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Interaction between *Serotonin Transporter* and *Serotonin Receptor 1 B* genes polymorphisms may be associated with antisocial alcoholism

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Abstract

Background: Several studies have hypothesized that genes regulating the components of the serotonin system, including *serotonin transporter (5-HTTLPR)* and *serotonin 1 B receptor (5-HT1B)*, may be associated with alcoholism, but their results are contradictory because of alcoholism's heterogeneity. Therefore, we examined whether the *5-HTTLPR* gene and *5-HT1B* gene *G861C* polymorphism are susceptibility factors for a specific subtype of alcoholism, antisocial alcoholism in Han Chinese in Taiwan.

Methods: We recruited 273 Han Chinese male inmates with antisocial personality disorder (ASPD) [antisocial alcoholism (AS-ALC) group ($n = 120$) and antisocial non-alcoholism (AS-N-ALC) group ($n = 153$)] and 191 healthy male controls from the community. Genotyping was done using PCR-RFLP.

Results: There were no significant differences in the genotypic frequency of the *5-HT1B G861C* polymorphism between the 3 groups. Although AS-ALC group members more frequently carried the *5-HTTLPR S/S*, *S/L_G* and *L_G/L_G* genotypes than controls, the difference became non-significant after controlling for the covarying effects of age. However, the *5-HTTLPR S/S*, *S/L_G* and *L_G/L_G* genotypes may have interacted with the *5-HT1B G861C C/C* polymorphism and increased the risk of becoming antisocial alcoholism.

Conclusion: Our study suggests that neither the *5-HTTLPR* gene nor the *5-HT1B G861C* polymorphism alone is a risk factor for antisocial alcoholism in Taiwan's Han Chinese population, but that the interaction between both genes may increase susceptibility to antisocial alcoholism.

Keywords: Antisocial alcoholism, Serotonin, *Serotonin transporter gene*, *5-HT1B gene*

Background

Alcoholism (also: alcohol dependence) is a complex, heterogeneous disorder that is influenced by multiple genes as well as sociocultural factors [1,2]. Twin studies have shown that genetic factors account for about 50-60% of the overall variance in alcoholism [3-5]. Many candidate genes have been studied; however, the results are controversial. Therefore, to reduce the heterogeneity of alcoholism by proper subtyping is important. In addition, a

gene-to-gene interaction approach may be more revealing than a single-gene approach for studying alcoholism. By using appropriate subtypes of alcoholism and the gene-to-gene interaction approach, the results may be more conclusive.

Cloninger (1987) identified two subtypes of alcoholism. Type I alcoholism is characterized by late onset, more psychological dependence, and anxious/depressed personality traits. Individuals with type II alcoholism are characterized by early onset, a higher familial risk for alcoholism, and antisocial personality traits [1]. We previously subdivided alcoholism into three categories: pure, anxious/depressive, and antisocial (AS) [6]. We showed that antisocial alcoholism (AS-ALC; alcoholism comorbid

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with antisocial personality disorder), which overlaps to some extent with Cloninger's type II alcoholism, have a strong genetic predisposition to alcoholism and different genetic polymorphisms than do other subtypes of alcoholism. The protective effect of the *ADH1B*2* allele against alcoholism vanished in antisocial alcoholics [6], and the protective effect of the *ALDH2*2* allele might disappear in individuals with antisocial personality disorder (ASPD) and carrying the MAOA-uVNTR 4-repeat allele [7]. Antisocial alcoholics had higher novelty-seeking scores with an interaction between the *DRD2 TaqI A1 (A1A1 or A1/A2)* and *5-HTTLPR S/S* genotype [8]. Moreover, antisocial alcoholics had different polymorphisms of the *serotonin (5-HT) receptor 1B A-161T* from other subtypes of alcoholics [9]. All the evidence supports the typology of alcoholism and the particular genetic vulnerability of antisocial alcoholics.

Numerous studies have reported reduced brain serotonin (5-hydroxytryptamine, 5-HT) function in patients who are similar to antisocial alcoholics. Lower cerebrospinal fluid concentration of the 5-HT metabolite 5-HIAA (5-hydroxyindoleacetic acid) has been found in early-onset antisocial alcoholics [10], impulsive alcoholic criminals [11,12], and alcoholic fire setters [13,14]. In addition, the variation of the central 5-HT system has been shown to modulate alcohol consumption and may contribute to the risk of alcoholism [15-17]. Genes encoding the 5-HTT gene (the serotonin transporter protein; genetic locus SLC6A4) and 5-HT receptor, which may cause different levels of activity of 5-HTT, have been examined as candidate genes for the risk of alcoholism. The polymorphism of the *5-HTT-linked promoter region (5-HTTLPR)*, which contains an insertion-deletion of two 22-bp repeats in a region of the promoter with multiple 22- to 23-bp repeats [18], affects the expression of serotonin transporter [19]. In vitro study, the short variant (S-allele) of the *5-HTTLPR* displayed reduced transcriptional activity [19] as well as decreased serotonin transporter expression and serotonin uptake in lymphoblast cell lines [18]. *5-HTTLPR* polymorphisms were also correlated to the function of serotonin transporter both in vivo imaging study [20] and in postmortem brain study [21]. Human SPECT study showed that homozygous carriers of the long allele (*L/L*) had an increase in the availability of raphe serotonin transporter [20]. Serotonin transporter mRNA levels in subjects with *S/L* and *S/S* genotypes were also lower than in subjects with the *L/L* genotype in postmortem brain study [21]. Although the S allele has been linked to a risk of alcoholism [22,23] and to antisocial alcoholics or type II alcoholics [24-26], this link is still controversial [20,27,28]. In addition to the long and short alleles, other subtypes of the L allele have been described [29]. The long variant with an adenosine at single nucleotide polymorphism (SNP) rs25531 (*L_A*) has been reported to have higher

activity than the long variant with a guanine at rs25531 (*L_G*) [30,31]. Furthermore, other uncommon alleles that are longer than the L variant were found in several studies [29,32-35]. The extra-long allele or "novel allelic variant" were called *XL* variants with 15, 18,19, 20 or 22 repeats with unknown function [29,32-35]. Hence, the past dichotomous classification of *5HTTLPR* into *S* and *L* allele may not be sufficient and the association between the tri-allelic 5-HTTLPR polymorphism (*S*, *L_A*, *L_G*) and the risk of alcoholism or antisocial alcoholism remains unknown.

The serotonin 1B receptors (5-HT1B) are located on presynaptic and postsynaptic terminals and function as both autoreceptors and heteroreceptors that mediate the release of serotonin [36]. The *5-HT1B* gene has been associated with alcohol intake and aggression in animal and human studies. An alcohol preference locus has been mapped in mice to the region where *5-HT1B* is located [37]. *5-HT1B* receptor knockout mice were found to have increased aggressive behavior [38] and increased spontaneous alcohol consumption [39]. In human studies, of 16 reported polymorphisms of *5-HT1B*, the results of association studies that used the *G861C* polymorphism remain inconsistent [40]. Some studies associated polymorphisms of *5-HT1B G861C* with alcoholism [41] or antisocial alcoholism [42]. People with the *5-HT1B 861C* allele also had 20% fewer *5-HT1B* binding sites in the prefrontal cortex, which was the same as the alcoholism group [43,44]. However, subsequent studies did not confirm the association [45-47]. The heterogeneity of alcoholism, as well as different populations and ethnicities, may cause the discrepancy.

The prevalence rate of ASPD and antisocial behaviors in individuals with alcohol use disorders ranges from 20% to 33% [48-50]. Because of their high comorbidity, most studies have not distinguished the effects of ASPD from those of alcoholism in Western populations [51]. Being able to specify whether the genes were involved in pure ASPD (ASPD not comorbid with alcoholism (AS-N-ALC)) or AS-ALC, or both, may clarify the nature of these disorders and elucidate whether past genetic associations with AS-ALC were merely contributed by ASPD.

The aims of current study is to (1) investigate whether the tri-allelic *5-HTTLPR* polymorphism and *5-HT1B G861C* polymorphism are associated with AS-ALC in the Han Chinese population in Taiwan, and (2) assess the serotonergic genetic interaction between *5-HTTLPR* and *5-HT1B G861C* polymorphisms and antisocial alcoholism.

Methods

Participants and clinical assessments

This study was approved by the Institutional Review Boards for the Protection of Human Subjects at the Tri-Service General Hospital and National Cheng Kung

University Hospital. Because of the low prevalence (0.1%) of ASPD in Han Chinese in Taiwan [52], it is difficult to recruit study participants with ASPD from clinics or from the community. Instead, we recruited 273 Han Chinese men with ASPD from a prison in Taipei (northern Taiwan) ($n=173$) and a prison in Tainan (southern Taiwan) ($n=100$). The research protocol was reviewed and approved by the Taiwan Ministry of Justice, which permitted us to collect blood samples from the participants. The procedures were fully explained to each participant before they gave written informed consent; however, there were no advocates for the inmates present when they were asked to sign the consent form. Participants were told that they were free to drop out of or to join the study at any time. We offered several compensatory benefits: 1) a free physical examination, 2) liver and renal function tests, and 3) and free treatment or assistance in transferring those with psychiatric or physical illnesses to hospitals. Each participant was initially evaluated by an attending psychiatrist and then interviewed by a psychologist well-trained in using the DSM-IV and the Modified Chinese Version of the Modified Schedule of Affective Disorder and Schizophrenia-Life Time (SADS-L) [53,54]. The inter-rater reliability of the Chinese version of the modified SADS-L was good with the kappa values range from 0.71 to 1.00 in several major mental illness [54]. The SADS-L was also found to be adequate for diagnosing ASPD with or without alcoholism [6,55-57]. Participants with major mental illnesses including schizophrenia, bipolar disorder, multiple substance abuse, or other organic mental disorders, and those who refused to participate ($< 10\%$) were excluded from the study. None of the participants was taking any kind of medication. All who met the criteria for a diagnosis of antisocial personality disorder with or without alcoholism were assigned to the AS-ALC group ($n=120$) or the AS-N-ALC group ($n=153$), respectively.

The healthy control group included 191 male volunteers recruited from the community. The Chinese version of the SADS-L was used to screen their psychiatric conditions. All controls were free of present and past major and minor mental illness (affective disorder, schizophrenia, anxiety disorder, personality disorder, alcohol abuse and dependence, and illegal substance use disorder), and none had a family history of psychiatric disorders among their first-degree relatives.

Blood samples and genotyping for 5-HTTLPR and 5-HT1B G861C polymorphisms

Twenty milliliters of blood was drawn from each participant, and standard methods were used to extract genomic DNA from the leukocytes. The genotypes of 5-HTTLPR functional polymorphisms were genotyped using polymerase chain reaction plus restriction fragment length

polymorphism (PCR-RFLP) analysis [18]. The A/G SNP of the *L* allele were determined using a modified protocol [58]. The G861C polymorphism of the 5-HT1B gene was genotyped using a modified protocol [42].

Statistical analyses

One-way analysis of variance (ANOVA) and the Bonferroni Post Hoc Test were used to determine the mean age differences between the subgroups. The difference in genotype frequency of 5-HTTLPR and 5-HT1B G861C polymorphisms between three groups was calculated using the Pearson χ^2 test (2-tailed). The 5-HTTLPR polymorphisms in AS-ALC and AS-N-ALC groups were further stratified by the 5-HT1B G861C polymorphism using the χ^2 tests. Logistic regression analysis was used to examine the main effect and gene-to-gene interaction of the 5-HTTLPR and 5-HT1B G861C polymorphisms for the risk of antisocial alcoholism. Both the S_A and the S_G alleles were recorded as the *S* allele because there is no functional difference in the SNPs [59]. Because past studies found that *S* and L_G carrier had lower function and expression of serotonin transporter [18-20,60], we divided the 5-HTTLPR polymorphisms into 2 groups, low-functional (S/S , S/L_G , L_G/L_G) and high-functional (S/L_A , L_G/L_A , L_A/L_A , S/XL , L_A/XL , L_G/XL), for data analysis. There was only 8 participants carried the $L_A L_A$ genotype in our study, so we did not separate the homozygous L_A carries from heterozygous L_A carriers into another group. SPSS for Windows 17.0 (SPSS Inc., Chicago, IL, USA) was used for all statistical analyses.

The power estimation was calculated using G-power 3.1 software [61,62]. Our total sample size ($n=464$) had a power of 0.58 to detect a small effect (effect size = 0.1), of 0.99 to detect a medium effect (effect size = 0.3), and of 1.00 to detect a large effect (effect size = 0.5) of genotype distributions.

Results

Healthy controls were significantly older than those in the AS-ALC ($P=0.004$) and AS-N-ALC groups ($P=0.009$; mean age: $F=6.87$; $df=2$, 461 (Table 1). A Bonferroni post hoc test did not show a significant age difference between the AS-ALC and AS-N-ALC groups.

Genotype distributions of 5-HTTLPR and 5-HT1B G861C were in Hardy-Weinberg equilibrium (5-HTTLPR: $P=0.75$ in AS-ALC and $P=0.22$ in AS-N-ALC; 5-HT1B G861C: $P=0.07$ in AS-ALC and $P=0.72$ in AS-N-ALC). The distribution of the 5-HTTLPR and 5-HT1B G861C genotypes was not significantly different between the three groups (Table 2). However, there was a higher frequency of the low-functional 5-HTTLPR polymorphism (S/S , S/L_G , L_G/L_G) in the AS-ALC group than in the control group (Table 2). After stratifying the 5-HTTLPR polymorphisms with the 5-HT1B G861C polymorphism, a

Table 1 Demographic data

Group	n	Age (years)	F	P-value
AS-ALC	120	32.4 ± 7.6	6.87	0.001
AS-N-ALC	153	32.8 ± 7.5		
Controls	191	35.5 ± 9.4		

AS-ALC, antisocial alcoholism; AS-N-ALC, antisocial personality disorder without alcoholism.

significant difference in the frequency of *5-HTTLPR* polymorphisms was found after controlling for the *5-HT1B G861C C/C* genotype for both AS-ALC versus controls and AS-ALC versus AS-N-ALC (Table 3). A logistic regression analysis, done to control the possible covarying effect of age, showed that the *5-HTTLPR* and *5-HT1B G861C* genotypes were not significantly different between the AS-ALC and control groups or the AS-ALC and the AS-N-ALC groups (Table 4). However, the regression did reveal a significant interaction between the *5-HTTLPR* and *5-HT1B G861C* genes. Compared with AS-N-ALC and control groups, the interaction between the *5-HTTLPR* low-functional group (*S/S, S/L_G, L_G/L_G* genotype) and the *5-HT1B G861C C/C* genotype may increase the risk of becoming an antisocial alcoholic ($P = 0.04$, odds ratio (OR) = 6.22 and 5.76, respectively) (Table 4).

Discussion

We found that interaction between the *5-HTTLPR S/S, S/L_G, L_G/L_G* genotypes and the *5-HT1B G861C C/C* genotypes was associated with the AS-ALC group compared with the control and AS-N-ALC groups. Unlike past studies [24-26,43], we did not find an association between the *5-HTTLPR* or the *5-HT1B G861C* polymorphisms with AS-ALC. Although the AS-ALC group seemed to have a higher frequency of the *5-HTTLPR S/S, S/L_G, L_G/L_G* genotype than did the control group, when the effect of age was controlled by logistic regression, the difference was not significant.

The interaction between the *5-HTTLPR S/S, S/L_G, L_G/L_G* and the *5-HT1B G861C C/C* genotypes may result in reduced brain serotonin function. Because

patients with the *5-HT1B 861 C* allele were found with fewer 5-HT1B binding sites in the prefrontal cortex [43,44] and the *5-HTTLPR S/S, S/L_G, L_G/L_G* was the low-functional group [30,31], our results imply that only the combined effect of serotonergic dysfunction from the studied genes, but not from individual genes, may be associated with risk for antisocial alcoholism. Several other reasons may also account for our positive finding. First, past studies compared the *5-HTTLPR* polymorphism between antisocial alcoholics and healthy (without ASPD and non-alcoholics) controls. Because ASPD has been linked to the *5-HTTLPR* polymorphism [63-65], we tried to control for the confounding effect of ASPD by comparing antisocial alcoholics with healthy controls and antisocial non-alcoholics. Second, our healthy control group belonged to a super-control group that excluded any other major or minor mental illness, while in other studies [25,26], the authors included only individuals from unscreened community controls and no antisocial non-alcoholics. Moreover, we used a tri-allelic functional *5-HTTLPR* polymorphism instead of a bi-allelic polymorphism. All these factors may have caused results different from those of other reports.

We found no association between antisocial alcoholism and the *5-HT1B G861C* polymorphism. A previous study [43] showed that the frequencies of the *5HT1B 861 C* allele varied widely among different ethnic groups, ranging from 20% in African-Americans to 36% in Hispanics and 42% in Asians. In our current study, the frequency of the *5-HT1B C* allele (48.9% in the control group, 49.6% in the AS-ALC group, and 47.1% in the AS-N-ALC group) was higher than in previous reports. We also added an AS-N-ALC group to decrease the effect of ASPD. Different populations and ethnicities may also partially explain our different results.

Our past study [66] showed that antisocial alcoholics and antisocial non-alcoholics shared a similar genetic vulnerability of dopamine-related genes, which was different from healthy controls. However, in the present study, we did not find the same distributions of the *5-HTTLPR* and *5-HT1B* genes in the three groups. It seems that the antisocial non-alcoholics and healthy

Table 2 Genotype distribution and frequency of the 5HTTLPR and 5HT1B G861C polymorphism

Group	n	5HTTLPR genotype frequency (%)				5HT1B G861C genotype frequency (%)				
		S/S, S/L _G , L _G /L _G	S/L _{A1} , L _G /L _{A1} , L _{A1} /L _{A1} S/XL, L _{A1} /XL, L _G /XL	χ ²	P-value	G/G	G/C	C/C	χ ²	P-value
AS-ALC	120	90 (75.0)	30 (25.0)	5.39	0.02 ^{a*}	25 (20.8)	70 (58.3)	25 (20.8)	4.58	0.10 ^a
AS-N-ALC	153	100 (65.4)	53 (34.6)	2.95	0.09 ^b	44 (28.8)	74 (48.4)	35 (22.9)	3.07	0.22 ^b
Controls	191	119 (62.3)	72 (37.7)	5.50	0.06 ^c	55 (28.8)	88 (46.1)	48 (25.1)	5.04	0.28 ^c

AS-ALC: Antisocial alcoholism;

AS-N-ALC: Antisocial personality disorder without alcoholism.

^a AS-ALC vs. Healthy Controls.

^b AS-ALC vs. AS-N-ALC.

^c AS-ALC vs. AS-N-ALC vs. Controls.

* $P < 0.05$.

Table 3 5HTTLPR genotype frequency after stratification of 5HT1B G861C genotype

G861C Genotype	Groups	5HTTLPR genotype (%)			χ^2	P
		S/S, S/L _G , L _G /L _G	S/L _A , L _G /L _A , L _A /L _A	S/XL, L _A /XL, L _G /XL		
AS-ALC vs. Controls						
G/G	AS-ALC	15 (60.0)	10 (40.0)	0.023	0.878	
	Controls	32 (58.2)	23 (41.8)			
G/C	AS-ALC	53 (75.7)	17 (24.3)	1.086	0.297	
	Controls	60 (68.2)	28 (31.8)			
C/C	AS-ALC	22 (88.0)	3 (12.0)	7.509	0.006*	
	Controls	27 (56.3)	21 (43.8)			
AS-ALC vs. AS-N-ALC						
G/G	AS-ALC	15 (60.0)	10 (40.0)	0.784	0.376	
	AS-N-ALC	31 (70.5)	13 (29.5)			
G/C	AS-ALC	53 (75.7)	17 (24.3)	3.075	0.079	
	AS-N-ALC	46 (62.2)	28 (37.8)			
C/C	AS-ALC	22 (88.0)	3 (12.0)	3.863	0.049*	
	AS-N-ALC	23 (65.7)	12 (34.3)			

*P < 0.05

AS-ALC, antisocial alcoholism; AS-N-ALC, antisocial personality disorder without alcoholism.

Table 4 Logistic regression analysis of 5HTTLPR gene and 5HT1B receptor gene G861C polymorphism and their interaction for the risk of antisocial alcoholism (AS-ALC)

Models	AS-ALC			
	B	OR	95% CI	P-value
Model 1				
5HTTLPR low functional group	-0.47	0.62	0.22-1.75	0.37
5HT1B G861C G/C	-0.21	0.81	0.29-2.27	0.69
5HT1B G861C C/C	-1.13	0.32	0.07-1.46	0.14
5HTTLPR low-functional group*5HT1B G861C G/C	1.09	2.97	0.84-10.44	0.09
5HTTLPR low-functional group*5HT1B G861C C/C	1.83	6.22	1.10-35.27	0.04*
Model 2				
5HTTLPR low-functional group	0.14	1.15	0.43-3.07	0.78
5HT1B G861C G/C	0.49	1.63	0.61-4.35	0.33
5HT1B G861C C/C	-1.26	0.28	0.07-1.19	0.09
5HTTLPR low-functional group*5HT1B G861C G/C	0.07	1.07	0.32-3.65	0.91
5HTTLPR low-functional group*5HT1B G861C C/C	1.75	5.76	1.09-30.44	0.04*

5HTTLPR low-functional group: S/S, S/L_G, L_G/L_G

5HTTLPR high-functional group: S/L_A, L_G/L_A, L_A/L_A, S/XL, L_A/XL, L_G/XL

B, coefficients; CI, confidence interval; OR, odds ratio

Model 1, reference groups are: 5HTTLPR high-functional group, 5HT1B G861C G/G genotypes, and AS-N-ALC group; covarying for age

Model 2, reference groups are: 5HTTLPR high-functional group, 5HT1B G861C G/G genotypes, and controls; covarying for age

*P < 0.05.

controls had a similar genotype distribution of these serotonin-related genes, one different from that of the antisocial alcoholics. We may speculate that other serotonin-related genes have the same distribution between individuals with ASPD and the general population. In addition, serotonin may modulate the reinforcement processes, possibly through complex interactions with the mesolimbic dopamine system [60,67] and the regulatory functions of several serotonin receptor subtypes, including 5-HT1B receptor [68-70]. The interaction of the dopaminergic and serotonergic neurotransmitter systems in the nucleus accumbens and the ventral striatum is thought to be critical in addiction [60,70,71]. Furthermore, enzymes involved in dopamine metabolism will also metabolize serotonin. Serotonin is initially metabolized to 5-hydroxyindole-3-acetaldehyde (5-HIAL) by monoamine oxidase (MAO) [72], and then oxidized to 5-hydroxyindole-3-acetic acid (5-HIAA) by aldehyde dehydrogenase (ALDH) [73]. We have reported [74] that the *dopamine D2 receptor* gene interacted with the *5-HTTLPR* gene in antisocial alcoholics with specific personality traits. Hence, additional studies to explore the associations among the *5-HTTLPR*, the *5-HT1B* gene, 5-HT metabolic enzyme-related genes, and dopamine-related genes will be required to study the pathogenesis of AS-ALC.

Our study has several limitations. First, the study populations in the AS-ALC and AS-N-ALC groups were small. In addition, the cell size with the significant finding was also small, which may cause the risk of false positive results. Larger study populations are needed. Second, there were few participants with the *5-HTTLPR* L_A/L_A genotype, and about 3.8% with the *5-HTTLPR* XL allele, which is different from other ethnic groups [30]. It may be feasible to divide those with the *5-HTTLPR* S/L_A, L_G/L_A, L_A/L_A, S/XL, L_A/XL, L_G/XL genotypes into a high-functional group and those with the S/S, S/L_G, L_G/L_G genotypes into a low-functional group for statistical analysis according to past study results [18-20,60]. Although the function of the *5-HTTLPR* XL allele remains unknown, one study had speculated that it may function as an L allele [75]. However, additional study will be required to test its function since a significant proportion of Han Chinese carry this allele [75]. Finally, it is very likely that individuals with ASPD may belong to a specific group since members of both the AS-ALC and AS-N-ALC groups were all recruited from jail, which caused sampling biases. In addition, we used controls without any past psychiatric history from the community. Regardless of alcoholism, the inmate participants with ASPD were substantially different from the healthy community participants [75]. It is necessary to be aware of the possibility of false-positive results when comparing these phenotypes with super-normal controls⁴².

Our results supported the notion that antisocial alcoholism is associated with the interaction between the 5-HTTLPR and 5-HT1B G861 polymorphisms. Additional studies to examine the 5-HTTLPR and 5HT1B G861C gene polymorphisms and their interaction effects with the dopaminergic gene in antisocial alcoholism are required.

Patient consent
Obtained.

Ethical approval

This study was approved by the Institutional Review Boards for the Protection of Human Subjects at the Tri-Service General Hospital and National Cheng Kung University Hospital.

Abbreviations

5-HTTLPR: Serotonin transporter; 5-HT1B: Serotonin 1 B receptor; ASPD: Antisocial personality disorder; AS-ALC: Antisocial alcoholism; AS-N-ALC: Antisocial non-alcoholism.

Competing interests

The authors declare no conflict of interests.

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Authors' contributions

The author, TYW wrote the first draft of this manuscript and designed this study with the author RBL. The authors, SYL and Y-HC managed the statistical analyses. The authors, SLC, SHC, CHC and CLC managed the lab work. The authors, TYW, SYH, NST, IHL, TLY, YKY and RBL managed the patients recruitment. All authors read and approved the final manuscript.

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References

1. Cloninger CR: Neurogenetic adaptive mechanisms in alcoholism. *Science* 1987, **236**:410–416.
2. Edenberg HJ, Foroud T: The genetics of alcoholism: identifying specific genes through family studies. *Addict Biol* 2006, **11**:386–396.
3. Kendler KS, Heath AC, Neale MC, Kessler RC, Eaves LJ: A population-based twin study of alcoholism in women. *JAMA* 1992, **268**:1877–1882.

4. Heath AC, Bucholz KK, Madden PA, Dinwiddie SH, Slutske WS, Bierut LJ, Statham DJ, Dunne MP, Whitfield JB, Martin NG: Genetic and environmental contributions to alcohol dependence risk in a national twin sample: consistency of findings in women and men. *Psychol Med* 1997, **27**:1381–1396.
5. Kendler KS, Prescott CA, Neale MC, Pedersen NL: Temperance board registration for alcohol abuse in a national sample of Swedish male twins, born 1902 to 1949. *Arch Gen Psychiatry* 1997, **54**:178–184.
6. Lu RB, Ko HC, Lee JF, Lin WW, Huang SY, Wang TJ, Wu YS, Lu TE, Chou YH: No alcoholism-protection effects of ADH1B*2 allele in antisocial alcoholics among Han Chinese in Taiwan. *Alcohol Clin Exp Res* 2005, **29**:2101–2107.
7. Lee SY, Hahn CY, Lee JF, Chen SL, Chen SH, Yeh TL, Kuo PH, Lee IH, Yang YK, Huang SY, et al: MAOA-uVNTR polymorphism may modify the protective effect of ALDH2 gene against alcohol dependence in antisocial personality disorder. *Alcohol Clin Exp Res* 2009, **33**:985–990.
8. Wu CY, Wu YS, Lee JF, Huang SY, Yu L, Ko HC, Lu RB: The association between DRD2/ANKK1, 5-HTTLPR gene, and specific personality trait on antisocial alcoholism among Han Chinese in Taiwan. *Am J Med Genet B Neuropsychiatr Genet* 2008, **147B**:447–453.
9. Lee SY, Lin WW, Huang SY, Kuo PH, Wang CL, Wu PL, Chen SL, Wu JY, Ko HC, Lu RB: The relationship between serotonin receptor 1B polymorphisms A-161T and alcohol dependence. *Alcohol Clin Exp Res* 2009, **33**:1589–1595.
10. Fils-Aime ML, Eckardt MJ, George DT, Brown GL, Mefford I, Linnoila M: Early-onset alcoholics have lower cerebrospinal fluid 5-hydroxyindoleacetic acid levels than late-onset alcoholics. *Arch Gen Psychiatry* 1996, **53**:211–216.
11. Linnoila M, Virkkunen M, Scheinin M, Nuutila A, Rimon R, Goodwin FK: Low cerebrospinal fluid 5-hydroxyindoleacetic acid concentration differentiates impulsive from nonimpulsive violent behavior. *Life Sci* 1983, **33**:2609–2614.
12. Linnoila M, De Jong J, Virkkunen M: Family history of alcoholism in violent offenders and impulsive fire setters. *Arch Gen Psychiatry* 1989, **46**:613–616.
13. Virkkunen M, Nuutila A, Goodwin FK, Linnoila M: Cerebrospinal fluid monoamine metabolite levels in male arsonists. *Arch Gen Psychiatry* 1987, **44**:241–247.
14. Virkkunen M, Rawlings R, Tokola R, Poland RE, Guidotti A, Nemeroff C, Bissette G, Kalogeras K, Karonen SL, Linnoila M: CSF biochemistries, glucose metabolism, and diurnal activity rhythms in alcoholic, violent offenders, fire setters, and healthy volunteers. *Arch Gen Psychiatry* 1994, **51**:20–27.
15. Kranzler HR, Anton RF: Implications of recent neuropsychopharmacologic research for understanding the etiology and development of alcoholism. *J Consult Clin Psychol* 1994, **62**:1116–1126.
16. Sellers EM, Higgins GA, Sobell MB: 5-HT and alcohol abuse. *Trends Pharmacol Sci* 1992, **13**:69–75.
17. LeMarquand D, Pihl RO, Benkelfat C: Serotonin and alcohol intake, abuse, and dependence: findings of animal studies. *Biol Psychiatry* 1994, **36**:395–421.
18. Lesch KP, Bengel D, Heils A, Sabol SZ, Greenberg BD, Petri S, Benjamin J, Muller CR, Hamer DH, Murphy DL: Association of anxiety-related traits with a polymorphism in the serotonin transporter gene regulatory region. *Science* 1996, **274**:1527–1531.
19. Heils A, Teufel A, Petri S, Stober G, Riederer P, Bengel D, Lesch KP: Allelic variation of human serotonin transporter gene expression. *J Neurochem* 1996, **66**:2621–2624.
20. Heinz A, Jones DW, Mazzanti C, Goldman D, Ragan P, Hommer D, Linnoila M, Weinberger DR: A relationship between serotonin transporter genotype and in vivo protein expression and alcohol neurotoxicity. *Biol Psychiatry* 2000, **47**:643–649.
21. Little KY, McLaughlin DP, Zhang L, Livermore CS, Dalack GW, McFinton PR, DelProposto ZS, Hill E, Cassin BJ, Watson SJ, Cook EH: Cocaine, ethanol, and genotype effects on human midbrain serotonin transporter binding sites and mRNA levels. *Am J Psychiatry* 1998, **155**:207–213.
22. McHugh RK, Hofmann SG, Asnaani A, Sawyer AT, Otto MW: The serotonin transporter gene and risk for alcohol dependence: a meta-analytic review. *Drug Alcohol Depend*, **108**:1–6.
23. Feinn R, Nellissery M, Kranzler HR: Meta-analysis of the association of a functional serotonin transporter promoter polymorphism with alcohol dependence. *Am J Med Genet B Neuropsychiatr Genet* 2005, **133B**:79–84.
24. Parsian A, Cloninger CR: Serotonergic pathway genes and subtypes of alcoholism: association studies. *Psychiatr Genet* 2001, **11**:89–94.
25. Hallikainen T, Saito T, Lachman HM, Volavka J, Pohjalainen T, Ryyanen OP, Kauhanen J, Sivalahti E, Hietala J, Tiitonen J: Association between low

- activity serotonin transporter promoter genotype and early onset alcoholism with habitual impulsive violent behavior. *Mol Psychiatry* 1999, **4**:385–388.
26. Sander T, Harms H, Dufeu P, Kuhn S, Hoehle M, Lesch KP, Rommelspacher H, Schmidt LG: Serotonin transporter gene variants in alcohol-dependent subjects with dissocial personality disorder. *Biol Psychiatry* 1998, **43**:908–912.
 27. Ishiguro H, Saito T, Akazawa S, Mitushio H, Tada K, Enomoto M, Mifune H, Toru M, Shibuya H, Arinami T: Association between drinking-related antisocial behavior and a polymorphism in the serotonin transporter gene in a Japanese population. *Alcohol Clin Exp Res* 1999, **23**:1281–1284.
 28. Schuckit MA, Mazzanti C, Smith TL, Ahmed U, Radel M, Iwata N, Goldman D: Selective genotyping for the role of 5-HT2A, 5-HT2C, and GABA alpha 6 receptors and the serotonin transporter in the level of response to alcohol: a pilot study. *Biol Psychiatry* 1999, **45**:647–651.
 29. Nakamura M, Ueno S, Sano A, Tanabe H: The human serotonin transporter gene linked polymorphism (5-HTTLPR) shows ten novel allelic variants. *Mol Psychiatry* 2000, **5**:32–38.
 30. Hu X, Oroszi G, Chun J, Smith TL, Goldman D, Schuckit MA: An expanded evaluation of the relationship of four alleles to the level of response to alcohol and the alcoholism risk. *Alcohol Clin Exp Res* 2005, **29**:8–16.
 31. Hu XZ, Lipsky RH, Zhu G, Akhtar LA, Taubman J, Greenberg BD, Xu K, Arnold PD, Richter MA, Kennedy JL, et al: Serotonin transporter promoter gain-of-function genotypes are linked to obsessive-compulsive disorder. *Am J Hum Genet* 2006, **78**:815–826.
 32. Delbruck SJ, Wendel B, Grunewald I, Sander T, Morris-Rosendahl D, Crocq MA, Berrettini WH, Hoehle MR: A novel allelic variant of the human serotonin transporter gene regulatory polymorphism. *Cytogenet Cell Genet* 1997, **79**:214–220.
 33. Gelernter J, Kranzler H, Cubellis JF: Serotonin transporter protein (SLC6A4) allele and haplotype frequencies and linkage disequilibria in African- and European-American and Japanese populations and in alcohol-dependent subjects. *Hum Genet* 1997, **101**:243–246.
 34. Kunugi H, Hattori M, Kato T, Tatsumi M, Sakai T, Sasaki T, Hirose T, Nanko S: Serotonin transporter gene polymorphisms: ethnic difference and possible association with bipolar affective disorder. *Mol Psychiatry* 1997, **2**:457–462.
 35. Narita N, Narita M, Takashima S, Nakayama M, Nagai T, Okado N: Serotonin transporter gene variation is a risk factor for sudden infant death syndrome in the Japanese population. *Pediatrics* 2001, **107**:690–692.
 36. Barnes NM, Sharp T: A review of central 5-HT receptors and their function. *Neuropharmacology* 1999, **38**:1083–1152.
 37. Crabbe JC, Belknap JK, Buck KJ: Genetic animal models of alcohol and drug abuse. *Science* 1994, **264**:1715–1723.
 38. Saudou F, Amara DA, Dierich A, LeMeur M, Ramboz S, Segu L, Buhot MC, Hen R: Enhanced aggressive behavior in mice lacking 5-HT1B receptor. *Science* 1994, **265**:1875–1878.
 39. Crabbe JC, Phillips TJ, Feller DJ, Hen R, Wenger CD, Lessov CN, Schafer GL: Elevated alcohol consumption in null mutant mice lacking 5-HT1B serotonin receptors. *Nat Genet* 1996, **14**:98–101.
 40. Cigler T, LaForge KS, McHugh PF, Kapadia SU, Leal SM, Kreek MJ: Novel and previously reported single-nucleotide polymorphisms in the human 5-HT (1B) receptor gene: no association with cocaine or alcohol abuse or dependence. *Am J Med Genet* 2001, **105**:489–497.
 41. Fehr C, Grintschuk N, Szegedi A, Anghelescu I, Klawe C, Singer P, Hiemke C, Dahmen N: The HTR1B 861 G>C receptor polymorphism among patients suffering from alcoholism, major depression, anxiety disorders and narcolepsy. *Psychiatry Res* 2000, **97**:1–10.
 42. Lappalainen J, Long JC, Eggert M, Ozaki N, Robin RW, Brown GL, Naukkarinen H, Virkkunen M, Linnoila M, Goldman D: Linkage of antisocial alcoholism to the serotonin 5-HT1B receptor gene in 2 populations. *Arch Gen Psychiatry* 1998, **55**:989–994.
 43. Huang YY, Oquendo MA, Friedman JM, Greenhill LL, Brodsky B, Malone KM, Khait V, Mann JJ: Substance abuse disorder and major depression are associated with the human 5-HT1B receptor gene (HTR1B) G861C polymorphism. *Neuropsychopharmacology* 2003, **28**:163–169.
 44. Underwood MD, Mann JJ, Arango V: Serotonergic and noradrenergic neurobiology of alcoholic suicide. *Alcohol Clin Exp Res* 2004, **28**:575–695.
 45. Huang YY, Grailhe R, Arango V, Hen R, Mann JJ: Relationship of psychopathology to the human serotonin1B genotype and receptor binding kinetics in postmortem brain tissue. *Neuropsychopharmacology* 1999, **21**:238–246.
 46. Kranzler H, Lappalainen J, Nellissery M, Gelernter J: Association study of alcoholism subtypes with a functional promoter polymorphism in the serotonin transporter protein gene. *Alcohol Clin Exp Res* 2002, **26**:1330–1335.
 47. Gorwood P, Aissi F, Batel P, Ades J, Cohen-Salmon C, Hamon M, Boni C, Lanfumey L: Reappraisal of the serotonin 5-HT(1B) receptor gene in alcoholism: of mice and men. *Brain Res Bull* 2002, **57**:103–107.
 48. Goldstein RB, Dawson DA, Saha TD, Ruan WJ, Compton WM, Grant BF: Antisocial behavioral syndromes and DSM-IV alcohol use disorders: results from the National Epidemiologic Survey on Alcohol and Related Conditions. *Alcohol Clin Exp Res* 2007, **31**:814–828.
 49. Ross HE, Glaser FB, Germanson T: The prevalence of psychiatric disorders in patients with alcohol and other drug problems. *Arch Gen Psychiatry* 1988, **45**:1023–1031.
 50. Hesselbrock VM, Hesselbrock MN, Stabenau JR: Alcoholism in men patients subtyped by family history and antisocial personality. *J Stud Alcohol* 1985, **46**:59–64.
 51. van den Bree MB, Sviki DS, Pickens RW: Genetic influences in antisocial personality and drug use disorders. *Drug Alcohol Depend* 1998, **49**:177–187.
 52. Hwu HG, Yeh EK, Chang LY: Prevalence of psychiatric disorders in Taiwan defined by the Chinese Diagnostic Interview Schedule. *Acta Psychiatr Scand* 1989, **79**:136–147.
 53. Endicott J, Spitzer RL: A diagnostic interview: the schedule for affective disorders and schizophrenia. *Arch Gen Psychiatry* 1978, **35**:837–844.
 54. Huang SY, Lin WW, Ko HC, Lee JF, Wang TJ, Chou YH, Yin SJ, Lu RB: Possible interaction of alcohol dehydrogenase and aldehyde dehydrogenase genes with the dopamine D2 receptor gene in anxiety-depressive alcohol dependence. *Alcohol Clin Exp Res* 2004, **28**:374–384.
 55. Goodwin RD, Hamilton SP: Lifetime comorbidity of antisocial personality disorder and anxiety disorders among adults in the community. *Psychiatry Res* 2003, **117**:159–166.
 56. Hesselbrock V, Stabenau J, Hesselbrock M, Mirkin P, Meyer R: A comparison of two interview schedules. The schedule for affective disorders and schizophrenia-Lifetime and the National Institute for Mental Health Diagnostic Interview Schedule. *Arch Gen Psychiatry* 1982, **39**:674–677.
 57. Rogers R, Cunnien AJ: Multiple schedule of affective disorders and schizophrenia (SADS) in the assessment of criminal defendants. *J Forensic Sci* 1986, **31**:222–230.
 58. Maron E, Tammiste A, Kallassalu K, Eller T, Vasar V, Nutt DJ, Metspalu A: Serotonin transporter promoter region polymorphisms do not influence treatment response to escitalopram in patients with major depression. *Eur Neuropsychopharmacol* 2009, **19**:451–456.
 59. Wendland JR, Martin BJ, Kruse MR, Lesch KP, Murphy DL: Simultaneous genotyping of four functional loci of human SLC6A4, with a reappraisal of 5-HTTLPR and rs25531. *Mol Psychiatry* 2006, **11**:224–226.
 60. Bowirrat A, Oscar-Berman M: Relationship between dopaminergic neurotransmission, alcoholism, and Reward Deficiency syndrome. *Am J Med Genet B Neuropsychiatr Genet* 2005, **132B**:29–37.
 61. Faul F, Erdfelder E, Buchner A, Lang AG: Statistical power analyses using G*Power 3.1: tests for correlation and regression analyses. *Behav Res Methods* 2009, **41**:1149–1160.
 62. Faul F, Erdfelder E, Lang AG, Buchner A: G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods* 2007, **39**:175–191.
 63. Lyons-Ruth K, Holmes BM, Sasvari-Szekely M, Ronai Z, Nemoda Z, Pauls D: Serotonin transporter polymorphism and borderline or antisocial traits among low-income young adults. *Psychiatr Genet* 2007, **17**:339–343.
 64. Reif A, Rosler M, Freitag CM, Schneider M, Eujen A, Kissling C, Wenzler D, Jacob CP, Retz-Junginger P, Thome J, et al: Nature and nurture predispose to violent behavior: serotonergic genes and adverse childhood environment. *Neuropsychopharmacology* 2007, **32**:2375–2383.
 65. Sadeh N, Javdani S, Jackson JJ, Reynolds EK, Potenza MN, Gelernter J, Lejuez CW, Verona E: Serotonin transporter gene associations with psychopathic traits in youth vary as a function of socioeconomic resources. *J Abnorm Psychol* 2010, **119**:604–609.
 66. Lu RB, Lee JF, Huang SY, Lee SY, Chang YH, Kuo PH, Chen SL, Chen SH, Chu CH, Lin WW, et al: Interaction between ALDH2*1*1 and DRD2/ANKK1 TaqI A1A1 genes may be associated with antisocial personality disorder not co-morbid with alcoholism. *Addict Biol*.
 67. Baumgarten HG, Grozdanovic Z: Psychopharmacology of central serotonergic systems. *Pharmacopsychiatry* 1995, **28**(Suppl 2):73–79.

68. Parsons LH, Weiss F, Koob GF: **Serotonin1b receptor stimulation enhances dopamine-mediated reinforcement.** *Psychopharmacology (Berl)* 1996, **128**:150–160.
69. Walsh SL, Cunningham KA: **Serotonergic mechanisms involved in the discriminative stimulus, reinforcing and subjective effects of cocaine.** *Psychopharmacology (Berl)* 1997, **130**:41–58.
70. Hallbus M, Magnusson T, Magnusson O: **Influence of 5-HT1B/1D receptors on dopamine release in the guinea pig nucleus accumbens: a microdialysis study.** *Neurosci Lett* 1997, **225**:57–60.
71. Yadid G, Pacak K, Kopin IJ, Goldstein DS: **Endogenous serotonin stimulates striatal dopamine release in conscious rats.** *J Pharmacol Exp Ther* 1994, **270**:1158–1165.
72. Cesura AM, Pletscher A: **The new generation of monoamine oxidase inhibitors.** *Prog Drug Res* 1992, **38**:171–297.
73. Keung WM, Vallee BL: **Daidzin: a potent, selective inhibitor of human mitochondrial aldehyde dehydrogenase.** *Proc Natl Acad Sci U S A* 1993, **90**:1247–1251.
74. Wang TJ, Huang SY, Lin WW, Lo HY, Wu PL, Wang YS, Wu YS, Ko HC, Shih JC, Lu RB: **Possible interaction between MAOA and DRD2 genes associated with antisocial alcoholism among Han Chinese men in Taiwan.** *Prog Neuropsychopharmacol Biol Psychiatry* 2007, **31**:108–114.
75. Goldman N, Gleit DA, Lin YH, Weinstein M: **The serotonin transporter polymorphism (5-HTTLPR): allelic variation and links with depressive symptoms.** *Depress Anxiety* 2010, **27**:260–269.

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