Bachelorarbeit

Fructose and weight gain

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Abstract

The prevalence of obesity is growing in the United States and worldwide. In this thesis the effects of the sugar fructose on weight gain and obesity are explored. The transport and uptake of fructose is based on two different systems, one that is related to insulin, and another that works in an insulin-insensitive manner. The role of the different glucose transport proteins, especially that of GLUT 5 in fructose uptake, are considered.

The hormones insulin and leptin could contribute to weight gain as they both act in the central nervous system in the regulation of eating behaviour and energy homeostasis. Fructose consumption induces dyslipidemia, insulin resistance, visceral adiposity and hypertension, which are all common features of the metabolic syndrome. Different markers of altered lipid metabolism due to fructose consumption have been under investigation, the main focus lying on TG, apoB, and LDLs. Fructose has also been shown to alter de novo lipogenesis. All these fructose-induced changes are associated with weight gain and obesity.

Abbreviations

2DG	2-deoxy-glucose
3OMG	3-O-methyl-glucose
apoB	apolipoprotein B-100
ATP	adenosine triphosphate
CB	cytochalasin B
DNL	de novo lipogenesis
FFA	free fatty acid
GAP	glyceraldehyde-3-phosphate
GI	glycemic index
GLUT	glucose transport protein
HFCS	high fructose corn syrup
LDM	low density microsome
LPL	lipoprotein lipase
MetS	metabolic syndrome
NAFLD	nonalcoholic fatty liver disease
NASH	nonalcoholic steatohepatitis
PI-3K	phosphatidylinositol 3-kinase
PKA	protein kinase A
PM	plasma membrane
PTP-1B	protein tyrosine phosphatase-1B
sdLDL	small dense LDL
SREBP-1c	sterol receptor element binding protein-1c
STZ	streptozotocin
TG	triacylglycerol, triglyceride
VLDL	very low density lipoprotein

1. Introduction

1.1 Fructose

The monosaccharide fructose was first discovered by Augustin-Pierre Dubrunfaut in 1847. As many fruits, such as berries or melons, contain fructose, it is also called fruit sugar. But fructose is not only an ingredient of fruits, also honey and vegetables contain fructose. Fructose is mainly used as a sweetener in foods and beverages and its content in soft drinks is high. An advantage of fructose is its lower glycemic index compared to that of the sugar glucose, making it ideal as a sweetener for patients with diabetes mellitus [1]. The effects of carbohydrates such as glucose or fructose on blood sugar levels are described by the glycemic index. By definition, the glycemic index is a measure of the effects of carbohydrates on blood sugar levels. A high glycemic index is correlated with a fast break-down of carbohydrates and thus a rapid release of glucose into the blood.

There is a distinction between crystalline fructose and high fructose corn syrup (HFCS). The purified monosaccharide is termed crystalline fructose. HFCS is produced by the enzymatic isomerization of dextrose to fructose [2] and refers to a mixture of fructose and glucose in varying amounts. However, fructose is mainly consumed in form of HFCS.

Fructose belongs to the group of carbohydrates. It is a naturally occurring chemical compound and its molecular formula is $C_6H_{12}O_6$. As it is a monosaccharide containing six carbon atoms, it belongs to the group of hexoses. Sugars having a ketone group are called ketohexoses, thus fructose is a ketohexose. Glucose belongs to the group of aldohexoses, because it has an aldehyde group. Fructose, which mainly occurs in combination with glucose, is an isomer. It has the same molecular formula $C_6H_{12}O_6$ as glucose, but their structures are different [3].

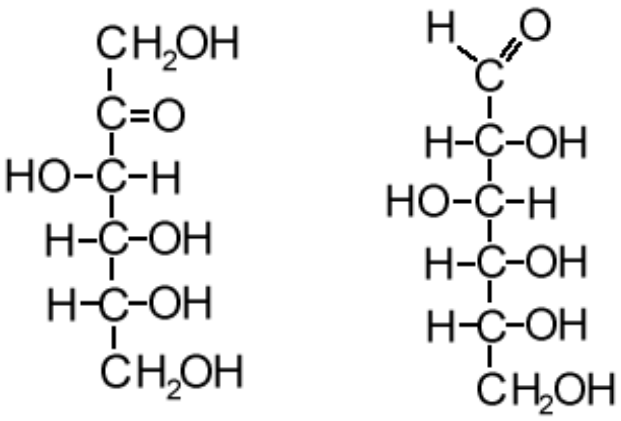

Figure 1. Structure of D-fructose and D-glucose

The condensation of those two components forms the disaccharide sucrose [1].
Along with glucose and galactose, fructose is one of the three most important blood sugars. Fructose is not only involved in glycolysis and gluconeogenesis, in general, it is essential for metabolic functions [4]. The metabolism of fructose appears to be different in different tissues depending on the enzymes involved. There are two different metabolic pathways for the degradation of fructose due to the diverse enzymes in muscles and liver [3]. However, the metabolism of fructose predominantly occurs in the liver [4].

1.2 Fructose metabolism

Via the portal vein absorbed fructose, whose uptake from portal circulation is greater than that of glucose [1], is delivered to the liver where fructose metabolism takes place [2]. In contrast to glucose, the uptake of fructose by the liver is not regulated by insulin [5], because fructose metabolism bypasses phosphofructokinase [1]. So fructose neither stimulates insulin secretion nor is insulin needed for the initial steps of its hepatic metabolism [1]. The fructose uptake by the liver is not limited by energy status like cytosolic ATP and citrate levels, because fructose metabolism does not depend on phosphofructokinase regulation [6].
The enzyme fructokinase accomplishes the phosphorylation of fructose by adenosine triphosphate (ATP) to form fructose-1-phosphate. In the liver aldolase B splits fructose-1-phosphate into triose phosphates, dihydroxyacetone phosphate and glyceraldehyde, which both can be converted to glyceraldehyde-3-phosphate (GAP) [2,3]. As mentioned above, the hepatic entry of fructose bypasses the main rate-controlling step of glycolysis catalyzed by phoshpofructokinase. So fructose is an unregulated source of GAP and thus also serves as an unregulated source of hepatic acetyl-CoA production.
In the glycolytic pathway of the liver fructose metabolism leads to 4 products termed glucose, glycogen, lactate, and pyruvate [2].
While seven enzymes are involved in fructose metabolism in liver, in muscles as well as in adipocytes, the enzyme hexokinase is responsible for the phosphorylation of fructose to fructose 6-phosphate [7,3]. In adipose tissue the type II hexokinase is the most abundant hexokinase [8].

2. Fructose transport and uptake in adipocytes

2.1 General facts

The transport systems of hexoses, especially those for fructose, into adipocytes have to be defined.

The transport of fructose is distinct from the transport of glucose into adipocytes. While the transport of glucose into adipocytes is based on a single transport system, fructose can enter adipocytes by two separate mechanisms. One of these transporters is sensitive to insulin and therefore probably the glucose transporter, while the other mechanism is insulin-insensitive [5].

The transport of glucose into adipocytes is sensitive to insulin [5]. In rat adipocytes the stimulation with insulin led to an increase of the maximum velocity of transport. However, insulin did not induce any change in the glucose concentration required for half-maximal rates of transport (K_t). In contrast to glucose, the influence of insulin on fructose metabolism is lower, indicating that there must also be an insulin-insensitive system. While a rising hexose concentration did not elicit any change in glucose uptake or metabolism, the uptake of fructose increased. The maximum velocity of transport and the concentration required for half-maximal rates of transport are higher for fructose than for glucose, but its affinity for the transport system is lower. However, in presence of insulin there is an interaction between the hexoses, leading to the assumption that one of the transport systems of fructose is dependent on glucose and therefore sensitive to insulin [5].

Different glucose transport proteins (GLUTs) being structurally related are in charge of up-taking sugars across the plasma membrane of most mammalian cells. There are five major forms of those transport proteins termed GLUT 1-5, of which GLUT 4 is the most studied one so far. GLUT 4 and GLUT 1 are both CB-sensitive transporters mediating glucose uptake [7]. GLUT 4 is the insulin-sensitive glucose transporter and therefore regulated by it. Depending on the amount of insulin in the blood, the localization of GLUT 4 changes. A meal causes an increase in blood insulin, which is correlated to a fast translocation of GLUT 4. Under conditions of low insulin, GLUT 4 is located in intracellular storage vesicles, from where GLUT 4 is translocated to the plasma membrane with increasing blood insulin levels. The cellular glucose uptake is stimulated by insulin due to the increased cell surface GLUT 4.

GLUT 1 is also expressed by adipocytes. Compared to GLUT 4, the proportion of GLUT 1 in the plasma membrane is already high in unstimulated cells. Therefore the influence of insulin is lower than for GLUT 4, which is considered to be the most important glucose carrier in fat cells [7].

As mentioned before, the uptake of fructose into fat cells depends on at least two separate transport mechanisms [7], because fructose metabolism was not stimulated by insulin in all studies. Compared to glucose, the magnitude of insulin stimulation on fructose metabolism was less. In presence of glucose, mannose, or 2-deoxyglucose, insulin could activate glycogen synthase in rat adipocytes, however no stimulation by insulin occurred when 5mM fructose was present [5]. So fructose uptake is just marginally stimulated by insulin while a carrier being insensitive to both insulin and cytochalasin B (CB) mediates a much larger proportion of fructose uptake into adipocytes. More precisely, about 80% of the fructose uptake is insensitive to CB [7]. Due to the fact that GLUT 1 and GLUT 4 are CB-sensitive and therefore not appropriate for the fructose uptake, it can be concluded that there must be another hexose transporter being specified on the transport of fructose. This was shown in a study, where GLUT 1 was expressed in Xenopus oocytes [7]. The carrier that is neither inhibited by CB nor regulated by insulin is the glucose transporter GLUT 5. GLUT 5 mRNA is abound in human adipose tissue and operates as a transporter of fructose. The plasma membrane localisation of GLUT 5 in both human skeletal muscle and rat adipocytes is similar [7]. Thus, GLUT 5 is expressed in rat adipocytes and therefore facilitates fructose uptake and metabolism in this tissue [7]. GLUT 5 mainly transports fructose, while its affinity for glucose is low. It is required for the absorption of fructose in the intestine. With higher fructose concentration in rats or mice, either achieved by feeding a diet high in fructose or by perfusing them with a high fructose concentration, the expression of GLUT 5 in the small intestine rises [4]. mRNA expression of the transporter GLUT 5 was also increased in brains of rats after feeding a diet high in fructose [1].

The expression of GLUT 2, another glucose transport protein, whose affinity for fructose is moderate and whose affinity for glucose is high, also increases in the intestine of rats or mice with increased fructose concentration. Due to an increase in fructose or glucose in the intestinal lumen, GLUT 2 is translocated from the basolateral membrane or the cytoplasm of enterocytes, which are found in the small intestine or colon, to the apical site of the membrane facing the lumen. Although it is certain that GLUT 2 and GLUT 5 transport fructose *in vitro*, the role of GLUT 5 *in vivo* in transporting fructose and being responsible for the absorption of fructose in the intestine is speculative [4].

2.2 GLUT 5 expression in rat adipocytes

The analytical technique immunoblotting, also called western blotting, was used for the assessment of the expression of GLUT 5 in rat adipocytes.

After incubation of rat adipocytes in presence and absence of insulin, they were subjected to subcellular fractionation. These fractions of either control or insulin-treated adipocytes were immunoblotted. In accordance with the immunoblots in *figure 2* it is obvious that insulin treatment has a great influence on GLUT 4. It is detected in intracellular membranes in unstimulated adipocytes. But after stimulation with insulin, GLUT 4 is translocated to the plasma membrane (PM) and simultaneously GLUT 4 decreases in the low density microsomes (LDM). GLUT 1 is hardly influenced by insulin treatment, but its amount in the PM of unstimulated adipocytes is already high.

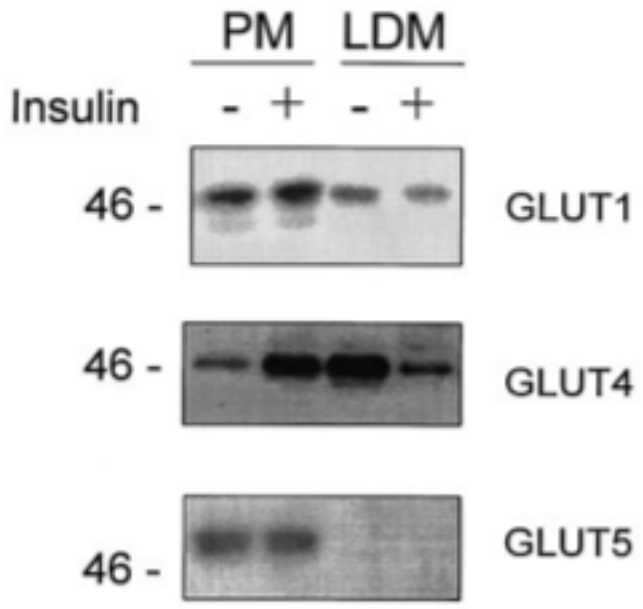

Figure 2. Influence of insulin on GLUT 1, GLUT 4 and GLUT 5 [7]

However, there was no immunoreactivity of GLUT 5 in the LDM fraction of the plasma membrane and no insulin-stimulated changes of GLUT 5 expression in the PM, proving that GLUT 5 is distinct from both GLUT 1 and GLUT 4. GLUT 5 is not detectable in intracellular regions. Insulin induces changes in GLUT 1 and GLUT 4 even if the changes are small. However, insulin does not elicit any alteration in GLUT 5 abundance in the PM due to the lack of intracellular GLUT 5. Membranes prepared from rat jejunum, which are enriched with GLUT 5, were used to prove that the detected GLUT 5 band did not represent an experimental artefact [7].

2.3 Fructose uptake

To show how fructose uptake into rat adipocytes takes place, the effect of insulin, cytochalasin B (CB), fructose, glucose, and the two glucose analogues 2-deoxy-glucose (2DG) and 3-O-methyl-glucose (3OMG) were determined. Cytochalasin B is a toxic metabolite inhibiting cytoplasmic and nuclear division as well as cell movement. *Figure 3* shows the time course for fructose uptake in isolated rat adipocytes in absence and presence of 2DG. The addition of 30mmol/l 2DG significantly suppressed the uptake of fructose. Without 2DG being present the fructose uptake was increasing linearly [7]. However, the influence of 2DG on fructose metabolism in adipocytes was rather small, while it strongly affects glucose metabolism [5].

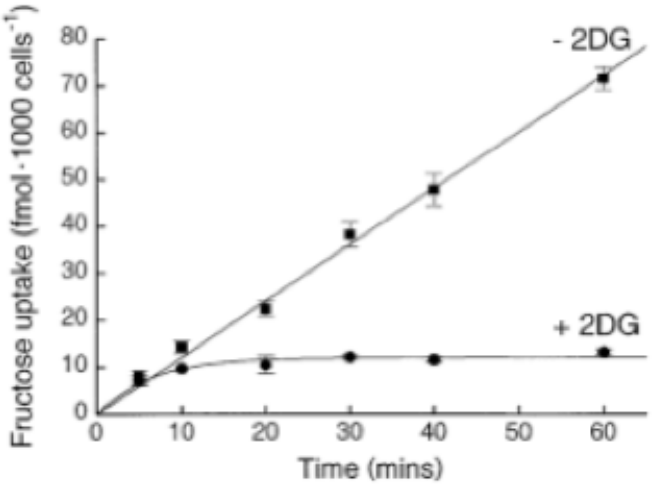

Figure 3. Time course for fructose uptake in isolated rat adipocytes in presence and absence of 2DG [7]

In the following study the effects on the uptake of 50 µmol/l [^{14}C]-fructose were investigated over a 40 min period.

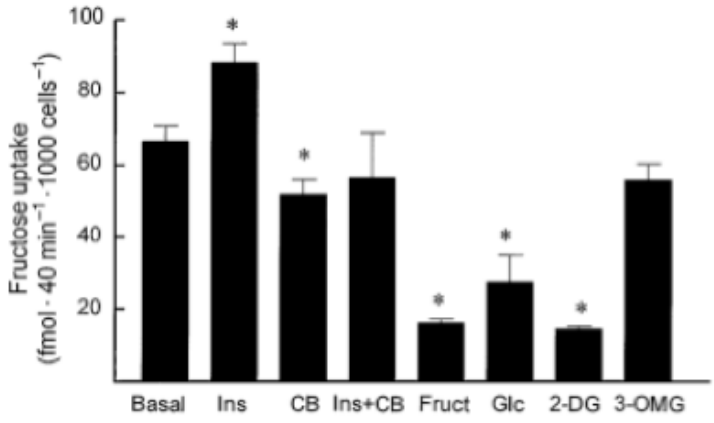

Figure 4. Effect of insulin, cytochalasin B (CB), fructose, glucose, 2DG, and 3OMG on fructose uptake in rat adipocytes [7]

While 100 nmol/l insulin stimulated fructose uptake in such a way that there was an increase about 35%, 10 µmol/l CB caused a decrease of fructose uptake by 21%. The presence of 30 mmol/l fructose, glucose or 2DG resulted in substantially suppressed fructose uptake, while fructose uptake by 3OMG was only inhibited by around 16%. These effects are shown in *figure 4* [7]. The lower the concentration of CB, which inhibited the transport of fructose, the higher was the inhibition [5].

Insulin increases the maximum velocity of fructose transport, which is shown in *figure 5*, where the fructose concentration is plotted against the rate of fructose transport inhibited by CB [5].

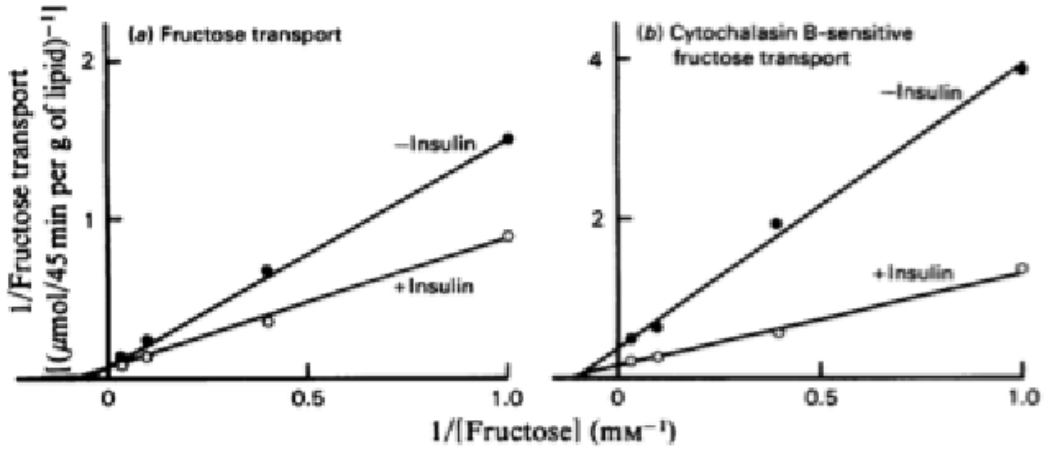

Figure 5. Effect of insulin on fructose transport and CB-sensitive fructose transport [5]

When both insulin and CB affect fructose uptake, CB abolishes the increase, which would be caused by insulin [7]. The effect of insulin on fructose transport is also abolished by 2DG [5]. These results show that fructose uptake in adipocytes is partly insulin sensitive, however, a much larger proportion of the total fructose uptake is insensitive to CB [7].

Not just 2DG, but also D-glucose and unlabelled fructose suppressed the uptake of [^{14}C]-fructose as can be seen in *figure 4*. An inhibition in hexokinase activity, being responsible for the phosphorylation of fructose to fructose 6-phosphate in adipocytes, may follow from the presence of 2DG or glucose during fructose uptake. Due to their presence, glucose 6-phosphate and 2DG phosphate is accumulated, thus leading to inhibition of hexokinase, which further results in the reduction of the transmembrane fructose gradient [7]. Therefore the uptake of sugar into fat cells, which the transmembrane fructose gradient is responsible for, is disturbed [7].

Since 3OMG is not phosphorylated, the effect of 3OMG is similar to that of CB. CB inhibits the fructose uptake by 21%, while 3OMG leads to a reduction of fructose uptake into fat cells by 16% (*figure 4*) [7]. 3OMG also inhibits the stimulation of insulin

on fructose transport. While unlabelled fructose, glucose, and its analogues have an inhibitory effect on fructose uptake into adipocytes, sucrose does not suppress the uptake. Thus osmotically-induced changes in cell volume are not related to the uptake of fructose seen in the presence of different hexoses [7].

2.4 GLUT 5 expression and fructose uptake in adipocytes from diabetic rats

According to several studies, the induction of diabetes reduces the expression of the insulin-regulated glucose transporter GLUT 4. It was investigated if the induction of diabetes also influences expression of GLUT 5 which is not regulated by insulin. Therefore streptozotocin, a naturally chemical compound inducing experimental diabetes in animals, is used. It has been observed if STZ-induced diabetes affects the expression of GLUT 5 and fructose uptake.

The immunoblots in *figure 6A* show both GLUT 4 and GLUT 5 in STZ-induced diabetic rats and control (C) rats in plasma membrane (PM) and low density microsome (LDM) fractions. As is observable in *figure 6A*, the injection of STZ resulted in a reduction in PM and LDM GLUT 4 content. The densiometric quantification of GLUT 5 in *figure 6B* shows that also the GLUT 5 abundance in the PM was reduced by about 75% after injection of STZ.

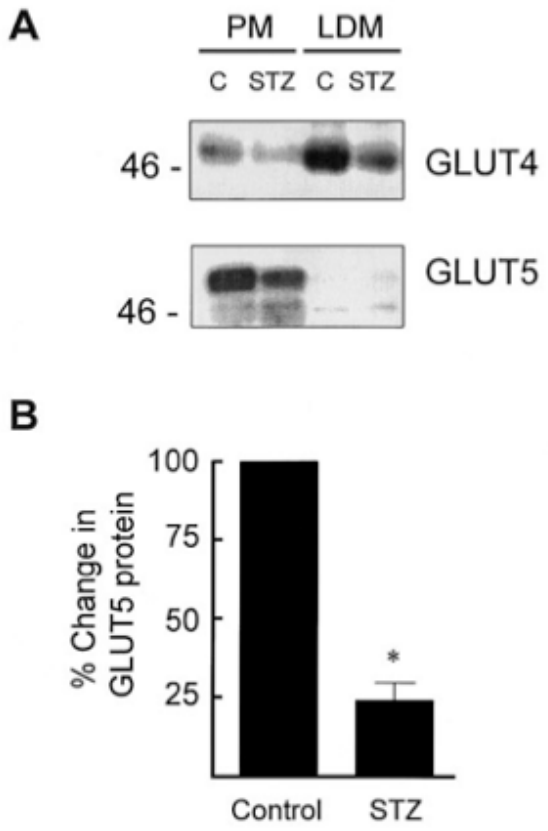

Figure 6A. GLUT 4 and GLUT 5 in control and diabetic rats
B. Densiometric quantification of GLUT 5 abundance in control and diabetic rats [7]

To further investigate if the expression of GLUT 5 may be responsive to blood insulin or glucose alterations, it has been observed if fructose uptake, which is related to a reduction in GLUT 5 expression [7], is reduced in adipocytes. Those adipocytes, which were isolated from diabetic rats, showed a decrease in fructose uptake of approximately 50% [7].

The relation between GLUT 5 expression and fructose uptake was also shown in a study with mice. Mice which were fed with a diet high in fructose had an up-regulated expression of the GLUT 5 transporter [4]. A high fructose perfusion into rats also increased the activity of GLUT 5 and abundance of mRNA [9]. Due to their higher GLUT 5 expression in their small intestine or jejunum, their capacity of uptaking fructose was increased [4]. The longer the influence of fructose lasts, the greater is the increase in fructose uptake [9].

The influence of glucose, when consumed along with fructose, on the fructose uptake was investigated [1]. Due to the increase in blood glucose in diabetic rats, glycaemia may also be involved in the regulation of GLUT 5 expression, of which a reduction is related to changes in fructose uptake. Vanadate, having insulin-like effects, was used to treat the high blood glucose in the diabetic rats. This correction with vanadate resulted in a restoration in GLUT 5 expression and fructose uptake due to the reduction in blood glucose. Blood glucose showed a decrease of about 40%. Although glucose is a modulator for expression of GLUT 5 in the human colonic and choriocarcinoma (placental) cell line, its effects on GLUT 5 expression in rat adipocytes is not yet completely understood. However, alterations in blood glucose may be responsible for the down-regulation of GLUT 5 in rat fat cells after inducing diabetes with streptozotocin [7].

2.5 Expression of GLUT 5 in mice

To ascertain the role of the glucose transport protein GLUT 5 in fructose transport and fructose absorption, mice were fed with a diet high in fructose. The role of GLUT 5 in fructose absorption was then determined comparing wild-type mice (GLUT $5^{+/+}$) with GLUT 5 knock-out mice (GLUT $5^{-/-}$), which were lacking the GLUT 5 gene (Slc2a5).

The expression of GLUT 5 in both the wild-type and knock-out mice is shown in *figure 7 (northern hybridization).*

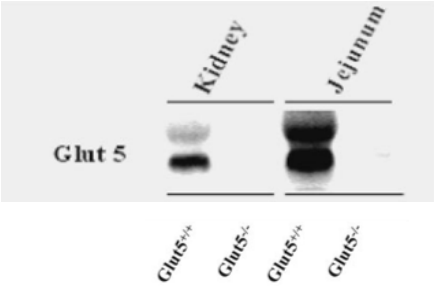

Figure 7. Expression of GLUT 5 in wild-type (GLUT $5^{+/+}$) and knock-out (GLUT $5^{-/-}$) mice [4]

In wild-type mice there is an expression of GLUT 5 in both, the jejunum and kidney, the abundance in the kidney being lower. However, in mice lacking the GLUT 5 gene, no expression of GLUT 5 is observable.

Additionally, in the jejunum of knock-out mice no labelling with immunofluorescence of GLUT 5 was detected, while the wild-type mice showed apical labelling as can be seen in *figure 8.*

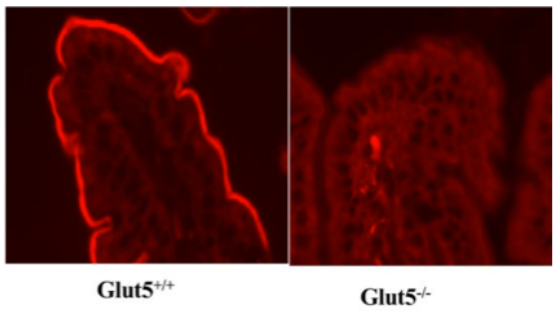

Figure 8. GLUT 5 labelling in the jejunum of wild-type and knock-out mice [4]

The wild-type as well as the GLUT 5-ko mice were fed a diet high in fructose (60% fructose) and as a reference a normal (60% starch) diet

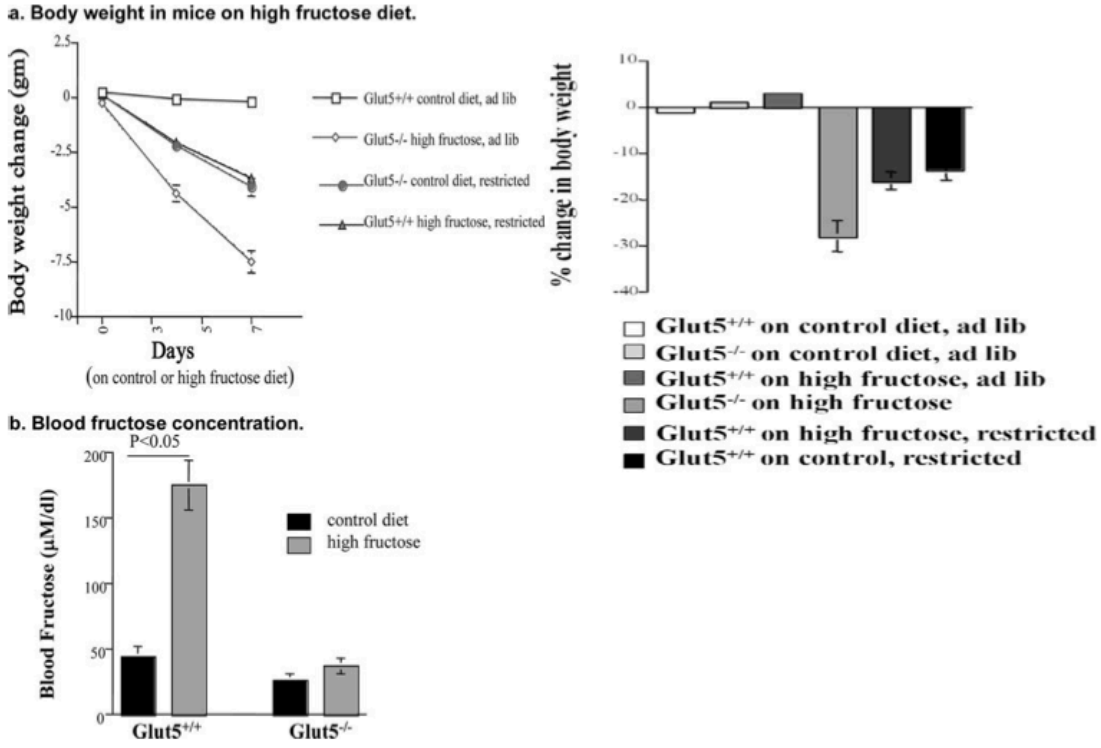

Figure 9a. Body weight change in GLUT 5 wild-type and knock-out mice fed a high-fructose and control diet
b. Blood fructose concentration in GLUT 5 wild-type and knock-out mice fed a high-fructose and control diet [4]

As is observable in *figure 9* the mice lacking the GLUT 5 gene lost weight fed with the diet high on fructose relative to the control diet with starch, while wild-type mice had a constant body weight. The weight loss of the GLUT $5^{-/-}$ is an indication for the role of GLUT 5 being an essential element for the absorption of fructose in the intestine. Due to the absence of the GLUT 5 gene, the fructose was not absorbed from the intestine leading to the loss of weight. For the further indication of GLUT 5 being essential for fructose uptake, fructose concentrations in blood were measured in both wild-type and knock-out mice. As GLUT 5 is required for fructose absorption, in GLUT $5^{+/+}$ mice having the GLUT 5 gene the concentration of fructose in blood increased with the feeding of the diet high in fructose. While in GLUT $5^{-/-}$ mice there was no increase of the fructose concentration in blood although the mice were fed with the diet high in fructose. As GLUT 5 is only essential for the transport of fructose, there was no discrepancy observable when mice were fed with a diet high in glucose [4]. So absence of GLUT 5 in mice has no influence on the absorption of glucose, whose transporter mainly is the insulin-regulated glucose transport protein GLUT 4 [7], but fructose malabsorption occurs [4]. Although it has been shown that the expression of GLUT 2 also increases with increased fructose concentration in the intestine of mice or

rats [4], GLUT 2 is not able to compensate the lack of GLUT 5. Compared to GLUT 5, the affinity of GLUT 2 to fructose is only moderate and thus GLUT 2 cannot fully adopt fructose absorption of GLUT 5 [4].

3. Effects of fructose

3.1 General facts

As the exposure to dietary fructose has increased 1.000% during the past 40 years [10], there is growing concern that the consumption of fructose may be a reason for the rising incidence of weight gain and obesity in humans all over the world. As there is a relation between the consumption of sugar-sweetened beverages and the risk of weight gain, the sugar fructose and its metabolism gained considerable attention [1]. Since 1970 the intake of fructose, either alone as a monosaccharide or together with glucose forming the disaccharide sucrose, consisting of 50% fructose and 50% glucose, has increased [1]. However, most of the fructose is consumed in form of high fructose corn syrup (HFCS) [10], whose ratios of glucose and fructose are similar to sucrose [1]. HFCS refers to varying amounts of free fructose and free glucose, but HFCS mainly (60%) consists of 55% fructose and 45% glucose. Referring to the amount of fructose being 55% it is also called HFCS-55 [10,11].

In most of the studies with human subjects, the amount of sugar was higher than the estimate for the average intake of added sugars by Americans, which is around 16% [6].

How fructose affects either animals or humans is subject of this thesis. There is a focus on the mechanisms for fructose-induced weight gain and the metabolic syndrome, whose risk factors contribute to obesity. All the features of the metabolic syndrome are associated with increased risk for cardiovascular disease [12]. Fructose also induces changes in de novo lipogenesis. Type 2 diabetes and fatty liver [12] are other metabolic consequences of the consumption of fructose [11].

Nonalcoholic fatty liver disease (NAFLD), being the most common hepatic disorder of industrialized countries is associated with obesity, insulin resistance, and type 2 diabetes, all features, which are linked to an excessive fructose intake [10].

3.2 Weight gain

High-fructose diets lead to increased body weight and adiposity in animals consuming diets high in fructose, however little information about the relation between fructose and adiposity in humans exists [2].

How fructose affects weight gain in humans has been observed in 3 studies. Although the aim of the studies was not to determine the effects of fructose on obesity, its effects and thus changed energy intake and body weight gained interest.

Both energy intake and body weight were increased in both male and female subjects drinking 1150 g soda sweetened with HFCS for 3 wk. If aspartame, an artificial sweetener, was used instead of HFCS, the increases were negligible [2].

In another study, 14 men of middle age incorporated 50-60g fructose/d into their diets for 24 wk. 11 of these men had type 2 diabetes mellitus, while the other 3 were suffering of type 1 diabetes mellitus. All 14 of them showed increased body weight [2].

The effects of fructose-intake on body weight were also investigated in overweight persons. Those overweight subjects showed increased energy intake, body weight, fat mass and blood pressure after consuming either sucrose ad containing 50% fructose or an artificial sweetener ad libitum for 10 wk [2]. In another study, glucose and fructose consumption providing 25% of daily energy requirement as glucose- or fructose-sweetened beverages resulted in increased body weight. Subjects consuming these beverages exhibited a higher fat mass and waist circumference. Both glucose and fructose led to increased adiposity, but the deposition of the adipose tissue differs depending on whether fructose or glucose are involved [6].

Food intake and energy expenditure is regulated by the 2 hormones insulin and leptin, whose production is not increased by fructose ingestion [2]. As consumptions of diets high in fructose may lead to increased caloric intake or decreased energy expenditure, decreased signalling in the brain of both hormones insulin and leptin could contribute to weight gain [11].

3.2.1 Insulin

The hormone insulin regulates fat and carbohydrate metabolism.

As already mentioned, the secretion of insulin from pancreatic β cells is not stimulated by fructose. A reason for this might be the low concentration of the glucose transport protein GLUT 5, being the main fructose transporter, in β cells.

Insulin plays an important role in the regulation of adiposity as it is one of the key signals concerning energy balance and food intake [2]. This regulation also works via the central nervous system (CNS), where insulin receptors are localized. Food intake in

animals is inhibited by insulin administration into the central nervous system. However, due to fructose ingestion the production of insulin is not increased therefore leading to a reduction in insulin administration into the CNS. This was shown in a study with dogs. A high-fat diet led to a reduced transport of insulin into the CNS of dogs and obesity is induced by the diet. So increased body weight and impairment of insulin transport are inversely related. As insulin is involved in the regulation of adiposity via its actions in the CNS, obesity in mice was observed as the insulin receptor in neurons was knocked out. Weight gain and thus the development of obesity are enhanced if the insulin delivery is reduced, e.g. by fructose [2].

3.2.2 Leptin

Leptin is one of the most important adipose-derived hormones that play a key role in regulating food intake [1]. Signalling through phosphatidylinositol 3-kinase (PI-3K), which is an enzyme also involved in the insulin signal transduction pathway, mediates the effects of leptin inhibiting food intake [13]. Via the insulin signal transduction pathway, actions of both insulin and leptin are mediated in the hypothalamic regulation in eating behavior and metabolic homeostasis [13]. Thus, they are transported as peripheral signals to the central nervous system (CNS) [13], of which insulin and leptin are key endocrine signals concerning energy balance [11]. The consumption of diets high in energy from fructose is leading to a reduction of insulin and leptin signalling in the brain. This is due to the increased caloric intake resulting from fructose consumption and the concomitantly reduced leptin and insulin production [11].

Like insulin, leptin administration reduces food intake. Defects in the leptin receptor or a failure in leptin production lead to obesity. Thus adiposity, resulting from leptin deficiency, is decreased by administration of leptin [2].

Insulin stimulates leptin gene expression and also the regulation of leptin production depends on insulin [2].

Glucose metabolism, which is strongly influenced by insulin signalling, regulates the release of leptin [1]. Due to glucose metabolism, the production of leptin indirectly depends on insulin, which increases glucose utilization and oxidative glucose metabolism in adipocytes [13]. If glucose transport or glycolysis is disturbed, leptin secretion, which is stimulated by insulin, is inhibited [2]. Neither plasma glucose nor the insulin concentration was increased after the uptake of fructose with a meal. The consumption of fructose does not lead to an insulin response or the production of leptin by adipose tissue [11]. Although leptin production in humans is stimulated by high-

carbohydrate meals (e.g. glucose), the effects of fructose are different because it does not stimulate insulin secretion [2].

Due to the assumption that beverages sweetened with fructose would lead to decreased leptin concentrations in comparison to beverages sweetened with glucose, 12 normal-weight young women consumed fructose-sweetened beverages with meals [11]. The leptin concentrations after consumption of fructose were measured over two separate 24h periods. 24-h circulating leptin concentrations were lower after consumption of fructose-sweetened beverages at 30% of energy requirements with three meals. The concentrations of glucose and insulin were also decreased [11]. Due to the decreased leptin concentration, the postprandial suppression of ghrelin, which is an orexigenic gastric hormone [2] and the counterpart of leptin, was less [11]. So fructose consumption leads to higher ghrelin levels [10].

According to another study, postprandial insulin responses to a diet containing fructose and 24-h circulating leptin concentrations were reduced, too. The concentration of leptin decreased by 35% [13].

Thus, the “feeding” of fructose led to a decreased secretion of leptin in human subjects. However, resistance to leptin was demonstrated by diet-induced obese mice as well as by obese humans [1]. Both fructose and dietary fat resulted in a decreased insulin secretion and leptin production, and the postprandial suppression of ghrelin was also lowered. The combined effects of lowered circulating leptin and concomitantly reduced satiety signalling and insulin led to the result that diets high in energy derived from fructose could lead to increased energy intake and thereby contribute to weight gain and obesity [13].

3.3 Metabolic syndrome

Increased fructose consumption is not only related to obesity, but also to the metabolic syndrome (MetS) [11]. However, obesity is associated with the metabolic syndrome. All the features of the MetS such as dyslipidemia, elevated waist circumference, insulin resistance and high blood pressure can thus also be linked to the consumption of the highly lipogenic sugar fructose in humans. And all these features of the MetS are risk factors related to obesity [1]. For the development of obesity and also insulin resistance, lipids play an essential role. Thus, as fructose is highly lipogenic, there is a focus on that characteristic.

The development of dyslipidemia, insulin resistance, or visceral adiposity is not related to the dietary glycemic index as consumption of fructose results in lower postprandial insulin and glucose [14].

3.3.1 Dyslipidemia

Dyslipidemia, which describes an abnormal amount of lipids in the blood, is a significant characteristic of the MetS. Several studies showed that dietary fructose induces hyperlipidemia being the most common form of dyslipidemia. In hyperlipidemia the lipids are elevated, which can be induced by feeding with fructose [2]. Elevated plasma triglycerides concentrations are typical of dyslipidemia [15].
Triacylglycerols, apo B, and also small dense LDL increased after consumption of 25% of energy in form of HFCS and sucrose. These effects are similar to that induced by fructose alone [11].

The effects of fructose consumption on TG levels in blood distinguish depending on the duration of the diet, classifying them in short-term (24 h) and long-term (>2wk) effects. Acute effects induced by fructose consumption differ from those occurring after some time. However, for the determination of the sustained metabolic effects of fructose consumption, long- term studies are indispensable [14]. For example, rats fed with a diet high in fructose showed significant increases of the serum triacylglycerol concentration and these elevated triacylglycerol concentrations are significant for obesity and diabetes [2]. However, the increase in postprandial TG concentrations was almost the same when HFCS or sucrose was consumed instead of 100% fructose alone. This was shown in a short-term study, in which data of fructose, glucose, HFCS, and sucrose were compared [11].
The human liver is rather capable of metabolizing fructose than glucose. Rats were fed with both fructose and glucose and the results were compared [8]. While glucose concentrations led to decreased circulating triacylglycerol concentrations, consumption of the same amount of fructose resulted in elevated triacylglycerol concentrations. When rats were fed with a standard diet, the TG concentrations fell and no hyperlipidemia developed. High-sucrose diets resulted in elevated serum triacylglycerol concentrations, however this is based on fructose, which is a component of sucrose and responsible for the effects of the disaccharide sucrose on the blood TG concentrations [2]. These results are consistent with a long-term study, where women consumed fructose-sweetened beverages at 25% of energy requirements for 10 wk. Results from this study indicated that 24 h TG exposure was increased by 140% in overweight women. As can be seen in *figure 10*, consumption of the same amount of glucose resulted in decreased TG concentrations [11].

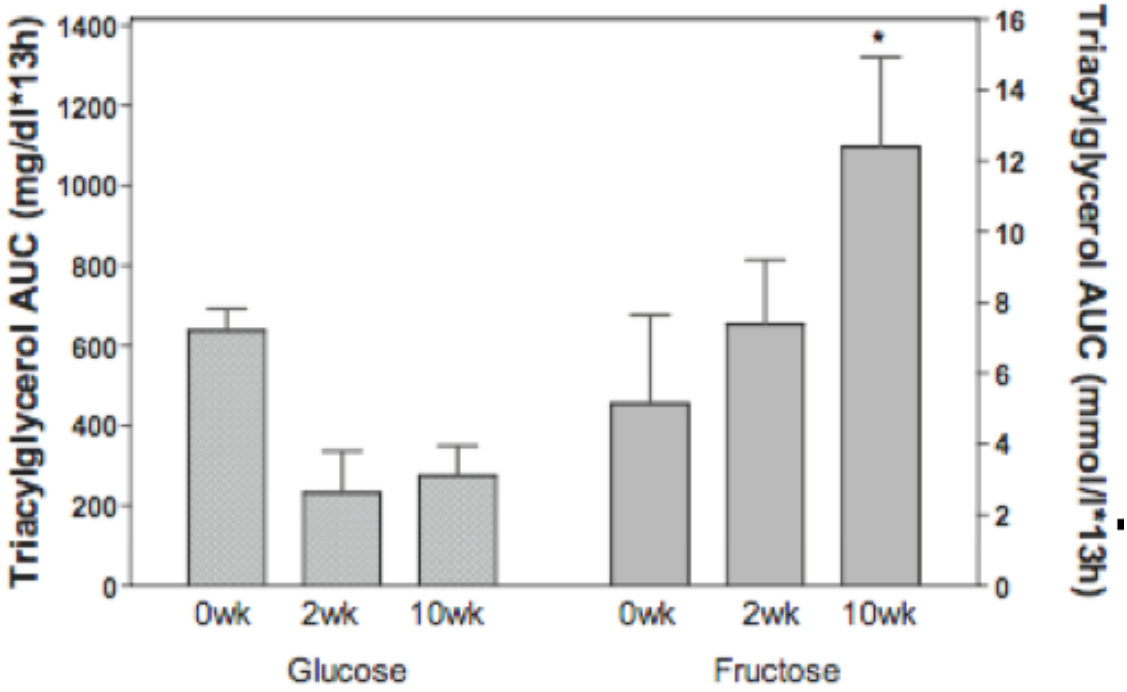

Figure 10. Changes in triacylglycerol concentrations after consumption of glucose and fructose [11]

To investigate the effects of different fructose concentrations, 12 hyperinsulinemic and 12 control subjects, both groups were male, consumed diets containing 0%, 7.5%, and 15% of energy from fructose. The duration of the study was 5 wk. When 7.5% or 15% of energy were consumed as fructose, total plasma cholesterol and LDL cholesterol were increased compared to the consumption of starch in control and hyperinsulinemic subjects. In hyperinsulinemic men, the plasma triacylglycerol concentrations were the higher, the higher the amount of fructose consumed [2].

In another 5 wk lasting study, male subjects being either hyperinsulinemic or non-hyperinsulinemic, consumed diets containing 20% of energy as fructose [2]. This was again compared to diets containing high-amylose cornstarch. In both hyperinsulinemic and non-hyperinsulinemic men, triacylglycerol and cholesterol concentrations were elevated. However, no increase in TG or cholesterol concentration was observed when the subjects consumed the cornstarch diet [2].

But elevated lipids induced by consumption of fructose were not observed in all studies. In the Turku sugar studies, the effects of fructose, xylitol, and sucrose were observed. The study lasted 2 y and the influence of fructose, xylitol, and sucrose on 127 healthy subjects was investigated. In this study, neither TG concentrations nor plasma cholesterol were manipulated when fructose or xylitol were substituted for sucrose, which is composed of 50% fructose. Chronic consumption of fructose and sucrose neither induced increased lipids nor were changes in body weight observed [2]. In a 10 weeks lasting study, subjects consumed both glucose and fructose

sweetened beverages providing 25% of energy requirements [6]. Results from this study report that while plasma lipid and lipoprotein concentrations increased during fructose consumption, no increase in fasting TG concentrations was detectable. But fasting TG concentrations were elevated after consumption of glucose. Fructose or glucose consumption on postprandial TG concentrations delivered opposite results. Fructose consumption resulted in markedly increased TG exposure, while TG concentrations remained unchanged during glucose consumption [6].

However, according to a study about the effects of dietary fructose on lipid metabolism, the mechanism of how triacylglycerol concentrations are influenced by dietary fructose are not well defined [2]. But the effects of fructose, consumed at about 20% of total energy, on total and LDL cholesterol concentrations are strongly evident [2].

In a 6 wk study, both fasting and postprandial TG concentrations were measured [2]. The effects of crystalline fructose providing 17% of energy were observed. The effects differed dependent on the age and sex of the subjects. While men older than 40 y showed increases in fasting, postprandial, and daylong plasma TG concentrations after fructose consumption, none of these effects was observable in men younger than 40 y and older women above an age of 40 y. In this study, no significant changes in plasma cholesterol, HDL cholesterol or LDL cholesterol were recognisable [2].

In a study with men and women consuming a diet providing 17% energy as fructose and glucose for 6 wk, differences dependent on the sex were observable [14]. While men showed increased 24 h TG postprandial profiles after fructose consumption, the effects after fructose and glucose consumption in women did not differ [14]. In women, body fat, age, and menopausal status are all characteristics influencing the responses of postprandial TG [6].

In another study, 12 young women with normal weight and without hypertriacylglycerolemia consumed fructose- and glucose-sweetened beverages [2]. The diets contained 30% of total energy from the sugars and were consumed with 3 meals over 24h. The consumption of fructose-sweetened beverages resulted in elevated TG concentrations and the peaks were higher. While fructose led to an increase in TG concentrations, TG concentrations declined after consuming glucose. In subjects with insulin resistance or hypertriacylglycerolemia, the increase of postprandial triacylglycerol concentrations may be even higher due to consumption of fructose [2].

A reason for the elevated TG concentrations caused by consumption of dietary fructose may be the increased TG synthesis. If both, VLDL particle size and secretion rate from the liver, are increased, increased TG synthesis follows [2].

As was shown in almost all studies, TG concentrations were increased after consumption of dietary fructose in both healthy and hyperinsulinemic men and patients with hypertriglyceridemia and type 2 diabetes [14].
But there were also reports that long-term consumption of fructose at 20-25% of energy did not increase fasting TG concentrations in neither older overweight women, nor healthy male and female subjects, nor hyperinsulinemic female subjects, nor male and female patients with type 2 diabetes, nor in men with hypertriglyceridemia [14]. These differences in the results may indicate that fasting TG concentrations may not be the inducer of dyslipidemia caused by fructose consumption [14].
However, long-term effects on circulating triacylglycerol concentrations and also HDL and LDL cholesterol after a diet high in fructose need to be clarified more precisely [2].

As for TG concentrations, the acute effects of fructose on apoB concentrations differ from those occurring after sustained exposure, too. There is a distinction between short-term and long-term studies [14].
Fructose increases the amount of total LDL cholesterol particles, however also when no increases in LDL cholesterol were present, apoB concentrations were elevated [11]. If hepatic lipids are increased, the degradation of apoB, which is necessary for the intracellular assembly of triglyceride into VLDL, is lowered [8]. While increases were observable after fructose consumption, glucose consumption did not induce any changes in fasting and postprandial apoB and total and LDL cholesterol [6].
Significant increases of both fasting and postprandial apoB concentrations, but also increases of fasting small dense LDL (sdLDL) and postprandial remnant lipoprotein concentrations were observed in a long-term study with both men and women consuming fructose [14]. The influence of preexisting metabolic syndrome risk factors (MSRF) was the highest considering the lipid parameter sdLDL, because in subjects exhibiting 3 MSRF the increases in sdLDL was 2-fold greater than in those with just 0 to 2 MSRF [6]. Fructose led to increases in both fasting and postprandial sdLDL concentrations. In subjects with MetS the concentrations of small dense LDL were doubled compared to subjects without the MetS [1].
No changes in fasting oxidized LDL concentrations were induced by glucose [6], but fructose consumption resulted in elevated concentrations of oxidized LDL, whose oxidation is easier for small dense LDL particles than for larger ones [1].
In short-term (24 h) studies, postprandial apoB concentrations decreased after consumption of fructose-sweetened beverages with 3 meals. This shows that the effects of fructose are different in short and in long term [14].

3.3.2 Insulin resistance

Insulin resistance, one of the main features of the MetS is induced by diets high in fructose [12].

Decreased insulin sensitivity associated with impaired hepatic insulin action was observed in a study with rats. This study lasted 4 wk and rats were fed a diet containing 35% of energy as fructose [2]. In a study with rats fed with fructose diets for 2 wk, adverse changes in glucose metabolism were observable. However, no effects or changes occurred after feeding the rats with a diet containing a comparable amount of starch. A study in hamsters showed similar results. Hamsters were fed with a high-fructose or a high-sucrose diet containing only 50% fructose. Glucose was administered intravenously into the hamsters and the reduction of the rate of glucose disappearance after injection was different in fructose-fed and sucrose-fed hamsters. More precisely, the consumption of fructose resulted in a much greater decrease of glucose disappearance [2].

In the long term, feeding of fructose induces insulin resistance and obesity in experimental animals, which thus induces hyperinsulinemia, however no stimulation of insulin secretion by fructose is observable in short term [2]. Dietary fructose showed to have effects on fasting serum insulin and fasting serum glucose, which were increased in rats after feeding 15% of energy as fructose. The study lasted 15 m, and the rats that consumed fructose were compared with rats that consumed cornstarch [2]. The food intake and body weight in both groups (fructose fed and cornstarch fed rats) did not differ [2]. The same results were achieved in a study with mice, which were fed with HFCS-55. With HFCS-55 enriched mice showed increased fasting serum insulin levels, which is a marker of insulin resistance [10].

In a 10 wk lasting study with humans, the participants consuming fructose providing 25% of energy requirements showed elevated fasting glucose and insulin concentrations [14] Moreover, consumption of fructose resulted in decreased insulin sensitivity and glucose tolerance, while glucose did not have any effect on neither insulin sensitivity nor glucose tolerance [6]. Both, men and women responded to the fructose consumption with decreased insulin sensitivity, however, in women insulin sensitivity was more decreased due to fructose consumption than in men [6]. While the changes of insulin sensitivity are greater in women, adverse results are given considering visceral adiposity, which was greater in men due to consumption of fructose. This suggests that visceral adiposity and FFA levels do not contribute to the decreased insulin sensitivity induced by fructose [6]. It has been demonstrated that in healthy objects the consumption of 1.000 extra kcal/d fructose along with ad libitum diet resulted in decreased insulin sensitivity. In contrast to fructose, 1.000 extra kcal/d

glucose did not induce any change in insulin sensitivity. It remains to ascertain if fructose consumption decreases insulin sensitivity to the same degree in absence of fat gain, because the subjects of the 10 wk lasting study, in which 25% of the energy was provided as fructose-sweetened beverages, gained body weight and fat. However, there was no correlation between the changes of body weight and the change of insulin sensitivity [6].

It has also been reported that in a 4 wk lasting study with 7 young normal-weight men increased fasting glucose levels were measured as fructose at 18% of the required energy was substituted by a moderate-fat diet [14]. While glucose levels changed, other changes indicating insulin resistance were not observable. These adverse results could be due to the different amounts of provided energy requirement or the duration of the study [14].

High fructose feeding for 7d induced decreased insulin sensitivity and insulin binding, but no similar changes were observed with diets high in glucose [2].

There is a relation between insulin resistance, increased fasting plasma insulin concentrations, glucose intolerance, and nonesterified fatty acid concentrations. Increased adiposity e.g. induced by fructose consumption might lead to elevated nonesterified fatty acid concentrations in the liver [2]. This metabolic consequence results from decreased insulin secretion and reduced leptin production, which are both related to fructose consumption and thus increased body weight and obesity [2].

As nonesterified fatty acid concentrations are increased in the liver by consumption of fructose, production of VLDL triacylglycerol is higher. Therefore fructose consumption may induce hypertriacylglycerolemia [2]. Decreased insulin sensitivity and also reduced postprandial exposure to insulin are related to postprandial lipoprotein lipase (LPL) activity. In subjects consuming fructose, postprandial LPL activity was reduced, which contributes to hypertriglyceridemia via a lowered clearance of TG [1]. So reduced clearance of TG may add to elevated postprandial TG [6]. Whereas fructose consumption affects postprandial postheparin lipoprotein lipase (LPL) activity, no fructose-induced changes appear considering fasting postheparin LPL activity [6].

The relation between hypertriacylglycerolemia and insulin resistance was observed in glycerolemic persons, which exhibited insulin resistance and reduced insulin binding. Hence, fructose diets may promote insulin resistance via increased amounts of triacylglycerol [2]. Fructose consumption is correlated with increased adiposity and insulin resistance in adipose tissue. Those features of the MetS promote an increase in both circulating and portal levels of FFA. As the hepatic FFA uptake is elevated, hepatic insulin resistance is promoted [1].

Humans consuming fructose showed no significant change in the concentrations of circulating FFA [1], while glucose led to increased FFA exposure [6]. In another study, no changes in FFA concentrations were observable. Glucose led to increased 24-h circulating FFA concentrations, while no changes occurred in subjects consuming fructose [14]. So increases in FFA levels and also visceral adiposity *(chapter 3.3.3)* have no influence on the hepatic insulin resistance following a lipid overload in the liver induced by fructose [14]. Insulin resistance might be promoted by fructose, but not due to the effect of fructose on systemic FFA, but due to de novo lipogenesis (DNL) [1].

As fructose feeding results in a positive energy balance, this may also contribute to decreased insulin sensitivity. When the production of TG exceeds the oxidation of free fatty acids (FFA) and VLDL production and secretion, TG accumulates in the liver being a mediator of hepatic insulin resistance [14]. This exceedance of the liver's capacity is due to the sufficient production of TG via DNL induced by fructose. This assumption is based on the elevated TG and postprandial apo B concentrations induced by fructose consumption [14]. While subjects showed increases of liver TG content and insulin resistance when a positive energy balance was present, the production of TG was not high enough to exceed FFA oxidation and VLDL production or secretion in an energy-balanced diet, although DNL may have increased.

Compared to fructose, glucose did not lead to increased DNL, thus not enough TG was produced in order to exceed FFA oxidation or VLDL production and secretion. Therefore, there is probably the need of both positive energy balance and increases of DNL in order to promote insulin resistance via increased liver TG content. However, the relation between fructose-induced DNL, increased liver TG content and insulin resistance remain to be further investigated [14].

Via fructose-induced DNL increased hepatic lipid content resulted in hepatic insulin resistance, which might be caused by elevated diacylglycerol levels. Insulin resistance induced by lipids is related to increases in diacylglycerol and also novel protein kinase C (PKC) activity, of which diacylglycerol is an activator [6].

Further, the influences of dietary fructose on circulating adiponectin concentrations are under investigation [2]. Mice fed with HFCS-55 diet showed reduced adiponectin levels [4]. Adiponectin is an adipocyte hormone and involved in the regulation of lipid and carbohydrate metabolism. Insulin action in animals is enhanced by administration of adiponectin and insulin sensitivity is related to circulating adiponectin. Insulin resistance is reduced by administration of adiponectin [13]. However, the decreased production of adiponectin leads to insulin resistance associated with obesity [2].

Fructose feeding induces insulin resistance being a main feature of the MetS, which is related to obesity, and the induction does not depend on the underlying mechanism [2].

3.3.3 Visceral adiposity

As fructose consumption is related to the metabolic syndrome, elevated waist circumference is one feature of it. Glucose consumption did not induce any significant changes in total and visceral adipose tissue (VAT) volume [6]. However, while consumption of glucose leads to a lipid deposition in subcutaneous adipose tissue (SAT), fructose consumption favours lipid deposition in both abdominal fat [6] and VAT, especially in men [1]. Increases in intraabdominal fat were larger in men than in women. In general, men showed a greater increase in fat mass than women [6]. *Figure 11* shows the changes in total abdominal adipose tissue, subcutaneous adipose tissue and visceral adipose tissue volume in subjects consuming either glucose or fructose. These subjects consumed glucose- or fructose-sweetened beverages for 10 weeks [6].

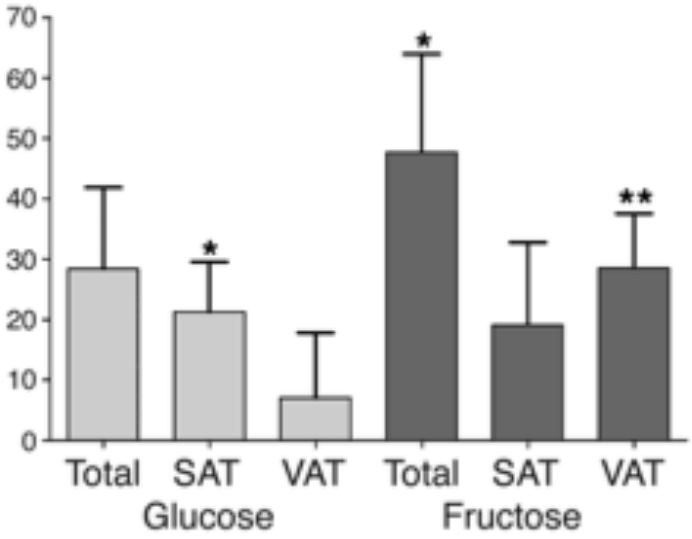

Figure 11. Lipid deposition after consumption of glucose and fructose [6]

A reason for the increased fat deposition in subcutaneous adipose tissue after glucose consumption might be the fact that SAT is more sensitive to the effects of insulin in activating LPL. Visceral adipose tissue, which is increased in subjects consuming fructose, is not as much influenced as SAT by the effects of insulin in activating LPL. The differential LPL responses to fructose and glucose may affect the differential lipid deposition [6].

To study the metabolic effects of fructose and also glucose, men and women with a BMI between 25 and 35, being overweight to obese, consumed beverages sweetened with the sugars fructose or glucose. These sugars provided 25% of the needed energy. Due to the consumption of either fructose or glucose sweetened beverages, the subjects gained around 1.4 kg of weight and fat mass increased about 0.8 kg [12]

during an 8wk lasting period. Thus, the subjects were in positive energy balance. As mentioned above, the consumption of glucose resulted in increased extra-abdominal fat area, while consumption of fructose had an influence on the intra-abdominal fat area. No lipid deposition in visceral adipose tissue was detectable after glucose consumption [14]. While the expression of the lipogenic genes stearoyl-CoA desaturase-1, fatty acid desaturase 1, and fatty acid desaturase 2 were elevated in subcutaneous gluteal fat after glucose consumption, no changes in the expression of those genes occurred after consumption of fructose [6,14]. The reasons for the distinguishing lipid deposition of fructose and glucose are unclear, but their different effects on postprandial exposure to TG and lipoproteins could be one explanation. Moreover, the effects of fructose and glucose on insulin sensitivity are different, thus providing a justification for the distinct regional adipose deposition [1]. Visceral adipose tissue, whose development is enhanced by ingestion of fructose, is the mesenteric adipose tissue. The rate of lipolysis, i.e. the breakdown of lipids, is high in mesenteric adipose tissue and mesenteric fat does not exhibit a strong response to the inhibition of lipolysis by insulin. In obese diabetic subjects, lipolysis did not work properly and the expression of lipolytic genes was restrained [1]. Mesenteric adipose tissue is anatomically located such that it takes part in the portal drainage of the gastrointestinal system. FFA would be directly delivered to the liver, if disturbances occurred in this adipose tissue. For example, increased lipolysis of endogenous lipid stores could be initiated by intake of a high amount of calories. This is described by the portal theory of insulin resistance [1].

Compared to SAT, there is an association between visceral adipose tissue, whose formation is promoted by consumption of fructose, and metabolic disease such as type 2 diabetes and cardiovascular disease [12].

The consumptions of diets high in fructose mainly promote visceral adiposity [1], which is important in the pathogenesis of the metabolic syndrome [14].

3.3.4 Hypertension

Like dyslipidemia, visceral adiposity, and insulin resistance, hypertension is a common feature of the metabolic syndrome and therefore related to obesity. It has been observed in many studies that hypertension is induced in animals which were fed with a diet high in fructose. Rats fed with fructose are often used as a model to observe and determine the effects of pharmacological agents used for the treatment of hypertension [2]. As described in *chapter 2.5,* wild-type mice (GLUT $5^{+/+}$) show after consumption of fructose an upregulated GLUT 5 expression, which is essential for the transport and uptake of fructose. These mice have increased capacity to take up fructose.

Interestingly, after fructose consumption, the blood pressure in GLUT $5^{+/+}$ was increased. While mice expressing GLUT 5 showed systemic hypertension after feeding fructose, mice lacking the GLUT 5 gene (GLUT $5^{-/-}$) did not exhibit hypertension, but a decreased blood pressure. So hypotension was developed in mice lacking the GLUT 5 gene when fed a high fructose diet. Hypertension is induced in mice after fructose consumption, however these results indicate that GLUT 5, being essential in fructose transport and uptake, also plays an important role in hypertension induced by fructose [4]. In human subjects, which consumed fructose-sweetened beverages no changes of blood pressure were observable during 10 weeks [6].

3.4 Lipogenesis

Comparing the changes in liver and adipose tissue, fructose feeding decreases fatty acid synthesis in adipose tissue, whereas it is increased in the liver. So lipid synthesis is shifted from adipose tissue to the liver under influence of fructose [8].

3.4.1 Lipogenesis – adipose tissue

In rats, fructose influences lipogenesis, which mainly takes place in adipose tissue. As mentioned before, adipose tissue lacks the enzyme fructokinase, instead hexokinase is used for the phosphorylation of fructose [8]. There are two isoenzymes of hexokinase present in adipose tissue of rats. However, the most abundant enzyme phosphorylating fructose in adipose tissue is type II hexokinase. This enzyme disappears on fasting, but appears again with fructose feeding.

The conversion of either glucose or fructose to fatty acids by adipose tissue was alleviated in rats which were fed with fructose. When sucrose was fed, lipogenesis from both fructose and glucose was depressed. Lipogenesis from fructose was minor and slower than that from glucose and while feeding of fructose alone resulted in a significantly depressed lipogenesis, depression of lipogenesis from glucose was not significant. Dietary fructose had an influence on lipid synthesis related enzymes as well. It depressed the activities of malic enzyme and citrate cleavage enzyme. In contrast, sucrose feeding just led to somewhat lower activities of malic enzyme and citrate cleavage enzyme, but instead the activity of 6-phosphogluconate dehydrogenase was depressed. Both feeding of sucrose and fructose resulted in depressed activity of adipose tissue hexokinase II, while glucose feeding did not elicit significant changes. All that changes induced by fructose feeding are indicators for the reduced capacity of the adipose tissue synthesizing fatty acids. As hexokinase II

phosphorylates fructose and also glucose, this enzyme might be responsible for the reduction in fatty acid synthesis induced by fructose feeding [8].

3.4.2 Hepatic lipogenesis

Fructose increases hepatic fatty acid synthesis, which is part of the lipogenesis. As the enzyme fructokinase is present in the liver, it phosphorylates fructose there, while hexokinase phosphorylates fructose in adipose tissue due to the lack of the enzyme fructokinase in adipose tissue. Both feeding of dietary fructose and sucrose resulted in an enhanced activity of hepatic fructokinase [8].

Fructose feeding affects hepatic lipogenesis, so the liver adapts to dietary fructose. Glucose did not induce any change in neither fasting nor postprandial hepatic DNL, while fructose did. Fructose feeding resulted in significantly increased postprandial DNL [6]. While dietary fructose did not have any effects on lipogenesis from glucose, lipogenesis from fructose was enhanced. Rats fed with fructose exhibited a more rapid fatty acid synthesis from fructose than those fed with glucose. In contrast to adipose tissue, the malic and citrate cleavage enzyme showed increased activity, being related to the rate of fatty acid synthesis in liver, after feeding with fructose [8].

Increased hepatic de novo lipogenesis follows ingestion of fructose. As fructose metabolism takes place in the liver, this might be one reason to promote lipogenesis. The unregulated uptake of fructose in the liver promotes increased DNL due to the increased production of the lipogenic substrates glycerol-3-phosphate and acetyl-CoA [12].

Moreover, fructose promotes lipogenesis due to the activation of genes like fatty acid synthase and acetyl CoA carboxylase being involved in de novo lipogenesis. The activation is based on the upregulation of the sterol receptor element binding protein-1c (SREBP-1c), which modulates lipid homeostasis [1]. The upregulation of SREBP-1c induced by fructose does not depend on insulin [11]. Moreover, fructose ingestion increases the activity of both carbohydrate regulatory element-binding protein (ChREBP) and nuclear SREBP-1 [1]. Fatty acid synthase (FAS), acyl coenzyme-A carboxylase (ACC) and stearoyl coenzyme-A desaturase-1 are all lipogenic genes, whose expression is increased by fructose feeding [1]. But according to another study [14], fructose feeding did not induce any increases in the expression of stearoyl coenzyme-A desaturase-1.

Due to increased hepatic de novo lipogenesis fatty acids are generated. These fatty acids can be incorporated into hepatic lipids, e.g. hepatic triacylglyceride [6]. Elevated hepatic lipid levels are related to increased VLDL synthesis and secretion and with reduced apoB degradation. Hepatic lipids are elevated by fructose-induced de novo

lipogenesis [1], so increased DNL contributes to TG being elevated due to the consumption of fructose [6]. Fructose-induced DNL, increased visceral adiposity [1], and a positive energy balance [6] are contributed to lipid overloading thus resulting in impaired insulin sensitivity and hyperlipidemia, which are both characteristic for the MetS [1].

3.5 Mechanisms underlying the effects of fructose

The following figure shows the mechanisms underlying the effects of fructose consumption and reviews the most important facts concerning the effects of fructose on lipids and lipid metabolism mentioned before in chapters 3.3.1, 3.3.2, and 3.4.

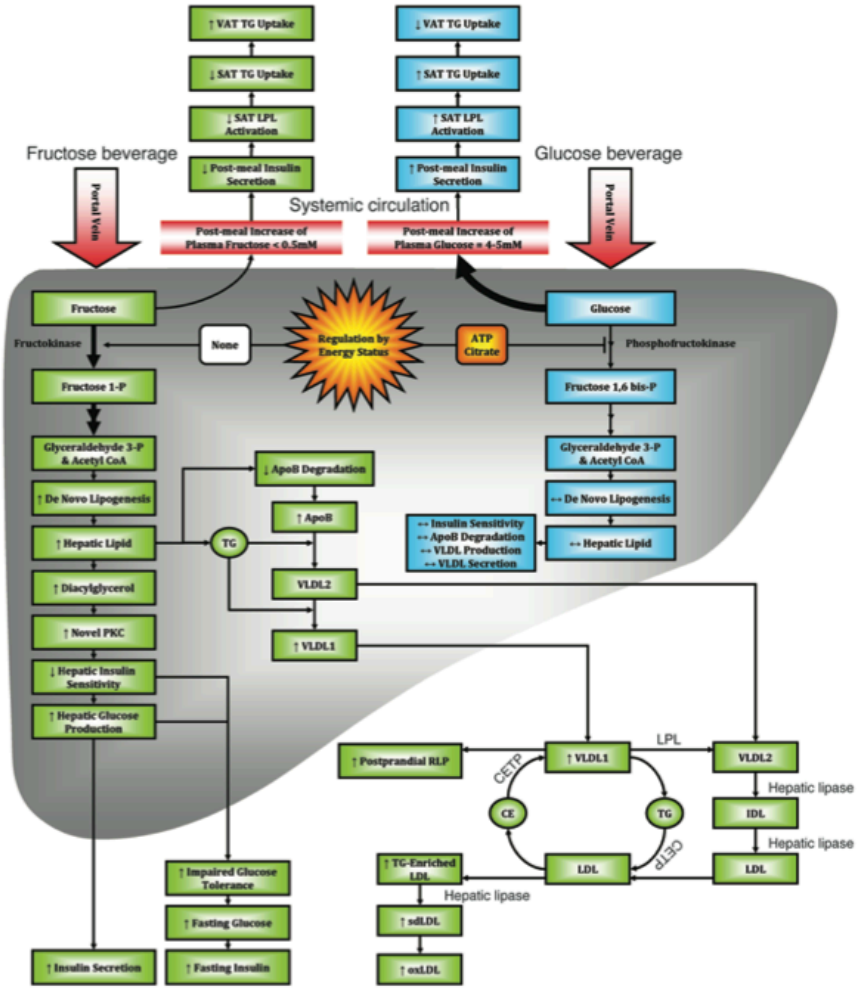

Figure 12. Mechanisms underlying the effects of fructose [6]

3.6 Link to PTP-1B

Protein tyrosine phosphatase-1B (PTP-1B) is an enzyme affecting insulin sensitivity and diet-induced weight gain [15]. It is involved in the downregulation of insulin receptor substrate-1 (IRS-1) and phosphatidylinositol 3-kinase (PI-3K), which

participates in the insulin signal transduction pathway and is stimulated by insulin. It has been reported that in insulin-resistant diabetes in humans tissue levels of PTP-1B were elevated. In rats the expression of PTP-1B was decreased when streptozotocin feeding, an inducer for experimental diabetes in animals, led to type 1 diabetes in rats.
Weight gain is also correlated to the adipose hormone leptin. PTP-1B-deficient mice show enhanced leptin-mediated loss of body weight.
As PTP-1B affects the insulin sensitivity, insulin resistance is promoted by increased PTP-1B expression. This was shown in hamsters, where increased PTP-1B mass and activity were associated with fructose-induced insulin resistance. In a study considering the relation between PTP-1B and fructose feeding, PTP-1B wild-type and knock-out mice were compared. Mice lacking PTP-1B are resistant to obesity and show enhanced insulin sensitivity. To further examine the part of PTP-1B, mice consumed a diet high in fructose for 3 weeks. In wild-type mice increases in VLDL-triglyceride and both LDL and HDL cholesterol were induced by the fructose-enriched diet, while in mice lacking PTP-1B no significant changes were observable upon fructose feeding. The same result was achieved considering total plasma triglyceride. Fructose induced an increase of TG in wild-type mice, but there was no increase in the knock-out mice. Moreover, PTP-1B knock-out mice exhibited a reduction of apoB-containing lipoproteins, associated with reduced hepatic apoB secretion from isolated hepatocytes, and reduction of plasma triglyceride. All these observations lead to the suggestion that the diabetogenic and lipogenic effects of fructose may be partly mediated by PTP-1B, whose reduced expression may protect against dyslipidemia. As insulin regulates lipid and lipoprotein metabolism, it may contribute to the influence PTP-1B has on lipid and lipoprotein metabolism and thus may take part in the development of dyslipidemia [15].

4. Conclusion

The sugar fructose and its effects are outlined in this thesis. Fructose consumption has increased during the last few decades and this increase coincides with increases in the prevalence of weight gain and obesity [1].

The uptake and transport of fructose deviates from that of glucose, which is sensitive to insulin [5]. The glucose transport protein being the most likely fructose transporter is GLUT 5. GLUT 5 is neither stimulated by insulin nor by cytochalasin B making it ideal for the transport of fructose. A large part of fructose is transported in an insulin-insensitive manner, while the residual part of fructose transport depends on insulin. Thus there is also a relation to the transport of glucose [7].

The uptake of fructose is associated with the expression of GLUT 5, which was shown in a study with mice. The expression of GLUT 5, which increases due to fructose ingestion, facilitates the uptake of fructose. The importance of GLUT 5 in the absorption of fructose has been demonstrated as mice lacking the GLUT 5 gene lost weight when being fed with a diet high in fructose [4].

As the concentration of GLUT 5 is low in pancreatic β cells, the secretion of insulin, which is an important hormone in the regulation of adiposity, is not stimulated by fructose. Fructose induces reduced insulin delivery to the CNS [2]. Like insulin, leptin is a key signal concerning energy balance and leptin signalling is decreased by fructose uptake, too [11]. Thus both hormones insulin and leptin promote weight gain and obesity as their concentrations are decreased following consumption of fructose [13].

Fructose consumption is associated with the metabolic syndrome, whose main features are dyslipidemia, insulin resistance, elevated waist circumference, and high blood pressure, all of which are related to obesity [1]. As fructose is a highly lipogenic sugar, there is a focus on lipids, whose amount is altered by ingestion of fructose [2]. Elevated plasma triglyceride concentrations are characteristic of dyslipidemia [15] and the influence on it was demonstrated in several studies. While in most of the studies with humans fructose feeding resulted in increased plasma TG concentrations thus leading to obesity, there were also studies disproving these results. Fructose consumption did not lead to an increase of fasting TG concentrations [6], however postprandial TG were elevated [11], though it is important to consider different facts such as age, sex, health status of subjects and the amount of energy requirements provided by fructose. The effects of fructose in short-term (24h) deviate from those in long-term [14]. ApoB concentrations decreased after consumption of fructose, which was demonstrated in a 24h study, while elevated apoB and also increased sdLDL were observed in a long-term study [14]. Fructose-induced de novo lipogenesis is associated

with elevated hepatic lipid levels such as elevated TG [6], increased VLDL synthesis and reduced apoB degradation [1].

Fructose consumption is correlated with increased adiposity and insulin resistance in animals and humans. Fasting serum insulin and fasting serum glucose, both markers of insulin resistance, are affected and elevated by fructose consumption. Insulin sensitivity, which was decreased by a diet high in fructose, is correlated with a positive energy balance and excess body weight [2]. The consumption of fructose leads to weight gain, however, mainly the development of intra-abdominal fat is enhanced and lipid deposition in visceral adipose tissue is favoured [6].

5. References

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