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Chapter 2

HIGH-FRUCTOSE CONSUMPTION AND METABOLIC DISEASES

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ABSTRACT

Renewed interest in the study of fructose arose after a number of papers on the harmful effects of ingesting high-fructose diets were published. Fructose intake has increased significantly in the United States and worldwide in the last four decades, which is mainly due to the increased consumption of high-fructose corn syrup and sucrose as a sweetener in beverages and in industrialized foods. There is substantial evidence from studies on rodents and on human adults suggesting that fructose intake in large amounts and for a long period of time leads to the development of the metabolic syndrome, insulin resistance, and dyslipidemia with increased plasma triacylglycerol, phospholipids and de novo lipogenesis. As a consequence, fructose may contribute to increased obesity, hypertension, type-2 diabetes and cardiovascular diseases. Fructose is a monosaccharide found in fruit, some vegetables and honey. Sucrose, a natural sweetener found in sugar cane and beets (50% fructose), is a disaccharide with two hexoses, one glucose molecule bonded to a fructose molecule by an alpha-1-4 glycoside. Although fructose has the same chemical formula as that of glucose (C_6 H_{12} O_6), it differs in its structure and metabolism. In the last few decades, sweeteners have been produced by corn starch hydrolysis, and part of the glucose produced is changed into fructose by enzymatic isomerization. This high-fructose corn syrup (42%-55% fructose) has a lower price than sucrose, and it has increasingly replaced sucrose as a sweetener in beverages and industrialized foods.

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Fructose can be produced from glucose by the sorbitol pathway. In endogenous fructose production, non-phosphorylated glucose is converted into fructose. Initially, glucose produces sorbitol in a reaction catalyzed by aldose reductase. In the next step, fructose is formed from sorbitol in a reaction catalyzed by sorbitol dehydrogenase. This pathway may be important in the embryonic stages of human life for regeneration of pyridine nucleotides in order to maintain ATP concentrations and cellular redox potentials when the fetus is in a low oxygen environment. After absorption, fructose is rapidly extracted by the liver and metabolized into fructose-1-phosphate in a reaction catalyzed by fructokinase, which is specific for fructose. Subsequently, fructose-1-P is split by fructose-1-phosphate aldolase in trioses, dihydroxyacetone phosphate and glyceraldehyde, a pathway to provide the backbone for triacylglycerol and phospholipid synthesis. Triose-P is converted into glucose and glycogen as well as into lactate, which can produce plasma lactic acidosis, and into glycerol, a precursor for triglycerides and de novo lipogenesis. Although considerable evidence suggests that high fructose intake can produce harmful effects, investigations where high-fructose diets are compared with high-glucose diets are necessary to clarify the real role played by free fructose in adverse metabolic effects. In epidemiological studies, the analysis of the effects of high-fructose corn syrup and high sucrose intake must consider that both products are composed of fructose and glucose.

INTRODUCTION

Fructose is a carbohydrate found in plants and humans. It was isolated in 1947 from sugar cane, where it is a component of sucrose. The word fructose originates from the Latin *fructus* as fruits are an important source of fructose. Fructose is considered a sugar and the word sugar is derived from the Sanskrit *çarkara* which means grain of sand. From this Sanskrit word came the Greek word *sakkaron*, the Latin word *saccharum*, and the Arabic word *sukkar*.

Free fructose is normally ingested with diet, as a component of honey, fruits, and some vegetables. It is also synthesized in the organism from glucose through the sorbitol pathway. In normal cellular metabolism this pathway is related to oxy-reductive process and regeneration of pyridine nucleotides.

Interest in studying fructose was renewed after many papers appeared on the harmful effects of high fructose diets. There is substantial evidence from studies in human adults suggesting that high fructose diets can induce metabolic syndrome, insulin resistance, and increased de novo lipogenesis which results in increased plasma triacylglycerol, phospholipids and cholesterol. As a consequence high fructose diets may contribute to obesity, Type 2 diabetes, hyperuricemia, high blood pressure, and cardiovascular diseases.

Early studies focused on its role as a glucose substitute in medical practice. Some studies suggested that fructose may be beneficial to individuals who are overweight, have non-insulin dependent diabetes mellitus, or participate in exercise activities [1]. However the clinical benefits conflict with harmful effects like ATP consumption, hypertriglyceridemia, and lactacidemia. Presently, its use is limited to specific situations such as a sport drink during endurance exercises.

In the last few years, fructose consumption has significantly increased as many Industrialized products use high-fructose corn syrups in place of sucrose to sweeten processed foods and beverages like bakery products, juices, and candies.

This chapter will present an overview of the fructose metabolism, possible uses, and the harmful effects when it is consumed in high quantities over long periods.

BIOCHEMICAL CHARACTERISTICS

Fructose is a monosaccharide with six carbon atoms linked, by simple covalent bonds, to hydroxyl groups formed by hydrogen and oxygen, and a carbonyl group with a double bond, linking carbon to oxygen. The position of this group determines whether carbohydrate hydrolysis will result in a ketone or an aldehyde. As fructose has the carbonyl group -CO- at the end of the chain, when hydrolyzed it produces ketone. For this reason fructose is considered a ketohexose. Although fructose has the same chemical formula as glucose (C_6 H_{12} O_6) it differs in structure and metabolism. Glucose has the carbonyl group in the first carbon and when hydrolyzed produces aldehyde. Carbohydrate oxidation is the main pathway for producing energy in most cells.

SOURCES OF FRUCTOSE

Fructose, also called levulose (D-fructose), can be found in fruits and honey as an isolated carbohydrate, or as a component in more complex carbohydrates like sucrose, and polymers like fructosan, e.g. inuline, found in certain tubers. Sucrose, a natural sweetener found in large quantities in sugar cane and beets (50% fructose), is a disaccharide with two hexoses, one molecule of glucose linked to one molecule of fructose by an alpha 1-4 glycosidic bond [2]. Raffinose, a trisaccharide found in cotton seed and sugar beet molasses, is composed of D-glucose, D-fructose, and D-galactose, and is formed by the transfer of galactose from UDP-galactose to sucrose.

Fructose can result from oxidation of sorbitol in a reaction mediated by sorbitol dehydrogenase. Sorbitol is present in many plants, particularly apple, pear, cherry, plum, and apricot. Table 1 shows the fructose, glucose, and sucrose content of several foods.

In the seventies, high performance liquid chromatography facilitated the separation of fructose from sugars. This method increased the production of a pure form of fructose and decreased its price. As well as isolating an isomerase which could transform D-glucose in D-fructose [4], contributed to the commercial introduction of high-fructose corn syrup (HFCS), a corn starch syrup rich in fructose. HFCS-42 (42% fructose) was introduced in 1967 and HFCS-55 (55% fructose) in 1977. HFCS-42 is 1.16 times and HFCS-55 1.28 times as sweet as sucrose [4]. This high-fructose corn syrup is cheaper than sucrose and has increasingly replaced sucrose as a sweetener in beverages and industrialized foods [6]. Fructose is highly soluble in water solutions and very sweet, 1.7 times the sweetness of sucrose, favoring its use over sucrose. Bakery foods contain between 1.0% and 2.05% fructose and if fruits are added this may increase to nearly 11.0%. Honey is high in fructose, 42.4%, and is considered a natural sweetener [6].

Table 1. Sugar contents of foods, g/100g

Foods	Fructose	Glucose	Sucrose
Apple	7.6	2.3	3.3
Apple juice	5.5	2.5	1.7
Apricot, raw	0.7	1.6	5.2
Apricot, dried	12.2	20.3	6.4
Bananas	2.7	4.2	6.5
Blackberries	4.1	3.1	0.4
Blueberries	3.6	3.5	0.2
Cherries	6.2	8.1	0.2
Figs, raw	2.8	3.7	0.4
Figs, dried	26.0	28.6	6.5
Grapes	6.9	6.6	1.4
Orange raw	2.5	2.2	4.2
Orange juice, raw	3.0	2.8	4.1
Pears raw	6.4	1.9	1.8
Pear juice, raw	7.1	1.6	-
Honey	41	34	2
Asparagus	1.3	0.9	0.2
Beets	0.2	0.2	6.1
Broccoli	0.7	0.6	0.3
Carrots	1.0	1.0	3.6
Cauliflower	0.8	0.9	0.5
Tomato	1.4	1.1	0.0

Adapted from Mathews et al. (1987), ref. [3], and Hallffrisch (1990), ref. [14].

FRUCTOSE CONSUMPTION

Carbohydrate intake in the Western World is 200-300g/day corresponding to between 40% and 50% of ingested energy. In the United States, mean daily fructose intake is 16g/day and 31g/day at the 90 percentile; if corn syrup is added this may reach 60-100g/day, or even 150g/day if sucrose is added [7].

Added sweeteners are important components of the American diet and from 1994 to 1996, represented 318kcal of dietary intake and 16% of total caloric intake. There is no doubt that fructose ingestion has increased in the last few decades in the United States, the highest consumers being teenagers and young adults, and sweet beverages the most important source of fructose intake [2].

In 1970, reports from the United States Department of Agriculture (USDA) indicated that the per capita added sugar in the American diet was 90/g day and no HFCS was consumed. Between 1970 and 1985 HFCS consumption increased and sucrose intake decreased [2]. USDA dietary records from 1977-1978 indicated mean daily fructose intake was 37g in the United States population [8]. In this survey sugar sweetened-beverages appeared as the mainly source of fructose in all age classes. The exception was for children under 6 years and adults over 50 years [8]. In an interesting review article, Tappy and Lê [2] analyzed the evolu-

tion of fructose intake. The authors found that from 1977 to 1990 average daily fructose intake was 54.7g, corresponding to an increase of 46% in a 10 to 16 year period (The Third National Health and Nutrition Examination Survey-NHANES III). Assessment of the evolution of fructose intake from 1999 to 2004 indicated that average fructose consumption was 49g/day and accounted for 42% of all calories from sweeteners consumed (NHANES 1999-2004) [2].

Although the United States is the main HFCS user in the world, HFCS is now produced and used in other countries, but data is scant. South America and Oceania are the highest sugar consumers; the lowest are Asia and Africa [2].

In Brazil, a sugar exporting country, mean fructose ingestion from fruit and vegetables is 4.34g/day, and from sucrose, 27.5g/day. This estimate was based on statistical data of alimentary ingested products, from the 1995/6 survey of Brazilian family budgets (IBGE-8) [9].

METABOLIC ASPECTS OF FRUCTOSE

Intestinal absorption

Fructose absorption across the small intestine is performed by two different mechanisms. Absorption takes place via a facilitated transport, independent from glucose, with a slower absorption rate than glucose and galactose, and by a glucose-dependent high capacity co-transport [10].

Fructose, present at the brush-border membrane, is transported into the enterocyte through a specific fructose transporter, GLUT5, located at the apical pole of the enterocyte. This transporter has a high affinity for fructose and low affinity for glucose. Fructose is transported throughout the enterocyte by the GLUT2 transporter, situated at the basolateral pole of the enterocyte.

The capacity of human adults to absorb fructose is limited and symptoms of malabsorption such as diarrhea and flatulence have been reported after fructose loading. This effect is largely abolished when fructose is ingested with glucose possibly due to activation of the fructose transporter by glucose [1, 11]. Fructose absorption is increased when ingested as sucrose or ingested with glucose. During glucose absorption small solutes, including fructose, move passively through opened junctions increasing fructose absorption by 29% [12]. Malabsorption rates for fructose and sorbitol are very similar in all studies. They vary according to the quantity and concentration of the sugar. After a 50g load of fructose in a 20% solution, 60% to 70% of patients display malabsorption [13].

Hepatic Metabolism

First Steps of Fructose and Glucose Metabolism

Fructose is mainly metabolized in the liver, although the intestines and kidneys have the enzymes necessary for its metabolism. A small proportion of orally administered fructose may be converted to glucose and lactate during transport through the intestinal wall [2].

After absorption, the fructose present in portal blood is rapidly extracted by hepatocytes. Although fructose is absorbed more slowly than glucose, it is more rapidly metabolized by the liver. The rapid fructose entrance into liver cells is mediated by the GLUT2 transporter. This process takes place without energy expenditure or insulin stimulation. Differently to glucose, fructose only has a modest effect on the stimulation of insulin secretion and does not require the presence of insulin to enter the intracellular compartment [1].

The initial steps of hepatic glucose metabolism differ from those of fructose, where different enzymes and reactions take place. Glucose is converted into glucose-6-P under hexokinase or glucokinase enzyme action. Glucose-6-P is converted to fructose-6-P and to fructose-1,6-di-P through a reaction catalyzed by the enzyme phosphofructokinase. Subsequently fructose-1,6-di-P is cleaved into two trioses, di-hydroxyacetone-P and glyceraldehyde-3-P (Figure 1). The metabolites from fructose-1,6-di-P will further be converted into pyruvate that enters the Krebs cycle to produce ATP and CO_2. The conversion of glucose to pyruvate is regulated by the energy status of cells, and by insulin that stimulates glucokinase gene expression and the activation of glycolytic enzymes [2].

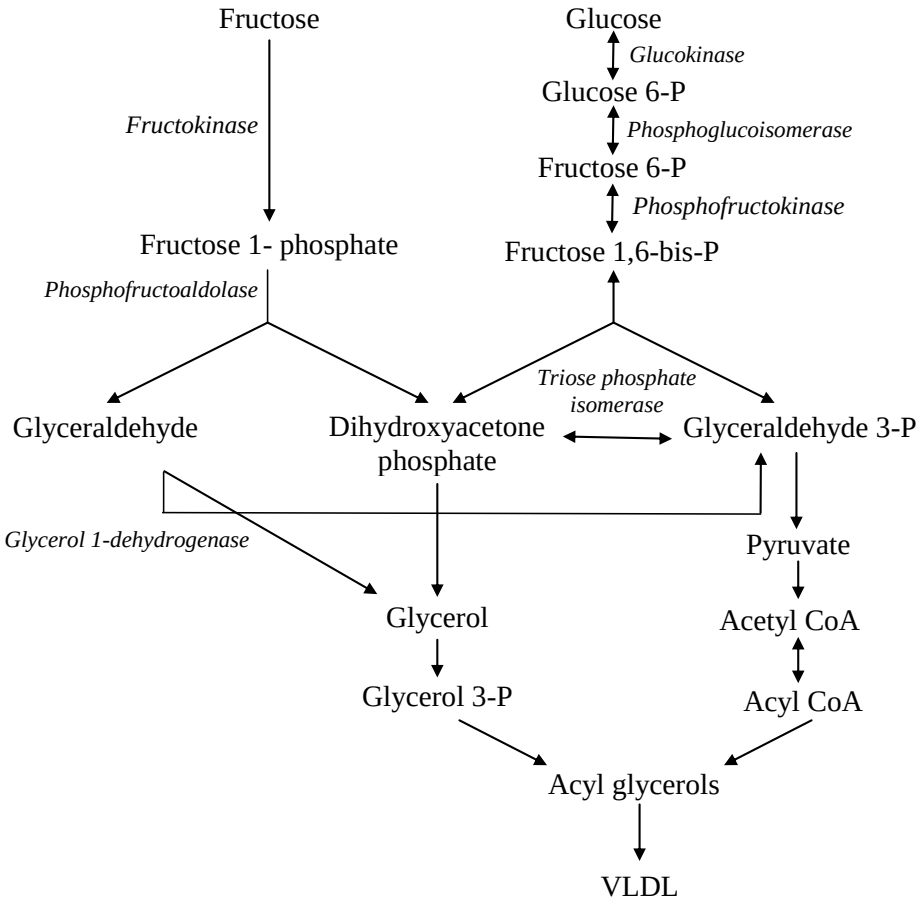

Figure 1. Fructose and glucose metabolism. Possible fructose metabolic pathway to generate triacylglycerol synthesis.

A large part of fructose uptake from the portal vein occurs immediately during the first pass in the liver. Once inside the cell, fructose is rapidly phosphorylated on carbon one in a reaction mediated by fructokinase, a fructose specific enzyme, and is converted to fructose-1-phosphate. This phosphorylation bypasses the initial limiting steps of insulin-stimulated transport needed for glucose metabolism. The increase in fructose intake stimulates increased fructokinase activity [14]. Further, fructose-1-phosphate is cleaved by aldolase B into two trioses: di-hydroxyacetone phosphate and glyceraldehyde, rather than glyceraldehyde-3-P which is directly produced in glucose metabolism. Di-hydroxiacetone phosphate can be isomerized into glyceraldehyde-3-phosphate. Glyceraldehyde from fructose is further phosphorylated to glyceraldehyde-3-P. This is the point where fructose metabolism is interrelated with glucose metabolism.

Metabolic Fate of Triose-P from Fructose

Triose-P derived from fructose can be further metabolized in different ways. The major part of triose-P is converted to glucose and glycogen through gluconeogenesis [2,15] (Figure 1). About two thirds of fructose is converted to glucose, which may accumulate as glycogen, be released from liver as glucose [1], or be oxidized in extrahepatic tissues [2].

Triose-P may be metabolized in the glycolytic pathway and then converted into lactate and pyruvate. A significant increase in plasma lactate can be observed after oral or intravenous fructose intake, and has been associated with lactic acidosis. Under normal circumstances, small amounts of fructose can be converted into pyruvate and oxidized to carbon dioxide through the tricarboxylic acid cycle. Before pyruvate entrance in the citric acid cycle it must be oxidatively decarboxylated to acetyl-Co-A, the source of carbon for fatty acid synthesis. Only a small proportion of fructose is converted to de novo fatty acids (Figure 1) [16].

Part of the glyceraldehyde produced is phosphorylated by a specific kinase to glyceroldehyde-3-P, a precursor of glucose production through gluconeogenesis. Part is reduced to glycerol by the action of glycerol 1-dehydrogenase and then phosphorylated to provide glycerol-3-P, a precursor in the synthesis of acylglycerols and other lipid products by de novo lipogenesis [14,17] (Figure 1).

The bulk of blood triacyglycerol is carried in 2 lipoprotein particles: chylomicrons and VLDL particles. Chylomicrons are produced by the intestine and are found in low concentrations in blood during fasting. These particles contain proteins, apolipoprotein B-48, and carry triacylglycerol derived from ingested fat. VLDL particles are produced by the liver, contain apolipoprotein B-100, and carry endogenous triacylglycerol. Concentrations of apo B-48, the apolipoprotein from chylomicrons, are normally 6 times lower (0.01μmol/L or 0.3mg/dL) than those of VLDL apo B-100 (0.06μmol/L or 3mg/dL) in the fasting state. Blood triacylglycerol (20% to 50%) can be carried in remnants of VLDL called intermediate-density lipoproteins (IDLs) [18].

The adverse effects of high fructose intake are related to the amount of fructose and the length of time that a person remains on a high fructose diet. Rats fed a high fructose diet, exhibit characteristics of metabolic syndrome which include insulin resistance, hyperinsulinemia, hypertriglyceridemia, and hypertension. The adverse effects were related to the amount and duration of fructose consumption [19]. There is evidence from studies of at least 4 weeks dietary control that diets containing ≥20% energy as fructose can cause lipid abnormalities like hypertriglyceridemia due to VLDL-triglyceride increases when associated with hyperinsulinemia, and LDL-cholesterol increases in normoinsulinemic subjects [20]. A discretely

lower amount of fructose, >15% of energy as fructose, was associated with increases in both fasting and postprandial triacylglycerol in humans [16]. In perfused liver, fructose inhibited hepatic lipid oxidation and facilitated fatty acid reesterification and VLDL-triglyceride synthesis [21].

As fructose can be metabolized by different pathways, more studies are needed to identify the contribution, and possible effects on human health, of each metabolic pathway. In a study investigating the effects of fructose or glucose meals on postprandial lipemia in 14 healthy subjects, the authors used stable isotopes, [$2H_2$]palmitate and [$U^{13}C$]D-fructose or [$U^{13}C$]D-glucose to trace the fate of dietary sugars [16]. Post-meal plasma triacylglycerol concentration was higher and rose earlier after fructose than glucose ingestion. The authors concluded that the contribution of de novo fatty acids to the increase of VLDL-triglycerides (VLDL-TG) by fructose is small. In contrast, de novo TG-glycerol formed from labeled fructose made up 38% of circulating VLDL-TG 240 minutes after fructose ingestion. Only 35% of the fructose load was oxidized over 6 hours. Approximately 40% of the remaining fructose was transformed into lactate and glycogen. The authors concluded that fructose contribution to de novo triacylglycerol synthesis through glycerol production is more significant than its contribution through fatty acid production. The conclusion was that a decrease in plasma-TG removal, via a lower activation of adipose tissue lipoprotein lipase, contributes to acute fructose-induced lipemia [16,18,20]. The exact contribution from lipoprotein lipase needs to be clarified. Not only in humans was decreased triglyceride clearance considered a contributory factor to high triacylglycerol levels, but studies using rats fed high-fructose diets also lead to the conclusion that both fructose and sucrose increased triglyceride production and decreased triglyceride clearance [16,22].

Endogenous Fructose Production

An interesting aspect of fructose metabolism is endogenous fructose production by the sorbitol pathway. Sorbitol is frequently ingested with food as it is found mainly in fruits, or can be produced from glucose as an alternative pathway in glucose metabolism (Figure 2). In this pathway, fructose is created in the liver directly from glucose supplied by the blood through the intermediate formation of its corresponding alcohol, D-sorbitol. This pathway has not been fully studied in humans and its real function and contribution to the total fructose pool is unknown.

It is has long been known that human spermatozoa live in a fructose environment produced by the prostate gland and seminal vesicles. The metabolic pathway includes aldose reductase and sorbitol dehydrogenase. The same process may occur in the liver, nerve tissue, and muscle [23]. The polyol pathway has also been identified in plants and animal tissues [24].

Ungulate animal placentas produce fructose and high fructose concentrations are found in fetal blood. Biochemical evidence has shown that fetal fructose in these animals is produced in the placenta from maternal glucose [25,26]. In the human fetus and newborn, data on polyols and sugars other than glucose, are scarce and their roles in placental, fetal, and postnatal metabolism need to be defined [27], Fructose, sorbitol, inositol, erythritol, and ribitol in coelomic and amniotic fluid from 5 to 12 week gestation embryos [28], and sorbitol

in cord blood from full-term newborns have been found at higher levels than maternal concentrations, suggesting that the polyol pathway is active in the fetus [29].

In endogenous fructose production, non-phosphorylated glucose is converted into fructose (Figure 2). In a first step, glucose, in a reaction catalyzed by aldose reductase, generates sorbitol and NADPH is oxidized to $NADP^+$. In the second step, fructose is formed from sorbitol in a reaction catalyzed by sorbitol dehydrogenase and NAD^+ is reduced to NADH, a co-factor that under anaerobic conditions is reoxidized by the conversion of pyruvate to lactate which can result in increased blood lactate [1,28,30]. NAD^+ allows the glycolisis process to continue and produce ATP [14,28,31]. This route bypasses the ordinary glucose metabolism pathways, in which phosphorylated derivatives are intermediates.

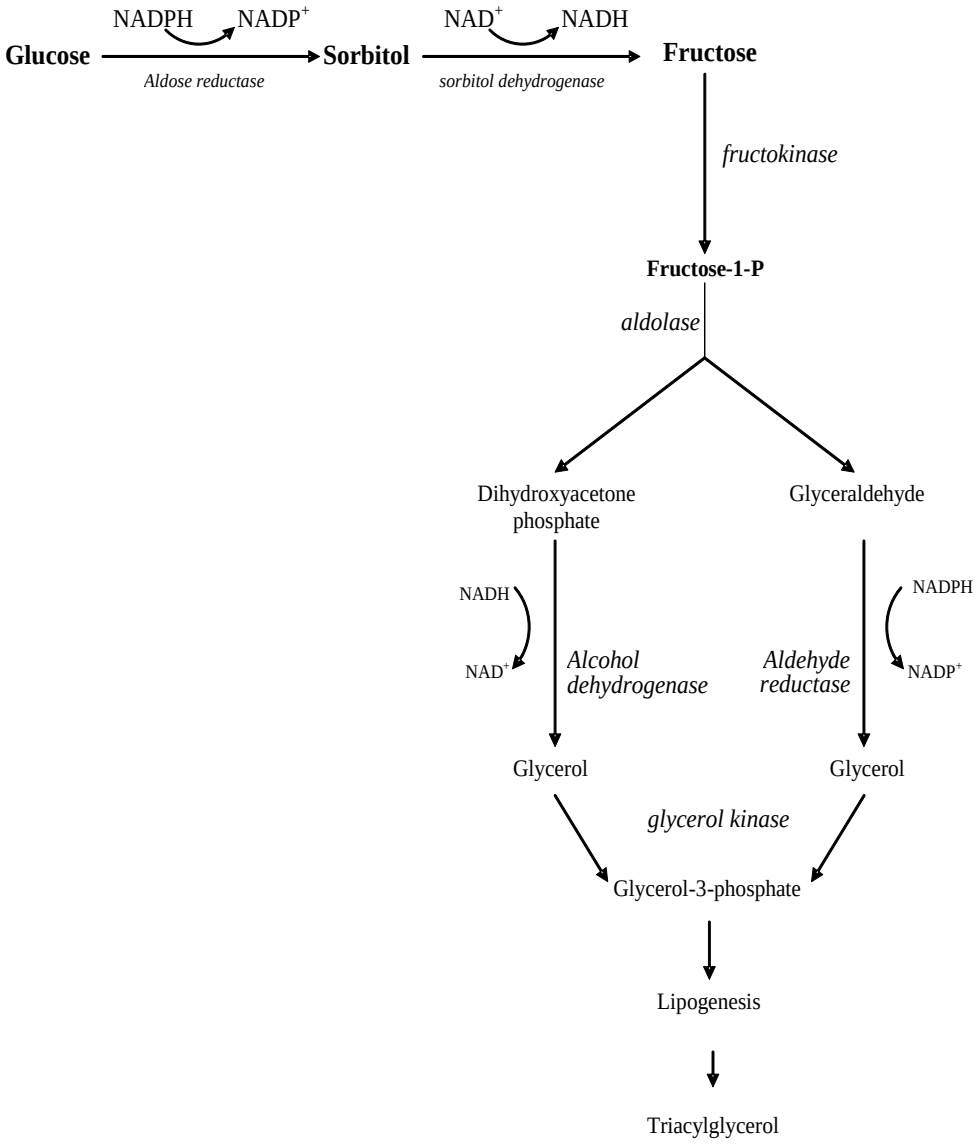

Figure 2. Schematic representation of fructose production and metabolism via the polyol pathway.

Many papers have been published concerning the harmful effects of high–fructose diets on human adults. In contrast few papers refer to obese children and high fructose diet [32], and one investigated fructose in coelomic and amniotic fluid from 5-12 week gestation embryos showing the presence of fructose in early life [28]. The lack of studies in this vulnerable period of life stimulated us to investigate fructose in fetus (cord blood) and the newborn infant to identify its presence in the early perinatal period and to find a physiological role for it.

In our study [33], investigating the presence of fructose in maternal blood and umbilical cord blood, and in peripheral blood from the normal full-term newborn 48h after delivery, we could observe that fructose concentrations in fetal blood were significantly higher than in maternal blood, suggesting endogenous fructose production by the fetus via the sorbitol pathway. The highest fructose values were found in newborns 48h after birth suggesting that fructose production is a continuous process from fetus to newborn infant. Considering that the newborns were breast-fed and that analysis of their breast milk showed it did not contain sorbitol or fructose, we concluded that fructose originated from glucose or glycogen through the sorbitol pathway [33]. From the beginning we had two questions: why the fetus uses an alternative pathway to the glucose metabolism to produce fructose, considering that glucose is a very important carbohydrate in producing energy for fetal and newborn brain and growth. The second was about the destiny of the endogenous fructose produced.

Besides a suggestion in literature [28] that fructose pathways play a role in reoxidation of pyridine nucleotides during the early stages of gestation, we hypothesized that fructose may be a precursor for lipid synthesis in the fetus and newborn. This hypothesis is based on the concept that fructose is more lipogenic than glucose and from analyzing the lipogenic effects of high-fructose diets in humans. At 50% to 60% gestation there is little lipid uptake by the fetus, mainly the essential fatty acids necessary for membrane development. In the last trimester of gestation large amounts of body fat are deposited (12% to 18% of body weight). The importance of this adiposity and the mechanisms that produce it are not well known. Considering that neutral lipids do not pass through the placental barrier, they must be produced by the fetus, thus fructose may contribute to the carbon backbone for triacylglycerol production in the fetus, and also in the newborn as seen in sheep fetuses [34]. In that article the [^{14}C] derived from [^{14}C] glucose and [^{14}C] fructose infused in fetal circulation was incorporated into neutral lipids and phospholipids from the heart, liver, kidney, brain, and adipose tissue. The incorporation patterns from fructose and glucose were similar.

Ectopic Lipid Deposition

Literature is scarce on intrahepatocellular (IHCL) and intramyocellular (IMCL) accumulation of lipids as a consequence of high-fructose diets in humans. Triglyceride deposition in hepatocyte cytoplasm leads to nonalcoholic liver disease. In rodents, a high-fructose diet induces hepatic insulin resistance, IHCL, and stimulates hepatic de novo lipogenesis [35]. High-fructose or sucrose ingested for long periods may induce hepatic steatosis, insulin resistance, and IMCL accumulation [36,37,38]. In healthy subjects with and without a family history of Type 2 diabetes submitted to a 7-day isocaloric diet or hypercaloric high-fructose diet, the authors concluded that the high-fructose diet increased fasting VLDL-triacylglycerol, induced ectopic lipid deposition in liver and muscle, and

decreased hepatic insulin sensitivity. The high-fructose diet increased VLDL-triacylglycerols and IMCL to a greater extent in healthy offspring of subjects with a family history of Type 2 diabetes. This group appeared more susceptible to the adverse effects of high-fructose diets [37]. These characteristics were considered indicators of genetically determined metabolic alterations [37].

FRUCTOSE AND EXERCISE

During prolonged exercise blood glucose levels fall and muscle glycogen depletion occurs. This limits athletic performance. Ingestion of carbohydrate solution, prior to 60 minutes or longer of high-intensity exercise can delay fatigue and improve endurance performance, by preventing hypoglycemia and by maintaining a high carbohydrate-oxidation rate. This is important during the later stage of exercise, when endogenous muscle and liver carbohydrate stores may be depleted [1]. The contribution by exogenous glucose to energy provision during prolonged high-intensity endurance exercise is limited to ~1.0-1.1g/min, even when ingested in quantities that exceed this rate. This could be explained by saturation of sodium-dependent glucose co-transporter (SGLT1) which is located in the brush-border membrane of the small intestine and is responsible for absorption of glucose by active transport [39].

Literature shows that glucose intake during endurance exercises has beneficial effects. However, glucose stimulates insulin and suppresses glucagon secretion. Fructose, on the contrary, produces minimal changes in blood glucose, insulin, and glucagon levels and is considered a more ideal energy source during prolonged exercises [1]. Although fructose has some advantages over glucose as an energy source, there are conflicting reports in literature, and it has been suggested that fructose, as a supplement in sports drinks, may have little advantage over glucose ingestion during endurance exercise [1].

There are benefits in the association of glucose and fructose. During exercise, fructose ingested with glucose increases exogenous carbohydrate oxidation rate up to 1.75g/min compared with 1g/min during exercise with glucose alone [40,41]. Fructose ingested with glucose increased intestinal glucose transport. This could explain the increase in glucose oxidation rate [40-42].

Plasma lactate increases during exercise when fructose and glucose are ingested together compared to isoenergetic amounts of glucose or glucose polymers. This could have a beneficial effect because lactate is efficiently oxidized by active muscles during exercise [43,44]. In a study with human volunteers, LeCoutre et al. observed that co-ingestion of fructose and glucose prior to exercise lead to an increase in carbohydrate oxidation rate, due essentially to enhanced lactate oxidation [45].

EFFECTS OF DIETARY FRUCTOSE

Dyslipidemia

Although many publications in literature address the effects of dietary fructose on lipid metabolism in humans, analyses must take into account that in high-fructose diets, fructose is

ingested together with glucose as a component of sucrose, or as HFCS which is composed of fructose and glucose. Administration of equivalent quantities of pure fructose, sucrose, a mixture of glucose and fructose, or HFCS resulted in similar postprandial increases in triglycerides [46]. Also many of the health problems induced by fructose are similar to diseases produced by overeating, high-fat diets, and high-carbohydrate diets. High-fructose/high-sucrose diets lead to adverse metabolic effects and cause dyslipidemia, insulin resistance, hypertension, hyperuricemia, and obesity [2]. Although animal studies have been described since 1970 [47,48], the bulk of literature on dietary fructose and human health has only appeared in the last twenty years.

Some human groups are more susceptible to presenting hypertriglyceridemia from fructose intake; these include the male gender, post menopause women, hyperinsulinemic men, previous hypertriglyceridemia, and Type 2 diabetes [14].

In 2000, Bantle et al. [49] reported gender differences when feeding men and women for 6 weeks on a diet providing 17% of energy as fructose, and a diet sweetened with glucose. In men, the fructose diet significantly increased fasting, postprandial, and daylong plasma triacylglycerol concentrations compared to the glucose diet. In women, fructose had no effect. The reason for this gender difference was not clear. Also, triacylglycerol concentrations were higher in men than women after 24h ingestion of HFCS or sucrose sweetened beverages [46]. These data suggest that men are more susceptible than women to the effects of sugars containing fructose [46].

In an investigation of the mechanisms of the acute effect of fructose on postprandial lipemia, the authors used stable isotopes to trace the metabolism and the fate of dietary fructose and glucose [16]. Plasma triacylglycerol and VLDL-triacylglycerol concentrations were higher whereas the concentration of insulin and [$^2H_2$]palmitate in nonesterified fatty acids were lower after fructose than after glucose diets. The conclusion was that the lower insulin secretion after fructose ingestion may result in less activation of adipose lipoprotein lipase and impairment of triacylglycerol clearance. Therefore, two mechanisms seem to be responsible for the increased triacylglycerol, de novo lipogenesis, and decreased triacyl-glycerol clearance. Also de novo lipogenesis from fructose contributes to the partitioning of fatty acids toward esterification [16].

A systematic review and meta-analysis assembled sixteen clinical trials and included 236 subjects to compare the effects of isocaloric fructose and carbohydrates. The following lipids were analyzed in Type 1 and Type 2 diabetic subjects: triglycerides, total cholesterol, LDL cholesterol, and HDL cholesterol. Increasing triglyceride and lowering total cholesterol was observed only in subjects with Type 2 diabetes when starch was the reference carbohydrate. These effects were not observed when fructose was compared to sucrose or mixed carbo-hydrate as reference because both sources contained fructose [50].

Fructose at >60g/day used as a sweetener raised triglycerides in ≥4 weeks follow-up [50]. Young adults and women who consumed glucose, fructose, and HFCS-sweetened beverages at 25% of energy requirements for 2 weeks, had the 24h triglyceride area under the curve increased during consumption of fructose compared with baseline, and fasting low-density lipoprotein (LDL) and apoB concentrations increased during ingestion of fructose and HFCS [51]. These authors estimated that 13% of the United States population consumes 25% or more of its energy from added sugar, and therefore this population are at risk of cardio-vascular diseases [51].

A meta-analysis which evaluated the effects of dietary fructose concluded that an intake >50g/day was associated with increased postprandial triglyceride excursions, whereas a fructose intake >100g/day was associated with increased fasting triglycerides [2,52]. The estimates of fructose intake, based on NHANES 1999-2004, revealed that young men in the 15-18 and 19-22 years age groups had the highest estimated mean intake of total fructose, 75g/day. Women had lower estimated fructose intakes than men in all age groups, 48.6g/day versus 62.8g/day. Total estimated mean usual daily fructose intake in both genders and all ages was 49g/day, very close to the amount considered a risk factor for increased postprandial triglyceride excursions [53].

Obesity and Fructose

The increase in obesity worldwide over the last decade has been considered an important public health matter, principally because obesity is associated to comorbidities like: metabolic syndrome, insulin resistance, Type 2 diabetes, hypertension, and coronary artery disease. The concomitant increase in soft drink consumption, sweetened with sucrose or HFCS, suggests a relationship between fructose and the obesity epidemic [2,5,54,55], especially in young people and children [56,57].

Epidemiological studies support the association between high sugar intake, adiposity, and body weight increase. Some reports are descriptive and results contradictory. Although there is no doubt that fructose may produce acute and long term increases in blood triglycerides and visceral lipids it is important to recognize that fat diets, sedentary lifestyle, socioeconomic factors, hormones, and genetics are also contributive factors to obesity. Obesity is a very complex state which involves the regulation of food intake, energy expenditure, and body fat stores by different nutrients, hormones, and the central nervous system [48]. Endocrine regulators of energy homeostasis, like leptin, insulin, ghrelin, and possibly peptide YY_{3-36} from the distal intestine, are released in proportion to body fat, to the quantity and composition of food consumed [48]. Some experiments support a relationship between fructose and the behavior of hormones that regulate energy homeostasis.

To understand the real effect of fructose on human health it is important to analyze studies in which free fructose is compared to glucose, even though these diets are not used in daily life, but only in experiments. Experimental studies in mice with free access to fructose (15% solution in water), sucrose 10%, or artificial sweetener (0% of calories), showed that animals in the fructose group had greater increases in body weight and body fat than the animals in the sucrose and artificial sweetener groups. Also, fructose supplementation impaired glucose tolerance, but had no influence on insulin, adiponectin, and ghrelin levels [58]. In human subjects the consumption of fructose-sweetened beverages compared to glucose-sweetened beverages reduced insulin secretion, leptin production, and attenuated ghrelin suppression [59]. Consumption of fructose or glucose-sweetened beverages at 25% of energy requirements demonstrated that fructose increased visceral adipose deposition and de novo lipogenesis, but decreased glucose tolerance, insulin sensitivity in older, overweight and obese men and woman [60]. These effects were not observed with glucose ingestion.

Insulin plays a role in regulating body adiposity by its action in the central nervous system which leads to inhibition of food intake, and increased energy expenditure [48]. Insulin receptors are expressed in several CNS areas related to the control of food intake and

energy homeostasis [61]. Fructose is a weak stimulator of insulin secretion probably because of the low expression level of fructose transporter GLUT5 in pancreatic β-cell. Leptin is produced by adipocytes and acts as a signal to the CNS in the regulation of food intake, energy expenditure, and body adiposity [62]. Together with insulin it acts in the hypothalamus and regulates food intake and energy metabolism via neuropeptide systems [51]. The leptin area under the curve over 24 hours was reduced by 20%-30% when normal weight women [59], and overweight men and women [63] consumed meals with fructose compared with glucose-sweetened beverages. In these studies, the presence of insulin resistance led to hypertriglyceridemia in response to glucose and fructose, but the intensity of the changes were higher with fructose.

Ghrelin is a peptide hormone produced by the stomach and upper small intestine. Its action is associated with food intake regulation [51]. Circulating ghrelin levels are inversely related to body weight and ghrelin increases after the intake of weight loss diets. Plasma ghrelin was 35% lower after meals accompanied by a glucose-sweetened beverage; but the decrease was greater after fructose-sweetened beverages [59].

Uric acid and Fructose

Increased dietary fructose intake has boosted research in various scientific fields. Studies on renal diseases and metabolic syndrome have linked increased fructose intake with high uric acid levels [64]. Fructose and its association with uric acid have been found in other disease situations; these include obesity/metabolic syndrome, gout, hypertension, diabetes, and heart and kidney diseases [2,65].

Uric acid seems to be an indicator of the toxic effect from high fructose intake [66], and its early formation, seen 30 minutes after ingesting the carbohydrate, reinforces the connection between fructose metabolism and uric acid formation. The action of hepatic fructokinase on that carbohydrate results in fructose 1-P, ADP, AMP, and uric acid [2]. Regulation of this pathway does not seem to suffer from the action of negative regulators, as observed in the glucose pathway, suggesting that excessive fructose intake, without proper control, can result in high uric acid levels.

Nakagawa et al (2005) observed that uric acid plays a fundamental role in the development of experimental metabolic syndrome. Treating these rats with allopurinol, a xanthine oxidase inhibitor which lowers uric acid levels, was able to decrease systolic blood pressure, increase insulin sensitivity and normalize triglyceride levels after the development of fructose-induced metabolic syndrome [67].

The capacity to produce uric acid from consuming large quantities of fructose seems to be influenced by genetic factors. Genetic polymorphism of the genes responsible for the synthesis of anion transporters such as Uric Acid Anion Transporter 1 (URAT-1) is associated with hyperuricemia [68]. Similarly to other enzymes or metabolic pathways, uric acid production also seems to be influenced by an individual's genetics and to be controlled by various genes and pathways [69]. Some individuals may produce high concentrations of uric acid whereas others may produce lower levels, suggesting a higher or lower susceptibility in certain individuals to the propensity and/or development of fructose-induced metabolic syndrome. Other studies have suggested that fructose may be produced by an individual's organism, and they report that this pathway may be an alternative pathway that complements

the toxic effect of fructose and metabolic syndrome. Individuals with diabetes and severe insulin resistance seem to synthetize fructose from glucose by using the polyol pathway [33]. Another negative effect of induced uric acid production in this situation is the inhibition of endothelial NO synthesis, which has been observed in cell cultures and experimental models [67,70]. The capacity of uric acid from the fructose metabolism to inhibit NO (endogenous vasodilator) has been associated with hypertension and heart disease. Uric acid also has a direct toxic effect on adipocytes, causing inflammation and inducing oxidative stress. These findings on the role of uric acid seem controversial, considering that the uric acid is one of the main antioxidants in the circulation. Finally, further studies on the fructose metabolism and the role played by uric acid in microenvironments and specifics diseases are still needed. Clarifying the actual role played by uric acid in various diseases, its relationship with high fructose intake, its production, and inhibition pathways is still a challenge to be overcome.

Hypertension and Fructose

Animal studies have shown a clear association between fructose ingestion and hypertension [71]. Experimental studies in dogs fed the same levels of high fructose and glucose diets, showed a net difference in metabolic response, with high fructose leading to hypertension and other adverse effects [72]. An association was also seen in rats between high-fructose ingestion and the development of cardiac hypertrophy [73], reduced baroreflex sensitivity [74], and renal damage [75]. Ingestion of acute quantities of fructose in humans elicits an increase in blood pressure which is probably mediated by increased cardiac output without compensatory peripheral vasodilation. The contrast between fructose ingestion response and the same amount of glucose may be explained by insulin hemodynamic action, as fructose has a negligible effect on insulin secretion. Physiological elevations in plasma insulin concentrations produce a rise in cardiac output and a reduction in muscle and systemic vascular resistance [76].

The association between fructose and hypertension may be explained by the uric acid pathway. Excessive fructose consumption leads to increased serum uric acid via fructose phosphorylation, and the generation of adenosine diphosphate which is metabolized to uric acid. Increased uric acid levels are associated to reduced nitric oxide levels and thus to blood hypertension. Sugar consumption is also associated to enhanced sympathetic nervous system activity, and sodium retention [77].

Frutose is the only sugar that raises uric acid concentration because fructose enters into hepatocytes and other cells, including tubular cells, adipocytes, and intestinal epithelial cells, where it is completely metabolized by frutokinase. In this process ATP is consumed without a negative regulatory mechanism to prevent ATP depletion. This leads to increases in lactic acid and uric acid levels which can increase 1-4mg/dL after a large ingestion of fructose [78]. Uric acid is associated with hypertension and a large number of deleterious effects, mainly by the stimulation of vascular smooth muscle cell proliferation and release of chemotactic and inflammatory substances [79,80]. The mechanism by which uric acid causes hypertension may involve a reduction in the concentration of endothelial nitric oxide, which results in systemic and intrarenal vasoconstriction, renal microvascular disease, and systemic hypertension [78,81].

Other experiments give support to the hypothesis that fructose plays a role in the pathophysiology of hypertension. Studies with mice have shown that diets rich in fructose stimulated salt absorption in the small intestine and kidney tubules, and resulted a state of salt overload which caused hypertension [82]. In the PREMIER study, a behavioral intervention trial of 810 prehypertensive and hypertensive individuals, reduced sugar-sweetened beverages or sugar intake over 18 months was associated with reduced blood pressure [83].

Metabolic Syndrome / Insulin Resistance / Diabetes

Different definitions have been proposed for metabolic syndrome in the last few decades [84,85]. Metabolic syndrome is a combination of clinical and laboratory findings which include central adiposity, hypertension, dyslipidemia, inflammation, and impaired glucose tolerance that predispose affected individuals to atherosclerotic cardiovascular disease and Type 2 diabetes [84,86,87]. Insulin resistance is the key metabolic feature of metabolic syndrome [88].

Currently, epidemiological studies report an alarming worldwide increase in cardiovascular disease and metabolic syndrome [84]. Excessive fructose consumption in diet has been epidemiologically linked to the development of metabolic syndrome [89]. More recent findings suggest that high fructose intake induces a series of metabolic and cardiovascular alterations in both animal and human models [90]. However, there is a metabolic difference between rodents and primates, mainly in the lipoprotein metabolism, the main site for lipogenesis and the physiology of thermogenesis [88]. Increased dietary fructose intake in rats and dogs for several weeks has also shown many parameters of metabolic syndrome, including hypertension, insulin resistance, and hyperlipidemia [90]. The mechanism responsible for the adverse effects of fructose diet is not quite clear, but there is evidence that such a diet promotes gluco-oxidative stress.

Studies in rhesus monkeys have shown that fructose-sweetened drinks are well tolerated, and that within 6 to 12 months of consumption, a high-fructose diet produces many of the characteristics of metabolic syndrome in humans, including central obesity, insulin resistance, inflammation, and dyslipidemia [88]. High fructose consumption in humans increases ectopic fat deposition in the liver and skeletal muscle in young healthy individuals without a family history of diabetes [37] and increased hepatic inflammation and fibrosis [91]. Inflammation is a corollary of obesity, which is marked by a broad inflammatory response. Inflammation cytokine TNF-α is constitutively expressed in adipose tissue and over-expressed in rodent models of obesity [92]. Experimental studies have shown that fructose increases production of tumor necrosis factor (TNF-α), a pro-inflammatory cytokine which can induce peripheral resistance to insulin and production of lipoproteins [93].

Some studies have suggested a strong association between insulin resistance and endothelial dysfunction. Endothelial dysfunction induces vascular relaxation, with a reduction in the bioavailability of nitric oxide (NO) and an increase in oxidative stress and O_2^- superproduction levels, a key event for the development of cardiovascular complications associated to insulin resistance in experimental fructose-fed rat models [90].

Elderly, overweight, and obese patients were evaluated for fructose consumption and adverse effects. When fructose consumption corresponded to more than 25% of their energy

needs, individuals presented increased visceral fat deposition and de novo lipogenesis, leading to dyslipidemia and reduced glucose tolerance [94]. Recent studies have reported that increased lipids is associated with increased diacylglycerol, which activates novel protein kinase C. Novel-PKC decreases tyrosine phosphorylation of the insulin receptor and/or insulin receptor substrate 1, resulting in increased hepatic glucose production, impaired glucose tolerance, and increased fasting glucose and insulin concentrations [84].

Fructose is also known to induce oxidative stress and mitochondrial dysfunction, resulting in the stimulation of peroxisome proliferator-activated receptor gamma coactivator 1-α and β (PGC1-α and PGC-1β) that drive both insulin resistance and lipogenesis [95,96]. The effect of fructose in lipogenesis has been reported as having the ability to alter the activity of key lipogenesis enzymes and transcription factors in the liver, such as pyruvate dehydrogenase kinase and sterol regulatory element-binding protein-1c (SREBP-1c), the main inducer of hepatic lipogenesis.

CONCLUSION

This chapter deals with the clinical and pathological aspects linked to high dietary fructose intake. The increase is specifically due to the introduction of HFCS as a sweetener in the United States, but less so in other parts of the world.

The adverse effects have been thoroughly demonstrated in experimental models, confirming the lipogenic effect of excessive fructose ingestion, with increased blood triglycerides, visceral lipids, insulin resistance, vascular dysfunction, and hypertension. However clinical studies, especially the epidemiological ones, associating the worldwide obesity epidemic [97], hypertension, and Type 2 diabetes to fructose, show several confounding factors and require better investigation.

Questions are raised in evaluating the studies; these include the amount of fructose ingested, diet duration, the previous presence of diabetes, and type of sweetener, remembering that HFCS and sucrose are similar in composition [98], Although free fructose is not in daily use, comparison between the effects of fructose and glucose have shown that the adverse effects are due to fructose [2]. Authors reported that the world obesity epidemic increases in the same proportion of sugar and sweeteners consumption. In addition, other authors have considered that fat consumption was reduced while obesity and fructose consumption increased in an epidemic manner [99]. Future studies can better elucidate the role of fructose in the pathophysiology of insulin resistance, metabolic syndrome, hypertension, and cardiac diseases.

REFERENCES

[1] Henry, RR; Crapo, P; Thornburn, AN. Current issues in fructose metabolism. *Annu Rev Nutr,* 1991;11:21-39.

[2] Tappy, L; Lê, K-A. Metabolic effects of fructose and the worldwide increase in obesity. *Physiol Rev,* 2010;90:23-46.

[3] Mathews, RH; Pehrsson, PR; Farhat-Sabet, M (1987). Sugar content of selected foods: individual and total sugars. *Home Economics Research Report no 48*, Human Nutrition Information Service USDA. pp. 3-14. Government Printing Office, Washington, D.C.

[4] Marshall, RO; Koo, ER. Enzymatic conversion of D-glucose to D-fructose. *Science,* 1957;125:648-649.

[5] Bray, A; Nielsen, J; Popkin, BM. Consumption of high-fructose corn syrup in beverages may play a role in the epidemic of obesity. *Am J Clin Nutr Nutr,* 2004;79:537-43.

[6] Wang, YM; Van Eys, J. Nutritional significance of fructose and sugar alcohols. *Ann Rev Nutr*, 1981;1:437-75.

[7] Rumessen, JJ. Fructose and related food carbohydrates. *Scand J Gastroenterol,* 1992;27:819-828.

[8] Park, YK; Yetley, EA. Intakes and food sources of fructose in the United States. *Am J Clin Nutr,* 1993;58(Suppl):737S-747S.

[9] Brazilian Institute of Geography and Statistics. *Familiar Budget Survey*, 1996. URL: http://www.ibge.gov.br/ (portuguese).

[10] Perman, JA. Digestion and absorption of fruit juice carbohydrates. *J Am Coll Nutr,* 1996;15(5Suppl):S12-S17.

[11] Kneepkens, CMF; Vonk, RJ; Fernandes, J. Incomplete intestinal absorption of fructose. *Arch Dis Child*, 1984;59:735-738.

[12] Shi, X; Schedl, HP; Summers, RM; Lambert, GP; Chang, RT; Xia, T; Gisolfi, CV. Fructose transport mechanism in humans. *Gastroenterology*, 1997;113:1171-1179.

[13] Born, P.The clinical impact of carbohydrate malabsorption. *Arab J Gastroenterol*, 2011;12:1-4.

[14] Hallffrisch, J. Metabolic effects of dietary fructose. *FASEB J*, 1990;4:332-339.

[15] Koo, HY; Wallig, MA; Chung, BH; Nara, TY; Cho, BH; Nakamura, MT. Dietary fructose induces a wide range of genes with distinct shift in carbohydrates and lipid metabolism in fed and fasted rat liver. *Biochim Biophys Acta*, 2008;1782:341-348.

[16] Chong, MF-F; Fielding, BA; Frayn, KN. Mechanisms for acute effect of fructose on postprandial lipemia. *Am J Clin Nutr,* 2007;85:1511-1520.

[17] McGilvery, RW. Fructose Metabolism. In: *Biochemistry. A functional approach*. Philadelphia. London. Toronto; W.B. Saunders Company; 1970, Pg 631-634.

[18] Parks, JE; Hellerstein, MK. Carbohydrate-induced hypertriacyl-glycerolemia: historical perspective and review of biological mechanisms. *Am J Clin Nutr,* 2000;71:412-433.

[19] Tran, LT; Yuen, VG; McNeil, JH. The fructose-fed rat: a review on the mechanisms of fructose-induced insulin resistance and hypertension. *Mol Cell Biochem,* 2009;330:219-228.

[20] Schaefer, EJ; Gleason, JA; Dansinger, ML. Dietary fructose and glucose differentially affect lipid and glucose homeostasis. *J Nutr,* 2009;139:1257S-1262S.

[21] Topping, DL; Mayes, PA. The immediate effects of insulin and fructose on the metabolism of the perfused liver. Changes in lipoprotein secretion, fatty acid oxidation and esterification, and lipogenesis, and carbohydrate metabolism. *Biochem J,* 1972;126:295-311.

[22] Kasumi, T; Vranic, M; Steiner, G. Triglyceride kinetics: effects of dietary glucose, sucrose, or fructose alone or with hyperinsulinemia. *Am J Physiol Endocrinol Metab,* 1986;250:E325-E330.

[23] Froesch, ER. Disorders of fructose metabolism. *Clin Endocrinol Metab,* 1976;5:599-611.

[24] Clements, RS; Morrison, AD; Winegrad, AI. Polyol pathway in aorta. Regulation by hormones. *Science,* 1969;166:1007-1008.

[25] Setchell, BP; Bassett, JM; Hinks, NT; Graham, NMcC. The importance of glucose in the oxidative metabolism of the pregnant uterus and its contents in conscious sheep with some preliminary observations on the oxidation of fructose and glucose by fetal sheep. *Q J Exp Physiol,* 1972;57:257-266.

[26] Teng, CC; Tjoa, S; Fennessey, PV; Wilkening, RB; Battaglia, FC. Transplacental carbohydrate and sugar alcohol concentrations and their uptakes in ovine pregnancy. *Exp Biol Med,* 2002;227:189-195.

[27] Bossolan, G; Trindade, CEP; Barreiros, RC. Blood galactose and glucose levels in mothers, cord blood, and 48-hour-old breast-fed full-term infants. *Neonatology,* 2007;91:121-126.

[28] Jauniaux, E; Hempstock, J; Teng, C; Battaglia, FC; Burton, G. Polyol concentrations in fluid compartments of the human conceptus during the first trimester of pregnancy: maintenance of redox potential in a low oxygen environment. *J Clin Endocrinol Metab,* 2005;90:1171-1175.

[29] Brusati, V; Józwik, M; Teng, C; Paolini, C; Marconi, AM; Battaglia, FC. Fetal and maternal non-glucose carbohydrates and polyols concentrations in normal human pregnancies at term. *Pediatr Res,* 2006;58:700-704.

[30] Brachet, EA. Presence of the complete sorbitol pathway in the human normal umbilical cord tissue. *Biol Neonate,* 1973;23:314-323.

[31] Meznarich, HK; Hay Jr, W; Sparks, JW; Meschia, G; Battaglia. FC. Fructose disposal and oxidation rates in the ovine fetus. *Q J Exp Physiol,* 1987;72:617-625.

[32] Aeberli, I; Zimermann, MB; Molinari, L; Lehmann, R; L'Allemand, D; Spinas, GA; Berneis, K. Fructose intake is a predictor of LDL particle size in overweight schoolchildren. *Am J Clin Nutr,* 2007;86:1174-1178.

[33] Trindade, CEP; Barreiros, RC; Kurokawa, C; Bossolan, G. Fructose in fetal cord blood and its relationship with maternal and 48-hour-newborn blood concentrations. *Early Hum Dev,* 2011;87:193-197.

[34] Scott, TW; Setchell, B; Bassett, JM. Characterization and metabolism of ovine fetal lipids. *Biochem J,* 1967;104:1040-1047.

[35] Carmona, A; Freedland, RA. Comparison among the lipogenic potential of various substrates in rat hepatocytes: the differencial effects of fructose-containing diets on hepatic lipogenesis. *J Nutr,* 1989;119:1304-1310.

[36] Chico, A; D'Alessandro, ME; Karabatas, L; Pastorale, C; Basabe, JC; Lombardo, YB. Muscle lipid metabolism and insulin secretion are altered in insulin-resistant rats fed a high sucrose diet. *J Nutr,* 2003;133:127-133.

[37] Lê, K-A; Ith, M; Kreis, R; Faeh, D; Bortolotti, M; Tran, C; Boesch, C; Tappy, L. Fructose overconsumption causes dyslipidemia and ectopic lipid deposition in healthy subjects with and without a family history of type 2 diabetes. *Am J Clin Nutr,* 2009;89:1760-1765.

[38] Tappy, L; Lê, KA; Tran, C; Paquot N. Fructose and metabolic diseases: New findings, new questions. *Nutrition,* 2010;26:1044-1049.

[39] Jeukendrup, AE. Carbohydrate intake during exercise and performance. *Nutrition,* 2004;20:669-677.

[40] Adopo, E; Peronnet, F; Massicotte, D; Brisson, GR; Hillaire, C. Respective oxidation of exogenous glucose and fructose given in the same drink during exercise. *J Appl Physiol,* 1994;76:1014-1019.

[41] Jeukendrup, AE; Moseley, L; Mainwaring, GI; Samuels, S; Perry, S; Mann, CH. Exogenous carbohydrate oxidation during ultraendurance exercise. *J Appl Physiol,* 2006;100:1134-1141.

[42] Currell, K; Jeukendrup, AE. Superior endurance performance with ingestion of multiple transportable carbohydrates. *Med Sci Sports Exerc,* 2008;40:275-281.

[43] Bergman, BC; Wolfel, EE; Butterfield, GE; Lopaschuk, GD; Casazza, GA; Horning, MA; Brooks, GA. Active muscle and whole body lactate kinetics after endurance training in men. *J Appl Physiol,* 1999;87:1684-1696.

[44] Van Hall, G; Jensen-Urstad, M; Rosdahl, H; Holmberg, HC; Saltin, B; Calbet, JA. Leg and arm lactate and substrate kinetics during exercise. *Am J Physiol Endocrinol Metab,* 2003;284:E193-E205.

[45] LeCoutre, V; Benoit, R; Carrel, G; Schutz, Y; Millet, GP; Tappy, L; Schneiter, P. Fructose and glucose co-ingestion during prolonged exercise increases lactate and glucose fluxes and oxidation compared with an equimolar intake of glucose. *Am J Clin Nutr,* 2010;92:1071-1079.

[46] Stanhope, KL; Griffen, SC; Bair, BR; Swarbrick, MM; Keim, NL; Havel, PJ. Twenty-four-hour endocrine and metabolic profiles following consumption of high-fructose corn syrup-, sucrose-, fructose-, and glucose-sweetened beverages with meals. *Am J Clin Nutr,* 2008;87:1194-1203.

[47] Herman, RH; Zakim, D; Stifel FB. Effect of diet on lipid metabolism in experimental animals and man. *Fed Proc,* 1970;29:1302-1307.

[48] Havel, PJ. Dietary Fructose: Implications for dysregulation of energy homeostasis and lipid/carbohydrate metabolism. *Nutr Rev,* 2005;63:133-157.

[49] Bantle, JP; Raatz, SK; Thomas, W; Georgepoulos, A. Effects of dietary fructose on plasma lipids in healthy subjects. *Am J Clin Nutr,* 2000;72:1128-1134.

[50] Sievenpiper, JL; Carleton, AJ; Chatha S; Jiang, HY; De Souza, RJ; Beyene, J; Kendall, CWC; Jenkins, DJA. Heterogeneous effects of fructose on blood lipids in individuals with type 2 diabetes. *Diabetes Care,* 2009;32:1930-1937.

[51] Stanhope, KL; Bremer, AA; Medici, V; Nakajima, K; Ito, Y; Nakano, T; Chen, G; Fong, TH; Lee, V; Menorca, RI; Keim, NL; Havel, P. Consumption of fructose and high fructose corn syrup increase postprandial triglycerides, LDL-cholesterol, and apolipoprotein-B in young men and women. *J Clin Endocrinol Metab,* 2011;96:E1596-1605.

[52] Livesey, G; Taylor, R. Fructose consumption and consequences for glycation, plasma triacylglycerol, and body weight: meta-analyses and meta-regression models of intervention studies. *Am J Clin Nutr, 2008;88:1419-1437.*

[53] Marriott, BP; Cole, N; Lee, E. National estimates of dietary fructose intake increased from 1977 to 2004 in the United States. *J Nutr,* 2009;139:1228-1235.

[54] Elliott, SS; Keim, NL; Stern, JS; Teff, K; Havel, PJ. Fructose, weight gain, and the insulin resistance syndrome. *Am J Clin Nutr,* 2002;76:911-922.

[55] Swarbrick, MM; Stanhope, KL; Elliott, SS; Graham, JL; Krauss, RM; Christiansen, MP; Griffen, SC; Keim, NL; Havel P. Consumption of fructose-sweetened beverages for 10 weeks increases postprandial triacylglycerol and apolipoprotein-B concentrations in overweight and obese women. *Br J Nutr,* 2008;100:947-952.

[56] Aeberli, I; Zimmermann, MB; Molinari, L; Lehman, R; L'Allemand, D; Spinas, GA; Berneis, K. *Am J Nutr,* 2007;86:1174-1178.

[57] L'Allemand-Jander, D. Clinical diagnosis of metabolic and cardiovascular risks in overweight children; early development of chronic diseases in the obese child. *Int J Obes* (Lond), 2010;34(Suppl):S32-S36.

[58] Jürgens, H; Haass, W; Castañeda, TR; Schürmann, A; Koebnick, C; Dombrowski, F; Otto, B; Nawrocki, AR; Scherer, PE; Spranger, J; Ristow, M; Joost, H-G; Havel, PJ; Tschöp, H. Consuming fructose-sweetened beverages increases body adiposity in mice. *Obes Res,* 2005;13:1146-1156.

[59] Teff, KL; Elliott, SS; Tschop, M; Kieffer, TJ; Rader, D; Heiman, M; Townsend, RR; Keim, NL; D´Alessio, D; Havel, PJ. Dietary fructose reduces circulating insulin and leptin, attenuates postprandial suppression of ghrelin, and increases triglycerides in women. *J Clin Endocrinol Metab,* 2004;89:2963-2972.

[60] Stanhope, KL; Schwarz, JM; Keim, Nl; Griffen, SC; Bremer, AA; Graham, JL; Hatcher, B; Cox, CL; Dyachenko, A; Zhang, W; Mc Graham, JL; Seibert, A; Kraus, RM; Chiu, S; Schaefer, EJ; Ai, M; Otokozawa, S; Nakajima, K; Nakano, T; Beysen, C; Hellerstein, MK; Berglund, L; Havel, PJ. Consuming fructose-sweetened, not glucose-sweetened beverages increases visceral adiposity and lipids and decreases insulin sensitivity in overweight/obese humans. *J Clin Invest,* 2009;119:1322-1334.

[61] Bantle, JP; Laine, DC; Castle, GW; Thomas, JW; Hoogvert, BJ; Goetz, FC. Postprandial glucose and insulin responses to meals containing different carbohydrates in normal and diabetic subjects. *N Engl J Med,* 1983;309:7-12

[62] Havel, PJ. Control of energy homeostasis and insulin action by adipocyte hormones: leptin, acylation stimulating protein, and adiponectin. *Curr Opin Lipidol,* 2002;13:51-59.

[63] .Teff, KL; Grudziak, J; Townsend, RB; Dunn, TN; Grant, RW; Adams, SH; Keim, NL; Cummings, BP; Stanhope, KL; Havel, PJ. Endocrine and metabolic effects of consuming fructose- and glucose-sweetened beverages with meals in obese men and women: influence of insulin resistance on plasma triglyceride responses. *J Clin Endocrinol Metab,* 2009;94:1562-1569.

[64] Cirillo, P; Sato, W; Reungjui, S; Heinig, M; Gersch, M; Sautin, Y; Nakagawa, T; Johnson, RJ. Uric acid, the metabolic syndrome, and renal disease. *J Am Soc Nephrol,* 2006;17(12 Suppl 3):S165-S168.

[65] Johnson, RJ; Perez-Pozo, SE; Sautin, YY; Manitius, J; Sanchez-Lozada, LG; Feig, DI; Shafiu, M; Segal, M; Glassock, RJ; Shimada, M; Roncal, C; Nakagawa, T. Hypothesis: could excessive fructose intake and uric acid cause type 2 diabetes? *Endocr Rev,* 2009;30:96-116.

[66] Johnson, RJ; Sanchez-Lozada, LG; Nakagawa, T. The effect of fructose on renal biology and disease. *J Am Soc Nephrol,* 2010;21:2036-2039.

[67] Nakagawa, T; Hu, H; Zharikov, S; Tuttle, KR; Short, RA; Glushakova, O; Ouyang, X; Feig, DI; Block, ER; Herrera-Acosta, J; Patel, JM; Johnson, RJ. A causal role for uric acid in fructose-induced metabolic syndrome. *Am J Physiol Renal Physiol,* 2006;290:F625-F631.

[68] Graessler, J; Graessler, A; Unger, S; Kopprasch, S; Tausche, AK; Kuhlisch, E; Schroeder, HE; Feig, DI; Johnson, RJ. The role of uric acid in pediatric hypertension. *J Ren Nut,* 2007;17:79-83.

[69] Feig, DD; Johnson RJ. The role of uric acid in pediatric hypertension. *J Ren Nutr,* 2007;17:79-83.

[70] Khosla, UM; Zharikov, S; Finch, JL; Nakagawa, T; Roncal, C; Mu, W; Krotova, K; Block, ER; Prabhakar, S; Johnson, RJ. Hyperuricemia induces endothelial dysfunction. *Kidney Int,* 2005;67:1739-1742.

[71] Bunag, RD; Tomita, T; Sasaki, S. Chronic sucrose ingestion induces mild hypertension and tachycardia in rats. *Hypertension,* 1983;5:218-225.

[72] Martinez, FJ; Rizza, RA; Romero, JC. High-fructose feeding elicits insulin resistance, hyperinsulinism, and hypertension in normal mongrel dogs. *Hypertension,* 1994;23: 456-463.

[73] Kamide, K; Rakugi, H; Higaki, J; Okamura, A; Nagai, M; Moriguchi, K; Ohishi, M; Satoh, N; Tuck, ML; Ogihara, T. The rennin-angiotensin and adrenergic nervous system in cardiac hypertrophy in fructose-fed rats. *Am J Hypertens,* 2002;15:66-71.

[74] Miller, AW; Sims, JJ; Canavan, A; Hsu, T; Ujhelyi, MR. Impaired vagal reflex activity in insulin-resistance rats. *J Cardiovasc Pharmacol,* 1999;33:698-702.

[75] Sanchez-Lozada, LG; Tapia, E; Jimenez, A; Bautista, P; Cristobal, M; Nepomuceno, T; Soto, V; Avila-Casado, C; Nakagawa, T; Johnson, RJ; Herrero-Acosta, J; Franco, M. Fructose-induced metabolic syndrome is associated with glomerular hypertension and renal microvascular damage in rats. *Am J Physiol Renal Physiol,* 2007;292:F423-F429.

[76] Brown, CM; Dulloo, AG; Yepuri, G; Montani, JP. Fructose ingestion acutely elevates blood pressure in healthy young humans. *Am J Physiol Regul Integr Comp Physiol,* 2008;294:R730-R737.

[77] Brown, IJ; Stamler, J; Van Horn, L; Robertson, CE; Chan, Q; Dyer, AR; Huang, C-C; Rodriguez, BL; Zhao, L; Daviglus, ML; Ueshima, H; Elliott, P. International Study of Macro/Micronutrients and Blood Pressure Research Group. Sugar-Sweetened Beverage, Sugar Intake of Individuals, and their Blood Pressure International Study of Macro/Micronutrients and Blood Pressure. *Hypertension,* 2011;57:695-701.

[78] Johnson, RJ; Segal, MS; Sautin, Y; Takahiko, N; Feig, DI; Kang, DH; Gersch, MS; Benner, S; Sánchez-Lozada, L. Potential role of sugar (fructose) in the epidemic of hypertension, obesity and the metabolic syndrome, diabetes, kidney disease, and cardiovascular disease. *Am J Clin Nutr,* 2007;86:899-906.

[79] Mazzali, M; Kanellis, J; Han, L; Feng, L; Xia, YY; Chen, Q; Kang, DH; Gordon, KL; Watanabe, S; Nakagawa, T; Lan, HY; Johnson RJ. Hyperuricemia induces a primary renal arteriolopathy in rats by a blood pressure-independent mechanism. *Am J Physiol Renal Physiol,* 2002;282:F991-F997.

[80] Watanabe, S; Kang, DH; Feng, L; Nakagawa, T; Kanellis, J; Lan, H; Mazzali, M; Johnson, RJ. Uric acid, hominoid evolution and the pathogenesis of salt-sensivity. *Hypertension,* 2002:40:355-60.

[81] Quiroz, Y; Pons, H; Gordon, KL; Rincón, J; Chávez, M; Parra, G; Herrera-Acosta, J. Mycophenolate mofetil prevents salt-sensitivite hypertension resulting from nitric synthesis inhibition. *Am J Physiol Renal Physiol,* 2001;281:F38-F47.

[82] Soleimani, M. Dietary fructose, salt absorption and hypertension in metabolic syndrome: towards a new paradigm. *Acta Physiol,* 2011;201:55-62.

[83] Chen, L; Caballero, B; Mitchell, DC; Loria, C; Lin, P-H; Champagne, CM; Elmer, PJ; Ard, JD; Batch, BC; Anderson, CAM; Appel, LJ. Reducing consumption of sugar-sweetened beverages is associated with reduced blood pressure: a prospective study among United States adults. *Circulation,* 2010;121:2398-2406.

[84] Meigs, JB. Epidemiology of type 2 diabetes and cardiovascular disease: translation from population to prevention: The Kelly West award lecture, 2009. *Diabetes Care,* 2010;33:1865-1871.

[85] Alberti, KG; Zimmet, P; Shaw, J. The metabolic syndrome: a new worldwide definition. IDF Epidemiology Task Force Consensus Group. *Lancet,* 2005; **366:**1059-1062.

[86] Antonoupolos, S. National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). Third Report of the National Cholesterol Education (NCEP) Expert Panel on Detection, Evaluation, and treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. *Circulation,* 2002;106:3143-3421.

[87] Grundy, SM; Cleeman, JI; Daniels, SR; Donato, KA; Eckel, RH; Franklin, BA; Gordon, DJ; Krauss, RM; Savage, PJ; Smith Jr, SC; Spectus, JA; Costa, F. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute Scientific Statement. *Circulation,* 2005;**112:**2735-2752 [published corrections appear in Circulation 2005;112:e297 and Circulation 2005;112:e298].

[88] Bremer, AA; Stanhope, KL; Graham, BS; Cummings, BP; Wang, W; Saville, BR; Havel, PJ. Fructose-Fed Rhesus Monkeys: A Nonhuman Primate Model of Insulin Resistance, Metabolic Syndrome, and Type 2 Diabetes. *Clin Trans Sci,* 2011; 4:243-252.

[89] Collino, M. High dietary fructose intake: sweet or bitter life? *World J Diabetes,* 2011;15:77-81.

[90] Korandji, C; Zeller, M; Guilland, JC; Collin, B; Lauzier, B; Sicard, P; Duvillard, L; Goirand, F; Moreau, D; Cottin, Y; Rochette, L; Vergel, YC. Time course of asymmetric dimethylarginine (ADMA) and oxidative stress in fructose-hypertensive rats: A model related to metabolic syndrome. *Atherosclerosis,* 2011;214:310-315.

[91] Abdelmalek, MF; Suzuki, A; Guy, C; Unalp-Arida; A; Colvin, R; Johnson, RJ; Diehl, AM. Increased fructose consumption is associated with fibrosis severity in patients with nonalcoholic fatty liver disease. *Hepatology* 2010; 51:1961-1971.

[92] Pradhan, A. Obesity, Metabolic Syndrome, and Type 2 Diabetes: Inflammatory Basis of Glucose Metabolic Disorders. *Nutr Rev,* 2007;65: S152-156.

[93] Kanuri, G; Spruss, A; Wagnerberger, S; Bischoff, SC; Bergheim, I. Role of tumor necrosis factor α (TNFα) in the onset of fructose-induced nonalcoholic fatty liver disease in mice. *J Nutr Biochem,* 2011; 22: 527-534.

[94] Stanhope, KL. Role of Fructose-Containing Sugars in the Epidemics of Obesity and Metabolic Syndrome. *Annu Rev Med,* 2011; Jan 26. [Epub ahead of print].

[95] Nagai, Y; Yonemitsu, S; Erion, DM; Iwasaki, T; Stark, R; Weismann, D; Dong, J; Zhang, D; Jurczak, MJ; Löffler, MG; Cresswell, J; Yu, XX; Murray, SF; Bhanot, S; Monia, BP; Bogan, JS; Samuel, V; Shulman, GI. The role of peroxisome proliferator-activated receptor gamma coactivator-1 beta in the pathogenesis of fructose-induced insulin resistance. *Cell Metab*, 2009;9: 252-264.

[96] Lin, J; Handschin, C; Spiegelman, BM. Metabolicc control through the PGC-1 family of transcription coativators. *Cell Metab*, 1: 361-370, 2005.

[97] White, S. Straight talk about high-fructose corn syrup: what it is and what it ain't. *Am J Clin Nutr*, 2008;88(Suppl):1716S-1721S.

[98] Wiernsperger, N; Geloen, A; Rapin, J-R. Fructose and cardiometabolic disorders: The controversy will, and must, continue. *Clinics*, 2010;65:729-738.

[99] Johnson, RJ; Segal, S M; Sautin, Y; Nakagawa, T; Feig, DI; Kang, D-H; Gersch, MS; Benner, S; Sánchez-Lozada, LG. Potential role of sugar (fructose) in the epidemic of hypertension, obesity and the metabolic syndrome, diabetes, kidney disease, and cardiovascular disease. *Am J Clin Nutr*, 2007;86:899-906.