation.

RESULTS

The alcohol proband group included 61 males and 26 females who met criteria for AD and 33 males and 34 females with AA. The average number of checklist symptoms was 7.7 in males and 6.1 in females, confirming their classification as having significant alcohol-related problems. Demographic characteristics of probands and cotwins were similar; fewer than one-third were currently married or cohabiting and about 40% had not completed high school. Logistic regression analyses predicting presence or absence of AAD in the cotwins from their age, race, educational level, and marital status did not yield any significant associations for either males or females.

As is expected from a sample ascertained primarily through psychiatric services, there was substantial psychiatric morbidity, and often the psychiatric disorder was the presenting problem. Lifetime prevalences for the most common categories among male and female probands, respectively, were: affective/anxiety disorder, 27% and 32%; antisocial personality, 53% and 28%; drug abuse/dependence, 49% and 47%; and schizophrenia, 16% and 20%. For comorbidity analyses we formed three subgroups: AAD + ASP: 59 pairs (19 MZ, 40 DZ) in which the probands had comorbid ASP but no affective or anxiety disorder; AAD + Aff/Anx: 36 pairs (13 MZ, 23 DZ) with an affective or anxiety disorder but not ASP; and AAD + other: 35 pairs (8 MZ, 27 DZ) with a comorbid diagnosis other than ASP or Aff/Anx disorders. Excluded were 16 pairs in which the proband's only diagnosis was AA or AD and 8 pairs in which the proband had both ASP and an affective/anxiety diagnosis.

The AAD + Aff/Anx group tended to be older than the other groups (mean 40 years vs. 33 years) and the majority were of European ancestry. The AAD + ASP probands were

predominantly male (75%) compared to an approximately equal distribution in the other groups. The ASP group tended to have more severe alcohol disorders, as evidenced by earlier onset (17.3 years vs. 23.9 years), more symptoms (means of 8.3 years vs. 5.6 years), a greater proportion with parental alcohol problems (63% vs. 52%), a higher proportion with AD versus abuse only (69% vs. 44%), and presence of another substance use disorder (64% vs. 39%).

Pair Resemblance and Model Fitting

Table I summarizes pair resemblance by zygosity and proband sex. Among males, MZ pairs were significantly more similar than DZ pairs for both AAD and AD. Concordance ratios for MZF and DZF pairs did not differ for any of the diagnostic definitions. In the opposite-sex twin pairs, the high prevalences of AAD and AD among male cotwins of female alcoholics and relatively lower values among female cotwins of male alcoholics are noteworthy, because they suggest a possible increased liability associated with having a female versus male affected sibling.

The third column in Table I displays twin pair resemblance information using a definition of AD in probands and AAD in cotwins. Using a narrow definition in probands increases confidence in the diagnosis, whereas a broader definition in cotwins decreases the possibility of false negatives [e.g., Gottesman and Carey, 1983]. The effect of the broader cotwin definition on the pattern of results in male same-sex pairs is negligible, but among females the twin pair resemblance increases markedly. However, the female MZ and DZ pair resemblance values remain similar to one another. Due to the overall similar pattern of results for different diagnostic definitions, we do not report further analyses using the proband AD–cotwin AAD definition.

Table II displays the results from twin model fitting to the same-sex twin data separately in males and females. For males, the majority of the familial resemblance is assigned to genetic variation. In contrast, the majority of the resemblance in female pairs is assigned to common environmental effects.

However, it is important to note the large standard errors around these estimates, reflecting the low precision associated with dichotomous definitions in these sample sizes.

We conducted a series of follow-up analyses to test the power to reject that the common environmental and genetic parameters differed from zero. There was sufficient power (at $P < 0.05$) to reject the hypothesis of no familial resemblance. Among males, the hypothesis that all pair resemblance was due to environmental variation was rejected for the AAD definition and was borderline for the AD definition. Among females, this hypothesis could not be rejected for either definitions indicating the genetic effect was not significantly different from zero.

Heterogeneity Associated With Sex, Ethnicity, and Clinical Features of Alcoholism

We conducted tests for two types of sex differences in transmission. First, we tested whether there was a sex of proband effect, such that risk for AAD was higher among relatives of female than male probands. This would be indicated by a higher concordance rate among DZ twins of female cotwins (i.e., OSFm > DZM and DZF > OSMf). Although the pattern of results was in the predicted direction, the only comparison to reach statistical significance was that the concordance rate in OSFm pairs (0.57) exceeded that in DZM pairs (0.13). Second, we tested for evidence of sex specific transmission, which would be indicated by same-sex pairs having stronger resemblance than opposite sex pairs. We found no evidence to support this.

We also tested whether the pair correlations could be equated across sex. A global test equating male and female correlations within zygosity (MZM = MZF; DZM = DZF = OSMf = OSFm) was not significant for either AAD ($\chi^2 = 3.5$, $df = 4$) or AD ($\chi^2 = 2.5$, $df = 4$). Based on these results, in subsequent analyses we used our broadest diagnostic definition (AAD in probands and cotwins) and pooled DZ twins from same- and opposite-sex pairs to give maximal power to test other sources of heterogeneity.

TABLE I. Twin Pair Resemblance for DSM-III Alcohol Diagnoses by Zygosity

Zygosity group	N	Diagnostic definition		
		Alcohol abuse or dependence (AAD)	Alcohol dependence (AD) ^a	AD in proband, AAD in cotwin
MZM	28	0.68 ± 0.09 (19/28)	0.40 ± 0.11 (8/20)	0.75 ± 0.10 (15/20)
DZM	26	0.79 ± 0.20 (12/26)	0.54 ± 0.19 (2/15)	0.87 ± 0.20 (7/15)
OSFm	19	0.46 ± 0.19 (11/19)	−0.03 ± 0.26 (4/7)	0.48 ± 0.13 (5/7)
MZF	17	0.58 ± 0.11 (8/17)	0.57 ± 0.19 (2/7)	0.71 ± 0.17 (6/7)
DZF	24	0.43 ± 0.14 (8/17)	0.55 ± 0.22 (2/7)	0.53 ± 0.23 (6/7)
OSMf	40	0.47 ± 0.12 (10/24)	0.29 ± 0.17 (3/12)	0.86 ± 0.13 (8/12)
		0.76 ± 0.16 (13/40)	0.61 ± 0.32 (4/26)	0.94 ± 0.29 (10/26)
		0.42 ± 0.10 (13/40)	0.25 ± 0.13 (4/26)	0.67 ± 0.14 (10/26)
		0.71 ± 0.13 (13/40)	0.56 ± 0.18 (4/26)	0.83 ± 0.17 (10/26)
		0.33 ± 0.07 (13/40)	0.15 ± 0.07 (4/26)	0.38 ± 0.10 (10/26)
		0.88 ± 0.16	0.61 ± 0.19	0.88 ± 0.16

Table entries are: proband-wise concordance ± standard error, (concordant pairs/unadjusted total pairs), tetrachoric correlation ± standard error.

Correlations and standard errors estimated following Smith [1974], based on Epidemiological Catchment Area Study (ECA) population prevalences weighted by age and ethnicity.

^aWith or without alcohol abuse.

TABLE II. Estimated Proportions of Variance and 95% Confidence Intervals for DSM-III AAD

Sex	Definition	Source of variation		
		Additive genetic (h^2)	Common environment (c^2)	Specific environment (e^2)
Male ^a	AAD	0.67 (0.00, 0.95)	0.12 (0.00, 0.78)	0.21 (0.06, 0.54)
	AD	0.48 (0.00, 0.80)	0.00 (0.00, 0.60)	0.52 (0.20, 0.97)
Female ^a	AAD	0.09 (0.00, 0.88)	0.64 (0.00, 0.86)	0.27 (0.07, 0.53)
	AD	0.10 (0.00, 0.93)	0.45 (0.00, 0.82)	0.45 (0.07, 0.92)
Sexes combined ^b	AAD	0.16 (0.00, 0.58)	0.60 (0.27, 0.79)	0.24 (0.10, 0.41)
	AD	0.30 (0.00, 0.80)	0.24 (0.00, 0.61)	0.46 (0.20, 0.77)

^aBased on data from same-sex pairs only.

^bBased on same- and opposite-sex pairs.

Our second major question of interest concerned ethnic differences in sources of variation for alcoholism. Tetrachoric correlations for AAD were calculated using age- and ethnic-adjusted prevalences. Among European Americans (EA), estimates were MZM: $\rho = 0.85$ (SE = 0.18, 15 pairs), DZM $\rho = 0.40$ (0.20, 23), OSFm $\rho = 0.80$ (0.21, 13); MZF: $\rho = 0.90$ (SE = 0.14, 12 pairs), DZF $\rho = 0.68$ (0.15, 18); OSMf: $\rho = 0.68$ (0.13, 24) and among African Americans were MZM: $\rho = 0.74$ (SE = 0.21, 13), DZM $\rho = 0.72$ (0.18, 3), OSFm $\rho = 0.18$ (0.44, 6); MZF: $\rho = 0.00$ (5 pairs), DZF $\rho = 0.78$ (0.23, 6); OSMf: $\rho = 0.48$ (0.19, 16). There are too few African American pairs to provide valid estimates of variance components, so we performed model fitting only in the EA group (Fig. 1). The results suggest a greater role for genetic factors among EA women than is evident from the full sample.

We examined evidence for heterogeneity associated with clinical features of alcoholism. We did not have population prevalence values for these features so could not validly calculate tetrachoric correlations or conduct model fitting. Therefore, we present the proband-wise concordances for the pairs split into high- and low-risk groups based on onset age, symptom severity, and psychiatric comorbidity. The number of MZF pairs (17) was prohibitively small for statistical tests, so the results are presented for their descriptive value. Also because of limited sample sizes, we pooled DZ same- and opposite-sex pairs, based on cotwin gender (DZM with OSFm, DZF with OSMf).

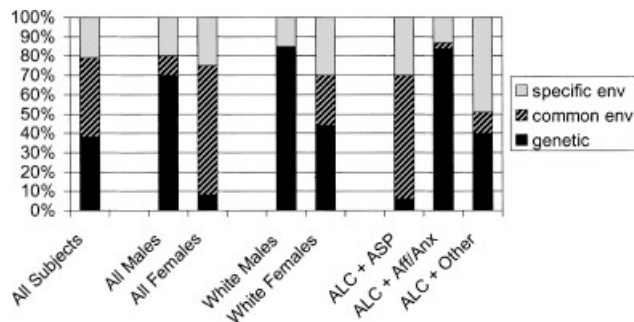


Fig. 1. Estimated genetic and environmental variance contributions to risk for alcoholism by subgroup. ALC, DSM-III alcohol abuse and/or dependence; ASP, antisocial personality; Anx/Aff, comorbid anxiety or affective disorder. See tables for sample sizes, group tetrachoric correlations, and confidence intervals.

The top portion of Table III shows proband-wise concordances based on splitting the pairs by median age at symptom onset in the proband (20 years). There was significantly greater similarity among later-onset MZ pairs than early-onset MZ pairs. The pattern of MZ–DZ differences is consistent with greater genetic contributions to later-onset alcoholism in males, a result that differs from findings reported by others. The female groups did not differ significantly, but the pattern of correlations was consistent with greater heritability in the early onset group.

We next examined evidence for heterogeneity associated with severity of the proband disorder. Pairs were split based on the median number of alcohol-related symptoms (seven in males, five in females). As expected, the proportion of affected cotwins was higher in the groups in which probands had more symptoms. However, this was not associated with a different pattern of MZ–DZ resemblance, suggesting there is no heterogeneity of genetic influence associated with different severity levels.

In 87 of the 154 pairs (56%) one or both parents were reported by the proband or cotwin to have experienced significant alcohol problems. Having an affected parent increased the risk of having an alcohol diagnosis for both males and females compared with the concordances in cotwins with unaffected parents. The pair concordance of MZ male pairs with a positive parental history was significantly higher than MZ males with unaffected parents. The pattern of male MZ–DZ differences also suggests stronger genetic influence in the positive family history group. The differences in the female pairs do not show much evidence of heterogeneity on the basis of parental alcohol history.

Heterogeneity Associated With Comorbidity

Pair resemblance for AAD among twin pairs grouped on the basis of proband comorbidity patterns is shown in Table III. Appropriate prevalences for comorbid disorders were available based on the ECA study, so we were able to calculate tetrachoric correlations and conduct model fitting based on these classifications. Because of the small sample sizes, we combined males and females for these analyses. Estimated pairwise correlations (\pm SE) were: AAD + ASP: MZ = 0.70 (\pm 0.20), DZ = 0.67 (\pm 0.21); AAD + Aff/Anx: MZ = 0.87 (\pm 0.16), DZ = 0.45 (\pm 0.18); and AAD + other: MZ = 0.51 (\pm 0.28), DZ = 0.31 (\pm 0.18). Estimated proportions of variance based on these values are shown in Figure 1. The estimated genetic proportions for the AAD + ASP and AAD + Aff/Anx groups are quite

TABLE III. Pairwise Concordance for AAD Associated With Clinical Features

Clinical feature	Zygosity group			
	MZM (n = 28)	DZM + OSFm (n = 45)	MZF (n = 17)	DZF + OSMf (n = 64)
Age of symptom onset				
Early (<20)	0.56 ± 0.12* ¹ (10/18)	0.48 ± 0.10 (13/27)	0.63 ± 0.17 (5/8)	0.40 ± 0.08 (14/35)
Later (≥20)	0.90 ± 0.09* ^{1,2} (9/10)	0.56 ± 0.12* ² (10/18)	0.33 ± 0.16 (3/9)	0.31 ± 0.09 (9/29)
Symptom severity ^b				
≤Median	0.62 ± 0.13 (8/13)	0.43 ± 0.09 (12/28)	0.29 ± 0.17 (2/7)	0.25 ± 0.08 (8/32)
>Median	0.73 ± 0.11 (11/15)	0.65 ± 0.12 (11/17)	0.60 ± 0.15 (6/10)	0.47 ± 0.09 (15/32)
Parental alcohol problems				
One or both affected	0.76 ± 0.09* ^{3,4} (16/21)	0.54 ± 0.10* ³ (13/24)	0.50 ± 0.16 (5/10)	0.44 ± 0.09* ⁵ (14/32)
Neither affected	0.43 ± 0.19* ⁴ (3/7)	0.48 ± 0.11 (10/21)	0.43 ± 0.19 (3/7)	0.28 ± 0.08* ⁵ (9/32)
Comorbidity group ^c				
AAD + ASP	0.60 ± 0.13 (9/15)	0.53 ± 0.12 (9/17)	0.50 ± 0.25 (2/4)	0.48 ± 0.10 (11/23)
	[0.69 ± 0.21]	[0.57 ± 0.21]	[0.78 ± 0.28]	[0.77 ± 0.12]
AAD + affective/anxiety	1.00 ¹ (6/6)	0.55 ± 0.15 (6/11)	0.43 ± 0.19 (3/7)	0.17 ± 0.11 (2/12)
	[1.00]	[0.61 ± 0.25]	[0.71 ± 0.23]	[0.39 ± 0.25]
AAD + other disorder	0.33 ± 0.27 (1/3)	0.33 ± 0.16 (3/9)	0.40 ± 0.22 (2/5)	0.22 ± 0.10 (4/18)
	[0.13 ± 0.64]	[0.17 ± 0.36]	[0.69 ± 0.28]	[0.42 ± 0.19]

Table entries are: proband-wise concordance ± standard error, (concordant pairs/total pairs), [tetrachoric correlations ± standard error]. Correlations and standard errors estimated following Smith [1974], based on ECA population prevalences for AAD + comorbid disorder weighted by age and ethnicity. Pair concordances are the proportion of cotwins with AAD.

*Test of difference between proportions ($P < 0.05$), superscript indicate groups compared.

¹No discordant pairs, standard errors not computed.

²Median was seven for males, five for females.

³Excludes six pairs in which the proband had both ASP and an affective or anxiety disorder.

different (6% vs. 84%); a test of equivalence across these groups could not be rejected ($\chi^2 = 2.1$, $df = 3$).

DISCUSSION

The major study findings are summarized in Figure 1, including estimated genetic and environmental contributions for the sample as a whole, separately for males and females, for EA males and females, and by comorbidity group. The results replicate the finding from prior twin studies that there are significant genetic contributions to risk for alcoholism among males. The estimated heritability is somewhat higher, but the confidence interval overlaps the estimates reported in other twin studies (50%–60%). The results in females are similar to those found in other studies using cases identified through treatment settings, but differ from those conducted in community-identified twin samples. Results based on treatment samples suggest a stronger role for common environmental effects, and less for genetic influences. It is noteworthy that when the sample was restricted to pairs of European ancestry, the results for females were more similar to those reported in other twin studies.

This study also provides further evidence related to the issue of differing heritability by clinical severity. Like the studies by Pickens et al. [1991] and Kaij [1960], this study employed proband ascertainment with clinical follow-up of cotwins and investigated multiple diagnostic definitions. Unlike those studies, this one did not find higher heritability among males for a narrower diagnostic definition (AD vs. AAD). Similar to these studies, we did find broader definitions were associated with (non-significantly) larger shared environmental effects for females.

Unlike clinically ascertained samples, twin studies based on population registries have not generally found evidence for heritability differences for different diagnostic definitions. This may reflect true etiological differences. The “cases” detected in population-based studies are generally milder and if they differ in etiology, their greater prevalence may obscure the processes underlying more severe cases. An

alternative explanation is ascertainment bias. Cases identified through treatment settings may not be representative of alcoholism in the general population and may find less evidence for genetic influence [e.g., Prescott and Kendler, 2000].

Evidence for Sex-Related Transmission of Alcoholism

This study found evidence consistent with greater risk to relatives of female than male alcoholics. For example, DZ cotwins of a proband with AD were more than twice as likely to have AD if their affected cotwin was female versus male. For AAD, the increase in risk was more modest, about 24%. These values are in the range of those reported in the family, adoption and twin literature for clinically ascertained samples. For example, in their review of the family study literature, McGue and Slutske [1996] found about 60% higher risk to relatives of female versus male alcoholic probands. The results from the adoption studies conducted in Sweden [Sigvardsson et al., 1996] indicated about 45% higher risk to children of alcoholic mothers versus alcoholic fathers. In the Minnesota twin sample, McGue et al. [1992] found a moderate increase in risk among cotwins of female versus male alcoholics.

The family data reported by McGue and Slutske and the Swedish adoption data are inconsistent with sex-specific transmission. The twin data vary, with non-significant negative evidence for sex-specific transmission in the Australian sample [Heath et al., 1997], non-significant positive evidence in the Minnesota sample [McGue et al., 1992], and positive evidence in the Virginia sample which was greater for broader than narrower definitions [Prescott and Kendler, 1999].

Heterogeneity Associated With Comorbidity, Clinical Features, and Ethnicity

The analysis of subgroups classified by comorbid diagnoses was suggestive of etiological heterogeneity. Although not significantly different in these sample sizes, twin pairs in which probands had alcoholism plus antisocial personality had

lower contributions from genetic variation than pairs with comorbid anxiety/depression. This differs from the findings by Cloninger et al. [1981] in which Type II alcoholism (characterized by earlier onset and antisocial personality) was heritable. Although our ASP subgroup was defined only on the basis of comorbid ASP, it was similar to Cloninger's Type II in being predominantly male and having earlier age of onset than the affective/anxiety subgroup. A difference is that our ASP subgroup tended to have more severe alcohol disorders, whereas the Type II group described by Cloninger et al. was intermediate in severity to other groups in their study.

Our findings were not consistent with those reported by McGue et al. [1992] in which males with earlier alcoholism onset had higher heritability. Some of the differences between our results and those reported by others may be related to differences in ascertainment method. In the studies by Cloninger et al. and McGue et al., probands were ascertained directly through treatment for alcoholism or registration for alcohol-related offences. In our study, although probands in the ASP subgroup were usually identified through their diagnosis of alcoholism, those in the other subgroups typically presented for treatment for a disorder other than alcoholism.

The finding that twins from concordant pairs were more likely to report parents with AA suggests these families have a stronger genetic loading. In contrast, pairs without an affected parent were more likely to be discordant, consistent with a sporadic etiology.

This was the first twin study of alcoholism to include a substantial number of African American participants. The sample sizes were still too small to permit precise estimates of genetic and environmental proportions of variance. However, based on the twin pair resemblance, there was no evidence for genetic influence; MZ and DZ pairs were equally similar. Etiological differences between ethnic groups could be due to variation in genetic and/or environmental factors. African Americans are more likely to start drinking later, and to belong to religious denominations, which discourage alcohol use, factors which may influence the risk for the development of alcoholism.

Limitations and Strengths

There are two major limitations associated with this sample: low power and phenotypic heterogeneity. Although the sample is small relative to studies from population-based twin registries, these subjects are particularly informative because they are clear-cut cases. The number of female alcoholic probands in this study ($n = 60$) is larger than that in all adoption studies of alcoholism combined and larger than the number of treated women in any one twin register study. Thus, although some comparisons were underpowered, it is unlikely that larger clinically based studies will be undertaken.

The ascertainment method resulted in substantially higher rates of comorbidity in this sample than would be expected in a sample of alcoholics from the general population. Comparing our alcoholism probands with individuals meeting criteria for AAD in the ECA study, prevalences were similar for affective and anxiety disorders, but differed for other comorbid disorders. For example, the prevalence of ASP was 14.3% in the ECA versus 53% among our probands.

Our analyses did not account for possible violations to the assumptions of the twin method, including random mating for alcoholism risk and equal environmental similarity for MZ and DZ pairs. The existence of assortative mating for alcohol-related phenotypes is likely [Maes et al., 1998] and would lead to underestimates of genetic influences. Violations of the equal environment assumption (EEA) would be suggested by greater similarity of alcoholism-relevant environments among MZ than DZ pairs and would produce over-estimates of genetic

influence. We cannot rule this out, but results from longitudinal studies suggests pair similarity for alcohol-related behaviors may be a cause (not a consequence) of within-pair contact and environmental similarity [Rose et al., 1990], which would not bias genetic estimates.

Despite its limitations, this study has several methodological strengths. First, ascertainment was prospective and nearly complete. Second, the diagnostic assessments were unusually thorough, including structured interviews, multiple sources of information, objective assessment of personality, and multiple experienced diagnosticians. Third, this is the only clinically based adult twin study of alcoholism, which includes a substantial number of minority twin pairs.

The results from this previously unpublished twin study of alcoholism confirm the high familiarity of alcoholism, however defined. Estimates of heritability varied with sex and diagnostic definition, but for the most part, the differences were not statistically significant, in part due to limited statistical power.

One explanation for inconsistent findings regarding genetic heterogeneity in the etiology of alcoholism is that the evidence arising from a particular study may depend on sample variation in prevalences and alcoholism subtypes. The ultimate untangling of the multiple paths to alcoholism may await the quantification of risk from measured genotypes and the assessment of sharing of risk alleles among family members.

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REFERENCES

- American Psychiatric Association. 1980. Diagnostic and statistical manual of mental disorders, 3rd edn. Washington DC: American Psychiatric Association.
- Asaka A. 1992. Drinking behavior and lifestyles in twins. In: Saitoh S, Steinglass P, editors. Alcoholism and the family. Tokyo: Psychiatric Research Institute of Tokyo. pp 24–47.
- Babor TF. 1996. The classification of alcoholics: Typology theories from the 19th century to the present. *Alcohol Health Res World* 20:6–14.
- Ball D, Collier D. 2002. Substance misuse. In: McGuffin P, Owen MJ, Gottesman II, editors. *Psychiatric genetics and genomics*. Oxford: Oxford University Press. pp 267–302.
- Bohman M, Sigvardsson S, Cloninger CR. 1981. Maternal inheritance of alcohol abuse: Cross-fostering analysis of adopted women. *Arch General Psychiat* 38:965–969.
- Cadoret RJ, Cain CA, Grove WM. 1980. Development of alcoholism in adoptees raised apart from alcoholic biologic relatives. *Arch General Psychiat* 37:561–563.
- Caldwell CB. 1992. Heterogeneity in the etiologies of alcoholism: A twin study (dissertation). Charlottesville, VA: University of Virginia.
- Caldwell CB, Gottesman II. 1991. Sex differences in the risk for alcoholism: A twin study. *Behav Genet* 21:563.

- Cloninger CR, Christiansen KO, Reich T, Gottesman II. 1978. Implications of sex differences in the prevalences of antisocial personality, alcoholism, and criminality for familial transmission. *Arch Gen Psychiat* 35:941–951.
- Cloninger CR, Bohman M, Sigvardsson S. 1981. Inheritance of alcohol abuse: Cross-fostering analysis of adopted men. *Arch Gen Psychiat* 38: 861–868.
- DiLalla DL, Gottesman II. 1995. Normal personality characteristics in identical twins discordant for schizophrenia. *J Abnorm Psychol* 104: 490–499.
- DiLalla DL, Gottesman II, Carey G. 1993. Assessment of normal personality traits in a psychiatric sample: Dimensions and categories. *Prog Exp Pers Psychopath Res* 16:137–162.
- Goodwin DW, Schulsinger F, Knop J, Mednick S, Guze SB. 1977. Alcoholism and depression in adopted-out daughters of alcoholics. *Arch Gen Psychiat* 34:751–755.
- Gottesman II, Carey G. 1983. Extracting meaning and direction from twin data. *Psychiatr Res* 1:35–50.
- Gynther LM, Carey G, Gottesman II, Vogler GP. 1995. A twin study of non-alcohol substance abuse. *Psychiatr Res* 56:213–220.
- Heath AC, Bucholz KK, Madden PAF, Dinwiddie SH, Slutske WS, Bierut LJ, Statham DJ, Dunne MP, Whitfield JB, Martin NG. 1997. Genetic and environmental contributions to alcohol dependence risk in a national twin sample: Consistency of findings in women and men. *Psychol Med* 27:1381–1396.
- Heath AC, Slutske WS, Madden PAF. 1998. Gender differences in the genetic contribution to alcoholism risk and alcohol consumption patterns. In: Wilsnack RW, Wilsnack SC, editors. Rutgers, NJ: Rutgers University Press. pp 114–149.
- Heath AC, Howells W, Bucholz K, Glowinski AL, Nelson EC, Madden PA. 2002. Ascertainment of a mid-western US female adolescent twin cohort for alcohol studies: Assessment of sample representativeness using birth record data. *Twin Res* 5:107–112.
- Jellinek EM. 1960. The disease concept of alcoholism. New Haven, CT: College and University Press.
- Kaj L. 1960. Alcoholism in twins. Stockholm: Almqvist and Wiksell.
- Maes HH, Woodard CE, Murrelle L, Meyer JM, Silberg JL, Hewitt JK, Rutter M, Simonoff E, Pickles A, Carbonneau R, Neale MC, Eaves LJ. 1997. Tobacco, alcohol, and drug use in eight-to-sixteen-year-old twins: The Virginia twin study of adolescent behavioral development. *J Stud Alcohol* 60:293–305.
- Maes HH, Neale MC, Kendler KS, Hewitt JK, Silberg JL, Foley DL, Meyer JM, Rutter M, Simonoff E, Pickles A, Eaves LJ. 1998. Assortative mating for major psychiatric diagnoses in two population-based samples. *Psychol Med* 28:1389–1401.
- McGue M, Iacono W. 2004. Behavior genetics principles: Perspectives in development, personality, and psychopathology. In: DiLalla LF, editor. Washington, DC: American Psychological Association.
- McGue M, Slutske WS. 1996. The inheritance of alcoholism in women. In: Howard JM, Martin SE, Mail PD, Hilton ME, Taylor ED, editors. Women and alcohol: Issues for prevention research. NIAAA Research Monograph No. 32. Bethesda, MD: USDHHS. pp 65–91.
- McGue M, Pickens RW, Svikis DS. 1992. Sex and age effects on the inheritance of alcohol problems: A twin study. *J Abnorm Psychol* 101:3–17.
- Neale MC, Boker SM, Xie G, Maes HH. 1999. Mx: Statistical modelling, 5th edn. P.O. Box 980126, Richmond, VA 23298-0126: Virginia Commonwealth University, Department of Psychiatry.
- Pickens RW, Svikis DS, McGue M, Lykken DT, Heston LL, Clayton PJ. 1991. Heterogeneity in the inheritance of alcoholism. *Arch Gen Psychiat* 48:19–28.
- Prescott CA. 1991. Clinical, psychometric, and biometric assessment of schizophrenia: A psychiatric twin study (dissertation). Charlottesville, VA: University of Virginia.
- Prescott CA. 2003. Alcoholism and drug addiction. Encyclopedia of the human genome. London: Nature Publishing Group.
- Prescott CA, Gottesman II. 1990. Biometric prediction of genetic liability for schizophrenia. *Behav Genet* 20:742.
- Prescott CA, Kendler KS. 1999. Genetic and environmental contributions to alcohol abuse and dependence in a population-based sample of male twins. *Am J Psychiat* 156:34–40.
- Prescott CA, Kendler KS. 2000. Influence of ascertainment strategy on finding sex differences in genetic influences from twin studies of alcoholism. *Neuropsychiat Genet* 96:754–761.
- Regier DA, Farmer ME, Rae DS, Locke BZ, Keith SJ, Judd LL, Goodwin FK. 1990. Comorbidity of mental disorders with alcohol and other drug abuse: Results from the Epidemiologic Catchment Area (ECA) Study. *JAMA* 264:2511–2518.
- Robins LN, Helzer JE, Croughan J, Williams JBW, Spitzer RL. 1981. Diagnostic Interview Schedule: Version III. Rockville, MD: National Institute of Mental Health.
- Robins LN, Helzer JE, Weissman MM, Orvaschel H, Gruenberg E, Burke JD, Regier DA. 1984. Lifetime prevalence of specific psychiatric disorders in three sites. *Arch Gen Psychiat* 41:949–958.
- Roe A, Burks B. 1945. Memoirs of the section on alcohol studies, Yale University, No. 3: Adult adjustment of foster children of alcoholic and psychotic parentage and the influence of the foster home. New Haven, CT: Quarterly Journal of Studies on Alcohol.
- Rose RJ, Kaprio J, Williams CJ, Viken R, Obrenski K. 1990. Social contact and sibling similarity: Facts, issues, and red herrings. *Behav Genet* 20:763–778.
- SAS Institute. 1999. SAS/STAT[®] Software: Version 8. Cary, NC: SAS Institute.
- Sigvardsson S, Bohman M, Cloninger CR. 1996. Replication of the Stockholm adoption study of alcoholism: Confirmatory cross-fostering analysis. *Arch Gen Psychiat* 53:681–687.
- Smith C. 1974. Concordance in twins: Methods and interpretations. *Am J Hum Genet* 26:454–466.
- True WR, Heath AC, Bucholz K, Slutske W, Romels JC, Scherrer JF, Lin N, Eisen SA, Goldberg J, Lyons M, Tsuang MT. 1996. Models of treatment seeking for alcoholism: The role of genes and environment. *Alcoholism: Clin Exp Res* 20:1577–1581.
- Vogler GP, Gottesman II, Knesevich JW. 1988. Ratings in a twin study of psychiatric disorders: Judging the judges. Presented at the annual meeting of the Behavior Genetics Association, Nijmegen Netherlands.