ELSEVIER

Atherosclerosis 184 (2006) 397–403

ATHEROSCLEROSIS

www.elsevier.com/locate/atherosclerosis

Lack of association between ADH3 polymorphism, alcohol intake, risk factors and carotid intima-media thickness

P. Marques-Vidal [a], C. Bal Dit Sollier [b], L. Drouet [b], H. Boccalon [c], J.B. Ruidavets [a], J. Ferrières [a,*]

[a] *INSERM U558, Faculté de Médecine, 37 Allées Jules Guesde, 31073 Toulouse cedex, France*
[b] *Department of Hematology, Hôpital Lariboisière, Paris, France*
[c] *Department of Vascular Medicine, CHU Rangueil, Toulouse, France*

Received 7 January 2005; received in revised form 28 April 2005; accepted 4 May 2005
Available online 6 June 2005

Abstract

Objective: We assess the relationships between alcohol dehydrogenase 3 (ADH3) polymorphism, alcohol consumption and cardiovascular risk factor levels.
Methods: In a representative population sample from Southwestern France (614 men, 567 women, age 49.7 ± 8.5 years), alcohol intake was assessed by questionnaire.
Results: Alcohol consumption was significantly related with higher levels of total and HDL cholesterol, triglycerides, apolipoprotein A-I in men and with higher levels of HDL cholesterol in women. Also, an inverse relationship between alcohol consumption and intima-media thickness was found in men. Conversely, in both genders, no differences were found between ADH3 genotypes regarding all cardiovascular risk factors studied and carotid intima-media thickness. Also, in both genders, no significant ADH3 × alcohol interaction was found for all variables, and further adjustment on age, body mass index, educational level, smoking status or after excluding subjects on hypolipidemic or antihypertensive drug treatment did not change the results.
Conclusion: We found no interaction between the ADH3 polymorphism and alcohol intake on cardiovascular risk factor levels and atherosclerotic markers in Southwestern France.

Keywords: Alcohol dehydrogenase; Polymorphism; Population sample; HDL; Alcohol consumption

1. Introduction

Moderate alcohol consumption has been shown to be protective against cardiovascular disease [1–4]. The major enzymes of alcohol metabolism are alcohol dehydrogenase (ADH) and aldehyde dehydrogenase (ALDH). ADH is a dimeric protein with two subunits and six ADH genes have been described, of which two (ADH2 and ADH3) are polymorphic. The ADH2 polymorphism has been shown to be related to cardiovascular risk factors, namely systolic blood pressure [5,6] and triglycerides [5]. The ADH3 polymorphism has two alleles, gamma 1 and gamma 2 (γ1 and γ2), and pharmacokinetic studies have shown that the homodimeric γ1 isoenzyme has a 2.5-fold higher maximal velocity of ethanol oxidation than the homodimeric γ2 isoenzyme [7]. Further, it has been shown that moderate drinkers who are homozygous for the slow-oxidizing ADH3 allele have higher HDL levels and a substantially decreased risk of myocardial infarction [8]. Still, those relationships between ADH polymorphisms and cardiovascular disease have been questioned [9–11], and it is currently unclear whether ADH polymorphisms do interact with alcohol consumption regarding the levels of cardiovascular risk factors or cardiovascular disease. France is characterized by a low incidence of myocardial infarction and a moderate-high intake of alcohol but, to our knowledge,

* Corresponding author. Tel.: +33 5 61 52 18 70; fax: +33 5 61 32 33 32.
E-mail address: jean.ferrieres@cict.fr (J. Ferrières).

0021-9150/$ – see front matter © 2005 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.atherosclerosis.2005.05.003

no study on the relationship between ADH3 polymorphism and alcohol consumption on the levels of cardiovascular risk factors has ever been conducted.

Thus, we used the data from the last MONICA (multinational monitoring of trends and determinants in cardiovascular diseases) population survey conducted in Southwestern France to assess the relationships between ADH3 polymorphisms, alcohol intake and cardiovascular risk factor levels. The relationships between ADH3 polymorphisms, alcohol intake and intima-media thickness, an indicator of preclinical atherosclerosis, were also assessed.

2. Population and methods

2.1. Population sample

The WHO–MONICA Project is a study that monitors deaths due to coronary heart disease, myocardial infarction (MI), coronary care and risk factors in men and women aged 35 to 64 years [12–14]. It consists of 39 MONICA Collaborative Centers (MCC) in 26 countries. Each MCC is in charge of carrying out two or three population surveys on cardiovascular risk factors in the beginning and at the end of the 10-year, and possibly in the middle of the study period. The sampling strategy was to have representative probability samples within each sex and 10-year age group, at least for the age range 35–64 years. The number of eligible subjects asked to participate was 50% higher than the required number in order to obtain the necessary quota of 200 persons for each sex and 10-year age group (allowing for subjects refusing to participate or not participating for any other reason). The informed consent to participate in the study was obtained from the subjects before the survey. The last Toulouse MONICA survey started in December 1994 and ended in July 1997 and concerned both genders; participation rates were 67% for men and 59% for women.

2.2. Data collection

Subjects were advised to restrain from physical exercising, smoking, eating or drinking anything other than water for at least one hour prior to the screening visit. Screening included standardized questionnaires on personal data and measurements of body height and weight and blood pressure. Body mass index (BMI) was calculated as weight (kg) height $(m)^{-2}$. Subjects were considered as smokers if they currently smoked, and the average amount of cigarettes smoked per day was assessed for each smoker. Subjects were considered as hypertensive if their systolic blood pressure was >160 mmHg and/or their diastolic blood pressure was >95 mmHg and/or they reported being on antihypertensive drug treatment.

2.3. Biological measurements

Lipids were measured on plasma EDTA plasma samples using automated enzymatic assays (Boehringer, Mannheim, Germany). External quality control by the MONICA reference laboratory indicated no relevant deviation. Subjects were considered as dyslipidaemic if their total cholesterol was >6.5 mmol/L and/or they reported being on hypolipidaemic drug treatment.

2.4. Intima-media thickness measurement

High-resolution B-mode ultrasonography was used to detect atherosclerotic plaques in carotid and femoral arteries and to measure common carotid IMT. An ATL UM9 system (Advanced Technology Laboratories Ultramark 9 High Definition Imaging) was used with a 7.4 MHz transducer. IMT was defined as the distance between the media ± adventitia interface and the lumen ± intima interface, avoiding the sites of plaque [15,16]. IMT was measured on the right and left common carotid arteries, on the far wall exclusively: three points at two locations on each artery, proximal and middle. Thus, 12 points were determined. The mean values at the 12 sites were combined to generate an overall mean value for common carotid IMT. In 26 subjects submitted to a second IMT assessment, the differences between the mean values of the measurements performed during the two examinations were small (−0.007 mm for the right common carotid artery and −0.022 mm for the left common carotid artery) and the intra-class correlation coefficients between the two measurements were 0.64 ($p < 0.001$) for the right common carotid artery and 0.53 ($p < 0.001$) for the left common carotid artery.

2.5. Alcohol consumption

Alcohol consumption was assessed by a validated questionnaire that recorded the subjects mean consumption (in units) of wine, beer, cider and spirits for each day of the week [17,18]. Intake of alcohol (expressed in ml of pure ethanol/week) was estimated from the average number of millilitres of ethanol in one unit of each type of alcoholic beverage: wine (10% or 12% alcohol, v/v) = 12 cl serving; beer (5% alcohol) = 12 cl serving; beer (6 or 8% alcohol) = 25 or 33 cl serving; cider (5% alcohol) = 12 cl serving; spirits (20% or 40% alcohol) = 2 or 6 cl serving. For each gender, alcohol consumption was further classified into three groups: teetotalers, below and above the median.

2.6. Alcohol dehydrogenase genotyping

DNA was obtained from leukocytes by salting out procedure, and ADH3 genotyping was performed in duplicate for each subject using polymerase chain reaction. Subjects were classified into three categories according to the presence of the γ2 (slow) allele: 11 homozygotes, 12 heterozygotes and 22 heterozygotes. The alcohol dehydrogenase 3 exon eight polymorphism was determined as followed:

A fragment of 130 bp in the ADH3 gene was amplified by polymerase chain reaction (PCR) using the following primers (forward 5′-GCTTTAAGAGTAAATCTGTCCCC-