ELSEVIER

Journal of Hepatology 43 (2005) 1055–1059

Journal of Hepatology

www.elsevier.com/locate/jhep

Phenotypic expression in detected C282Y homozygous women depends on body mass index

Fabrice Lainé[1,*], Anne-Marie Jouannolle[2], Jeff Morcet[1], Alain Brigand[3], Michel Pouchard[4], Bernard Lafraise[5], Jean Mosser[6], Véronique David[2,6], Yves Deugnier[1,7]

[1]*Centre d'Investigation Clinique INSERM 0203, CHU Pontchaillou, 35033 Rennes, France*
[2]*Laboratoire de Génétique Moléculaire et d'Hormonologie, CHU Pontchaillou, 35033 Rennes, France*
[3]*Centre d'Examen de Santé des Côtes d'Armor, 22000 Saint Brieuc, France*
[4]*Centre d'Examen de Santé d'Ille et Vilaine, 35000 Rennes, France*
[5]*Centre d'Examen de Santé de Loire Atlantique, 44000 Saint Nazaire, France*
[6]*UMR 6061 CNRS, Faculté de Médecine, 35000 Rennes, France*
[7]*Service des Maladies du Foie, CHU Pontchaillou, 35000 Rennes, France*

***Background/Aims*: In HFE-related haemochromatosis, a large proportion of C282Y homozygotes, especially women, are not detected by phenotypic screening using transferrin saturation. The aim of this study was to identify the clinical and biochemical factors associated with non-expression of the disease as defined as transferrin saturation <45%.**

***Methods*: The study was performed in 78 (57 women and 21 men) C282Y homozygotes identified through a large-scale screening program conducted on 19,644 French subjects. Biometric, clinical and biochemical variables including those susceptible to influence body iron stores were tested for association with transferrin saturation levels <45%.**

***Results*: Non-expression was observed in 26/57 (46%) women and in 5/21 (24%) men. At multivariate analysis, female gender (OR: 16.5, 95%CI 1.8–146.5; $P=0.012$), body mass index (OR: 1.21, 95%CI 1.02–1.44; $P=0.027$), haemoglobin levels (OR: 0.88, 95%CI 0.81–0.97; $P=0.012$) and serum ferritin levels (OR: 0.99, 95%CI 0.98–1.00; $P=0.007$) were significantly and independently associated with a non-expressing phenotype.**

***Conclusions*: Excess body mass is commonly associated with the lack of phenotypic expression in detected C282Y homozygotes. This should be kept in mind with respect to the design and cost-effectiveness of phenotypic screening programs for haemochromatosis.**

***Keywords*: Body mass index; Transferrin saturation; Screening; Haemochromatosis**

1. Introduction

Homozygosity for the C282Y mutation on the HFE gene accounts for more than 90% of cases of genetic haemochromatosis (GH) in Caucasians [1]. However, the penetrance of this genotype remains rather low as stressed by Beutler et al. [2]. Both genetic factors and acquired conditions are likely to modulate the expressivity of HFE-GH. With respect to acquired factors, dietary habits, blood donation, gender-related events (pregnancy, menopause and mode of contraception) and associated disorders such as digestive malabsorption or blood loss, have been anecdotally reported to influence phenotypic expression in haemochromatotic subjects but have not been extensively studied in C282Y homozygotes [3]. The present study was aimed at identifying the clinical and biological factors associated with non-expression of C282Y homozygosity in subjects diagnosed through a systematic genetic screening program conducted on 19,644 French subjects from general population. Non-expression was defined as a transferrin saturation (TS) lower than 45%, which corresponds to the threshold commonly used in phenotypic GH screening [4,5].

Received 1 March 2005; received in revised form 13 April 2005; accepted 25 May 2005; available online 28 June 2005

* Corresponding author. Tel.: +33 2 9928 4297; fax: +33 2 9928 4112.
E-mail address: fabrice.laine@chu-rennes.fr (F. Lainé).

0168-8278/$30.00
doi:10.1016/j.jhep.2005.05.027

2. Subjects and methods

From 1998 to 2003, a large-scale genotypic screening for HFE-GH was performed on 19,644 Caucasian subjects, aged from 25 to 70 years, who attended the Health Appraisal Centres of Rennes, Saint-Brieuc and Saint-Nazaire (Brittany, France). The present study was conducted on the ninety unrelated subjects (60 women and 30 men) who were identified as C282Y homozygotes through this program whose design and preliminary results have been previously published [6]. Twelve subjects were excluded because of the lack of TS determination or previous venesection therapy. Among the remaining 78 subjects, there were 21 men and 57 women of whom 5 (24%) and 26 (46%), respectively, were considered as non-expressing on the basis of a TS level lower than 45%.

2.1. Clinical data

The following clinical data were recorded in all subjects using the same questionnaire: age, dietary habits, medical history including past and present symptoms such as fatigue, arthralgias and diarrhoea, current medications, alcohol consumption and history of blood donation. In addition, number of pregnancies, use of oral contraception or of intrauterine device, and menstrual history were collected in women. Body mass index (BMI—weight/height^2 as kg/m^2) and waist circumference were recorded. Abdominal obesity was defined as waist circumference >88 cm in women and >102 cm in men [7].

2.2. Laboratory tests

Samples for laboratory tests including serum iron, TS, ferritin, alanine amino transferase, gamma-glutamyl transpeptidase, glucose, total cholesterol, triglycerides and blood cell count were drawn from fasting subjects between 8.00 and 10.00 a.m. Biochemical assays were performed in each Health Appraisal Centre using the same technical process. Intra and interlaboratory control of quality was performed using ASQUALAB™. TS were determined on the basis of serum iron and serum transferrin assayed by immunoturbidimetric method (Kone instrument). Serum ferritin was determined using an immunoenzymatic method (KitAbbot, Milford, CT, USA Imx B22192).

2.3. Statistical analysis

Subjects were divided into two groups according to a transferrin saturation threshold of 45%. Those with TS $<45\%$ are referred as non-expressing homozygotes along the manuscript. Univariate analyses were performed in which the data were compared between expressing and non-expressing groups, using the t test (or Wilcoxon test as appropriate) for continuous variables and chi square test for categorical variables (or Fisher's exact test as appropriate). Only variables with $P<0.1$ were introduced into multivariate analyses which were performed using a forward logistic regression model to estimate the odds ratio (OR) of being non-expressing (along with a 95% confidence interval (CI(). A P value <0.05 was considered as significant. All statistical analyses were performed using SAS software ™ (Florida, USA).

All subjects gave a written consent after full information by a medical doctor. The study was approved by the Local Committee of Ethics of Rennes.

3. Results

3.1. Whole population

3.1.1. Characteristics of subjects

Main clinical and biochemical characteristics of the *78* C282Y homozygous subjects are summarized in Table 1. Mean age was 44.2 years (range: 25–70). Twenty-four subjects (30.7%) had BMI >25 kg/m^2 of whom six were obese (BMI >30 kg/m^2) and eight had abdominal obesity. Only one subject was considered as excessive alcohol drinker. None was vegetarian and none was diabetic nor had clinical evidence of digestive blood loss, malabsorption or inflammation.

3.2. Factors associated with non-expression

3.2.1. Univariate analysis

As shown in Table 1, abdominal obesity was significantly and positively associated with non-expression of C282Y homozygosity while serum iron, serum ferritin, haemoglobin levels, and mean globular volume were significantly and negatively associated with non-expression. Age, genital events, blood donation, alcohol consumption and white cell count did not differ significantly between the expressing and non-expressing subjects. There was a trend for a positive association of female gender and elevated BMI with non-expression.

3.2.2. Multivariate analysis

All non-collinear variables associated with non-expression of C282Y homozygosity with a P value ≤ 0.1 at univariate analysis (gender, body mass index, abdominal obesity, blood donation, serum ferritin and haemoglobin levels, and mean globular volume) were entered into multivariate analysis. Female gender, decrease of serum ferritin, decrease of haemoglobin level and increase of BMI were independently associated with non-expression (Table 2) even after adjustment on age.

Because gender was identified by multivariate analysis as a strong independent factor associated with phenotypic expression, and because putative causes of blood loss are different between women and men, analyses were performed in men and women, separately.

3.3. Male subjects

Univariate analysis showed that non-expressing men had significantly lower serum ferritin (182 ± 134 vs. 621 ± 435 μg/l; $P=0.007$) and lower haemoglobin levels (147 ± 8 vs. 155 ± 6 g/l; $P=0.05$) than expressing men. There was no significant difference between expressing and non-expressing men regarding age (52.6 ± 15 vs 38.4 ± 12; $P=0.07$), BMI (20.8 ± 2.7 vs. 24.2 ± 3.4; $P=0.08$) and waist circumference (75 ± 8 vs. 84 ± 9; $P=0.15$).

However, these results must be kept with caution due to the small number of non-expressing men which precluded any multivariate analysis.

3.4. Female subjects

Characteristics of the 57 homozygous women are summarized in Table 3. Eleven (19.3%) women were