**RAPID COMMUNICATION**

**ALCOHOL DEHYDROGENASE POLYMORPHISMS IN NATIVE AMERICANS: IDENTIFICATION OF THE ADH2*3 ALLELE**

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**Abstract** — Alcohol dehydrogenase (ADH) polymorphisms were evaluated among 95 Native American Mission Indians. Approximately equal frequencies of ADH3*1 and ADH3*2 alleles were found. Twelve individuals were heterozygous for ADH2*3, an allele previously identified only in persons of African origin. None of the individuals with ADH3V alleles was of purely Native American descent, although none had known African ancestry. These results suggest that these candidate genes deserve broader study among Native Americans and may provide increased understanding of the likely polygenic contributions to alcohol-related disorders in this population.

**INTRODUCTION**

The major enzymes involved in alcohol metabolism, alcohol dehydrogenase (ADH) and mitochondrial aldehyde dehydrogenase (ALDH2), both exist as multiple isoenzymes that differ in their kinetic properties. Because the frequency of these isoenzymes varies across population groups, their genes have been suggested as candidate genes that are likely to contribute to ethnic differences in alcohol and acetaldehyde degradation, variability in response to alcohol, and differential vulnerability for developing alcoholism and alcohol-related illness (Bosron et al., 1993).

The inactive form of the mitochondrial enzyme, encoded by the ADH2*2 allele, is associated with elevated acetaldehyde levels, an alcohol-induced flushing reaction, and lower rates of alcohol use and alcoholism in individuals of Asian heritage with this gene mutation (Takeshita et al., 1994; Higuchi et al., 1995). ADH is polymorphic at two loci, ADH2 and ADH3. There are three alleles at the ADH2 loci and two alleles at the ADH3 loci with distinct differences in the kinetic properties of the isoenzymes that form from the associations of the subunit proteins encoded at these loci (Bosron and Li, 1987).

Prior to the ability to genotype subjects for ADH2, ADH3 and ALDH2, some investigators reported that Native American Indians exhibit ALDH enzyme deficiency, alcohol-induced flushing, and other physiological reactions to alcohol that are similar to those of Asians (Wolff, 1973; Zeiner et al., 1976, 1985; Goedde et al., 1986). This apparent similarity in alcohol responsivity, however, is difficult to reconcile with the findings that Native Americans have some of the highest and Asians some of the lowest reported rates of alcoholism (Brod, 1975; Helzer et al., 1990). These studies prompted subsequent investigations genotyping for variation in the alcohol metabolizing enzymes in several Native American Indian groups (Rex et al., 1985; Bosron et al., 1988; O'Dowd et al., 1990; Chen et al., 1992; Dyck, 1993; Novoradovsky et al., 1995). The results from these studies indicate that the distribution of ADH2, ADH3 and ALDH2 alleles in previously studied Native Americans appears to be more...
Table 1. Distribution of alcohol dehydrogenase (ADH) alleles in 95 Native American Mission Indians

<table>
<thead>
<tr>
<th>ADH2*1 (β1)</th>
<th>ADH2*2 (β2)</th>
<th>ADH2*3 (β3)</th>
<th>ADH3*1 (γ1)</th>
<th>ADH3*2 (γ2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>177</td>
<td>1</td>
<td>12</td>
<td>107</td>
</tr>
<tr>
<td>Frequency</td>
<td>0.932</td>
<td>0.005</td>
<td>0.063</td>
<td>0.563</td>
</tr>
</tbody>
</table>

similar to that of Caucasians than to that of Asians. The present study was conducted to determine ADH2 and ADH3 allele frequencies in Mission Indians from southern California, a Native American group with a high prevalence of alcoholism and alcohol-related disorders.

METHODS

The subjects were 95 Native American Mission Indians who were recruited from reservations in southern California as part of a larger study. All subjects provided written informed consent (approved by The Scripps Clinic and Research Foundation human subjects committee) prior to participation. All were unrelated as first-degree relatives. Subjects completed a structured self-report instrument that gathered information including demography and usual quantity and frequency of alcohol consumption over the previous 6 months. As determined by self-report of their Bureau of Indian Affairs status, all subjects were at least 12.5% Native American heritage. A blood sample was drawn from each subject and leukocyte DNA was extracted for genotyping. The relevant portions of the ADH loci were amplified using the polymerase chain reaction and allele-specific oligonucleotide primers followed by hybridization with radiolabelled oligonucleotide probes (Xu et al., 1988). Relationships among percent Native American heritage, ADH2 genotype, ADH3 genotype, and drinking history variables were indexed using Pearson correlation coefficients.

RESULTS

Demographic characteristics revealed that the average Native American heritage for the subjects was 45% (range, 12.5–100%), but only one subject was of purely Native American descent. Other ethnicities identified included: Latino (primarily Mexican, 55 (58%); Caucasian (primarily European), 42 (44%); Asian (Filipino), 1 (1%); African, 1 (1%); and other, but unknown, 12 (13%). Percentages exceed 100 because some subjects reported more than one other ethnic heritage.

ADH2 genotyping indicated that twelve subjects had ADH2*1/2*2 genotype, one subject had ADH2*1/2*2 genotype, and the remaining subjects had ADH2*1/2*1 genotype. This is the first report of the ADH2*3 allele in individuals not of African heritage. Although none of the individuals with ADH2*3 alleles were of purely Native American descent, none had any known African ancestry. Five of the twelve subjects with the ADH2*2 allele reported that they were of mixed Mexican heritage, six reported that they were of mixed Caucasian heritage, and two reported that they were also of other, but unknown, heritage.

ADH3 genotyping indicated that 37 subjects had ADH3*1/3*1 genotype, 33 subjects had ADH3*1/3*2 genotype, and 25 subjects had ADH3*2/3*2 genotype. The distribution of ADH3 alleles is similar to previously studied Native American tribes, particularly Navajo and Sioux (Rex et al., 1985; Bosron et al., 1988), as well as Caucasian populations (Goedde et al., 1992). Table 1 provides the ADH allele frequencies in this population.

Drinking history variables revealed that the subjects drank alcohol in the prior 6 months on an average of 5.7 ± 6.9 days per month (range, 1–30) and consumed 5.4 ± 5.0 drinks per occasion (range, 1–30). As shown in Table 2, quantity and frequency of drinking over the previous 6 months did not correlate significantly with percentage of Native American heritage, ADH2 genotype, or ADH3 genotype. Thus, in this sample, it does not appear that there are distinct phenotypic differences in recent alcohol use between individuals of
Table 2. Correlations between genetic variables and drinking history variables in 95 Native American Mission Indians

<table>
<thead>
<tr>
<th>Variable</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Native American</td>
<td>-0.15</td>
<td>0.03</td>
<td>0.13</td>
<td>0.11</td>
<td>0.16</td>
<td></td>
</tr>
<tr>
<td>ADH2 genotype</td>
<td>-</td>
<td>-0.23*</td>
<td>0.04</td>
<td>0.12</td>
<td>0.07</td>
<td></td>
</tr>
<tr>
<td>ADH3 genotype</td>
<td>-</td>
<td>-0.05</td>
<td>0.01</td>
<td>-0.09</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alcohol frequency (days/month)</td>
<td></td>
<td>0.48*</td>
<td>0.70*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alcohol quantity (drinks/occasion)</td>
<td></td>
<td></td>
<td>0.78*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alcohol frequency x quantity</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-</td>
</tr>
</tbody>
</table>

*P <0.05 (two-tailed test).

**DISCUSSION**

The catalytic differences among the isoenzymes encoded by the ADH2 and ADH3 alleles have led to interest in their distribution and their influence on drinking behaviour and development of alcoholism and alcohol-related disease. Relevant to the present findings, ADH2*3 alleles have been associated with fetal alcohol syndrome (Sokol et al., 1989) and with faster alcohol metabolism in individuals of African heritage (Thomasson et al., 1995) and ADH3*1 alleles have been associated with alcoholic liver disease in a Caucasian population (Day et al., 1991). Native Americans, as a group, have higher rates of fetal alcohol syndrome, alcoholism, and alcoholic liver disease than other population groups in the United States (Indian Health Service, 1993). There have also been reports that Native Americans metabolize alcohol differently (generally faster) than other ethnic groups (Schaefer, 1981; Reed, 1985). Future work will determine whether ADH polymorphisms contribute to differences in alcohol metabolism or response to alcohol in this heterogeneous population. These results indicate that larger studies examining the relationship between ADH gene variants and alcohol-related behaviours in Native Americans deserve broader study and might help to explain, in part, the likely polygenic contributions to the high prevalence of alcoholism and other alcohol-related health problems in this population.

**REFERENCES**


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Tackmann, D., Singh, S., Beckmann, G., Bhatia, K.,
Chen, L. Z., Fang, B., Lisker, R., Paik, Y. K.,
Rothhammer, F., Saha, N., Segal, B., Srivastava, L.
M. and Czeizel, A. (1992) Distribution of ADH2 and
ALDH2 genotypes in different populations. Human
Genetics 88, 344–346.

Helzer, J. E., Canino, G. J., Yeh, E.-K., Bland, R. C,
Alcoholism — North America and Asia. Archives of
General Psychiatry 47, 313–319.

Higuchi, S., Matsushita, S., Murayama, M., Takagi, T.
dehydrogenase polymorphisms and the risk for
alcoholism. American Journal of Psychiatry 152,
1219–1221.

Indian Health Service (1993) Trends in Indian Health —
1993. Department of Health and Human Services,
Washington, DC.

Novoradovsky, A. G., Kidd, J., Kidd, K. and Goldman,
D. (1995) Apparent monomorphisms of ALDH2 in
seven American Indian populations. Alcohol 12,
163–167.

Genotyping of mitochondrial aldehyde dehydrogen-
ase locus of Native American Indians. Alcoholism:
Clinical and Experimental Research 14, 531–533.

Reed, T. E. (1985) Ethnic differences in alcohol use,
abuse, and sensitivity: a review with genetic

Rex, D. K., Bosron, W. F., Smialek, J. E. and Li T.-K.
(1985) Alcohol and aldehyde dehydrogenase iso-
enzymes in North American Indians. Alcoholism:
Clinical and Experimental Research 9, 851–855.

of findings and some new directions. Journal of
Studies on Alcohol 42, 99–117.

Sokol, R. J., Smith, M., Ernhart, C. B., Baumann, R.,
Marrier, S. S., Ager, J. W. and Morrow-Tlucak, M.
(1989) A genetic basis for alcohol-related birth
defects? Alcoholism: Clinical and Experimental
Research 13, 343.

Takeshita, T., Morimoto, K., Mao, X. Q., Hashimoto, T.
and Furuyama, J.-l. (1994) Characterization of the
closest genotypes of low Km aldehyde dehydrogenase
in a Japanese population. Human Genetics 94, 217–
223.

ADH2 gene polymorphism are determinants of
alcohol pharmacokinetics. Alcoholism: Clinical and
Experimental Research 19, 1494–1499.

Wolff, P. H. (1973) Vasomotor sensitivity to alcohol in
diverse mongoloid populations. American Journal of
Human Genetics 25, 193–199.

Xu, Y., Carr, L. G., Bosron, W. F., Li, T.-K. and
dehydrogenase at the ADH2 and ADH3 loci
following DNA sequence amplification. Genomics
2, 209–214.

Physiologic responses to ethanol among Tarahumara
Indians. Annals of the New York Academy of
Sciences 273, 151–158.

Zeiner, A. R., Girardot, J. M., Jones-Saumty, D. and
among American Indians in Oklahoma. Japanese
Journal of Alcohol and Drug Dependence 20, 359–
366.