Nutrition 21 (2005) 240–248

www.elsevier.com/locate/nut

Basic nutritional investigation

Changes in lipid metabolism and antioxidant defense status in spontaneously hypertensive rats and Wistar rats fed a diet enriched with fructose and saturated fatty acids

Aurélie Girard, M.D.[a], Sihem Madani, Ph.D.[a], Es Saddik El Boustani, Ph.D.[b], Jacques Belleville, Ph.D.[a], Josiane Prost, Ph.D.[a,*]

[a] *Université de Bourgogne, UPRES Lipides Nutrition EA 2422, Faculté des Sciences Gabriel, Dijon, France*
[b] *Nutrition et Santé, Université Caddi Ayyad, Faculté des Sciences Semlalia, Marrakech, Morocco*

Manuscript received March 13, 2003; accepted April 20, 2004.

Abstract

Objective: Larger doses of fructose and saturated fat have been associated with oxidative stress and development of hypertension. The effects of modest amounts of fructose and saturated fatty acids on oxidative stress are unknown.

Methods: To increase knowledge on this question, 10-wk-old spontaneously hypertensive rats and Wistar rats were fed for 8 wk with a control diet or an experimental diet enriched with fructose (18%) and saturated fatty acids (11%; FS diet). The total antioxidant status of organs and red blood cells was assayed by monitoring the rate of free radical-induced red blood cell hemolysis. Sensitivity of very low-density lipoprotein and low-density lipoprotein (VLDL-LDL) to copper-induced lipid peroxidation was determined as the production of thiobarbituric acid-reactive substances. Antioxidant enzymes and vitamins were also measured to establish the oxidative stress effect.

Results: The FS diet did not affect blood pressure in either strain, but it increased plasma insulin concentrations only in Wistar rats without affecting those of glucose of either strain. The FS diet significantly enhanced plasma and VLDL-LDL triacylglycerol concentrations without affecting concentrations of VLDL-LDL thiobarbituric acid-reactive substances. The decreased content of arachidonic acid and total polyunsaturated fatty acids in VLDL-LDL by the FS diet may have prevented lipid peroxidation in this fraction. Moreover, FS consumption by both strains was accompanied by a significant increase in total antioxidant capacity of adipose tissue, muscle, heart, and liver. This may have resulted from increased tissue ascorbic acid levels and glutathione peroxidase and glutathione reductase activities in tissues.

Conclusions: These findings clearly indicate that the FS diet did not alter blood pressure of spontaneously hypertensive rats and Wistar rats. The FS diet resulted in hypertriglyceridemia but increased the total antioxidant status, which may prevent lipid peroxidation in these rats.

Keywords: Fructose; Saturated fatty acids; Spontaneously hypertensive rats; Antioxidant status

Introduction

Oxidative stress is obtained when the pro-oxidant challenge overwhelms the antioxidant defenses. Oxidative stress may contribute to the initiation and maintenance of hypertension by inactivation of nitric oxide (NO), which acts as vasodilator [1,2], the generation of vasoconstrictive isoprostanes [1], and vasopressor action [3]. Treatment with antioxidants may prevent or reverse abnormalities associated with hypertension and its complications. Many studies have reported that dietary supplements such as antioxidants, vitamins, and minerals prevent or at least attenuate organic impairment originated by excess oxidative stress [4,5]. In

This work was supported by the Council of Burgundy.

* Corresponding author. Tel: +33-03-80-39-63-11; fax: +33-03-80-39-63-30.

E-mail address: jprost@u-bourgogne.fr (J. Prost).

0899-9007/05/$ – see front matter
doi:10.1016/j.nut.2004.04.022

addition, modification in the macronutrient composition of the diet is an intervention that has been advocated by some to avoid oxidative stress, specifically, diets with decreased total fat, saturated fat, sugar, and relatively increased fiber. Clinical trials [6] that have used dietary approaches to stop hypertension have shown that a diet low in refined sugar with decreased intake of saturated fat and increased intake of fruits and vegetables decreases oxidative stress and blood pressure in hypertensive and normotensive individuals. In the same way, there is evidence that hyperlipidemia [7] and high-sugar diets [8], high-fat diets [9], or both induce oxidative stress. Larger amounts of fat and refined carbohydrate in diet also have been shown to promote hypertension and endothelial dysfunction in rats [10]. However, in this situation, the investigators used diets very high in carbohydrates (60%) or fats (20%) in animal models for hypertension studies [11–13] and to induce syndromes of insulin resistance to test hypoglycemic agents [14–16]. The role of modest amounts of carbohydrate and saturated fats on oxidative stress and blood pressure is unknown.

This study investigated the effects of a diet that combined fructose (18%) and saturated fatty acids (11%; FS diet) without distinguishing between the effects of either nutrient separately in normotensive and hypertensive rats. It is representative of the Western diet that is rich in refined sugar and saturated fat, and it appeared interesting to know the effects of such a combined diet. Fructose is a glucide that can be found in foods as a simple glucide and as a component of sucrose. Because of the use of high-fructose corn sweeteners and of sucrose in manufactured foods, the dietary consumption of fructose has increased several-fold from that present in natural food [17]. In the present study, the consequence of this FS diet in rats was assessed by measuring several variables related to lipid metabolism, blood pressure, and oxidative stress in blood and tissues.

Materials and methods

Animals and diets

Ten-week-old male, spontaneously hypertensive rats (SHRs; $n = 12$) and Wistar rats ($n = 12$) were purchased from Janvier (Le Ginestet St Isle, France). They were maintained at 24°C and constant humidity (60%) with a 12-h light, 12-h dark cycle. Each group of rats was divided into two groups of six rats each and fed different diets for 8 wk. One group received the control diet. The other group received the FS diet (Table 1). Sucrose and some starch were substituted with fructose and saturated fat in the FS diet. Food and tap water were freely available. Levels of selected vitamins and mineral salt were in line with recommended requirements [18]. We followed the general guidelines for the care and use of laboratory animals recommended by the Council of European Communities.

Table 1
Composition of the diets

Constituents (g/kg diet)	Control diet	FS diet
Casein*	260	260
Methionine†	3	3
Starch*	549.3	299.3
Sucrose*	44	—
Fructose*	—	184
Cellulose*	50	50
Vitamin mix‡	3	3
Mineral mix§	40	40
Isio 4‖	50	50
Végétaline‖	—	110
Choline chloride (98%)†	0.7	0.7

Végétaline provided the following fatty acids (% total fatty acids): C6:0, 0.53; C8:0, 7.57; C10:0, 6.02; C12:0, 47.87; C14:0, 18.19; C16:0, 9.27; C18:0, 9.65; C18:1, 0.90.

FS, enriched with fructose and saturated fatty acids

* Purchased from UAR (Villemoisson, Epinay sur Orge, France).

† Purchased from Prolabo (Paris, France).

‡ Purchased from UAR 200 (Villemoisson). This vitamin mix provided the following nutrients (mg/kg of dry diet): retinol, 1.8; cholecalciferol, 0.019; thiamine, 6; riboflavin, 4.5; pantothenic acid, 21; pyridoxine, 3; inositol, 45; cyanocobalamin, 0.015; ascorbic acid, 240; DL-α-tocopherol, 51; menadione, 12; nicotinic acid, 30; para-aminobenzoic acid, 15; folic acid, 1.5; biotin, 0.09.

§ Purchased from UAR 205B (Villemoisson). This mineral mix provided the following nutrients (g/kg of dry diet): Ca, 4; K, 2.4; Na, 1.6; Mg, 0.4; Fe, 0.12; elements (traces): Mn, 0.032; Cu, 0.005; Zn, 0.018; Co, 0.00004; I, 0.00002, completed to 40 000 with cellulose.

‖ Commercial products. Fatty acid compositions of Isio 4 expressed as percentages: C16:0, 6.1; C16:1 (ω-7), 0.1; C18:0, 3.63; C20:0, 0.3; C22:0, 0.7; C18:1 (ω-9), 38.6; C20:1 (ω-9), 0.23; C18:2 (ω-6), 44.73; C18:3 (ω-3), 1.3. Total saturated fatty acids: 10.8; total monounsaturated fatty acids: 40.2; total polyunsaturated fatty acids (ω-6): 47.2; total polyunsaturated fatty acids (ω-3): 1.7; total saturated fatty acids/total monounsaturated fatty acids; 0.30; total saturated fatty acids/total polyunsaturated fatty acids (ω-6/ω-3): 0.22; total polyunsaturated fatty acids (ω-6)/total polyunsaturated fatty acids (ω-3): 27.8.

Blood pressure measurement

Systolic blood pressure of conscious rats after 4 and 8 wk on the diets was measured by a non-bloody tail-cuff method [19]. Blood pressure values are the means of at least four measurements per rat, and only measurements at 8 wk are reported in Table 2.

Analytical procedures

Blood samples and tissue preparation

After the 8-wk dietary period, rats were deprived of food for 12 h and then anesthetized with sodium pentobarbital (60 mg/kg of body weight). Blood was collected from the abdominal aorta into tubes containing ethylene-diaminetetraacetic acid, and plasma was prepared by low-speed centrifugation (1000*g* for 20 min). Plasma concentrations of total cholesterol, triacylglycerol, and phospholipid were determined with enzyme kits (Boehringer, Meylan, France). Fasting glycemia was determined with glucometer (Life-