CHARLES UNIVERSITY IN PRAGUE
THIRD FACULTY OF MEDICINE

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Adipokines (leptin and adiponectin) as predictors of weight loss and weight regain during one year period following the 10-week low caloric diet (NUGENOB study)

Vliv adipocytárních hormonů (leptin a adiponectin)
na predikci váhového úbytku a následného přírůstku (weight regain)
v průběhu jednoho roku po ukončení 10-týdenní nízkokalorické diety
(studie NUGENOB)

Diploma thesis

Prague, May 2010

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Master's programme of study

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Medicine, 3LF

Date and year of defence: 07.06.2010

Written Declaration

I declare that I completed the submitted work individually and only used the mentioned sources and literature. Concurrently, I give my permission for this diploma thesis to be used for study purposes.

Jan Drahoš

In Prague on May 26th, 2010

Acknowledgements

I would like to thank to doctor Jan Polák, MD PhD for making me involved in his work at the department of Sport Medicine and for many useful tips regarding this thesis.

I'd also like to thank all the other members of the team at the Department of Sport Medicine for all the support they have shown to me.

I'd like to thank Eva Němcová who worked on the same project as me and help me a lot.

Contents

Introduction

For many years I was thinking of becoming an internist and I among all the internal subspecialties I was interested in diabetes, metabolism and nutrition. That made me some years ago to start volunteering in the laboratory of Department of Sport medicine at The 3rd faculty of Medicine of Charles University, as this department focuses mainly on the topic of overweight. So I've decided to use this colaboration and set myself a project in this laboratoy that would be a basis for my thesis. I was approved to participate on a follow up of a multicentric study called Nugenob, sponsored by the European Union which could serve as a sign that it's topic is generally recognized as important. This study was focusing on the changes in gene expression under the influence of different hypocaloric meals. But except the data gathered for genetic analysis, there was a lot of other important information about the metabolic state of the subjects. The database that was created could be a source for many different analysis out of which I've been approved to perform one. The goal of my thesis is to have a closer look on the effect of adipose tissue hormons, leptin and adiponectin, and asses their part in weight loss and subsequent weight maintenance. That could possibly tell the doctor right at the beginning of weigh-loosing counselling, whether the patient is going to be successful using the basic treatment or whether he or she needs to get some extra care in order to achieve satisfactory results.

1. Obesity

1.1. Introduction

I'm starting this thesis with an introduction to the obesity issue, including some information about the broadly discussed topic of it's treatment, i.e. weight loss but also a bit less considered but at least as important problem of weight maintenance.

Storing fat was evolutionary useful but in the modern society it leads to obesity and deteriorates our health.

1.2. Definition

Obesity is a stage of increased body fat percentage. It's most widely evaluated by BMI in kg/m2. Because the prognoses start to deteriorate with BMI of 25, this is considered a beginning of overweight and has clear health consequences correlate with BMI over 30, people who meat this criterion are classified as obese.

1.3. Prevalence

Among the US population, 30% are obese and another 35% are overweight. The prevalence of obesity in the US is higher among women and poor. Also in the Czech republic, the prevalence of obesity is higher in women than men, but the prevalence of overweight is higher in men than in women.

Figure 1 – Prevalence of obesity and overweight in the Czech republic

Prevalence of obesity and overweight in the Czech republic

men   **women**

48,8   22,6   28,6

30,9   25,6   43,5

normal weight (BMI 18,5-25)

overweight (BMI 25-30)

obesity (BMI >30)

n = 1174   n = 1275
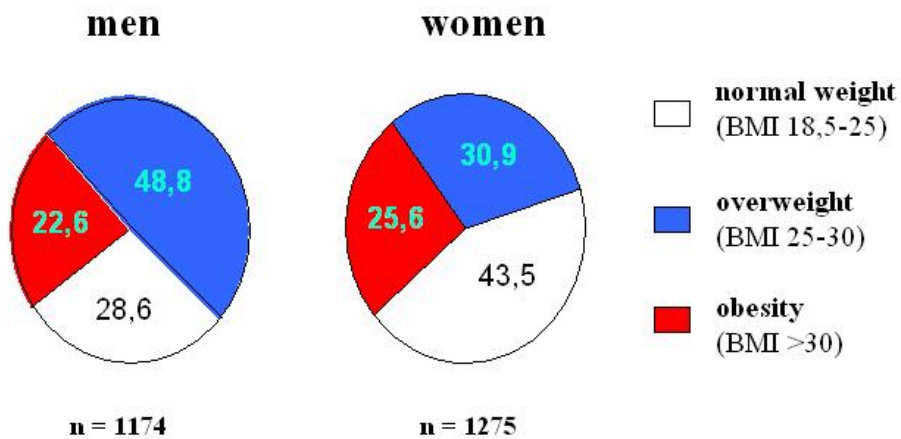
Hainer V. et al., Progress in obesity research, 1999 according to Czech Monica 1997

1.4. Etiology

Sometimes obesity can be secondary to some other underlying disease. It could be seen in Cushing syndrome, hypotyroidism, insulinoma or damage to hypophysis such as in craniopharingeoma, postraumatic or an immune system mediated damage.

But most often, obesity is caused by a primary failure of body weight regulation, which will be discribed in the second chapter.

1.5. Complications

Obesity comes with many adverse effects on health.

Figure 2 – High BMI means a higher risk of diseases

High BMI means
a higher risk of diseases

Relative risk of death

Cardiovascular diseases
Cancer
All other causes of death

BMI

Calle EE et al. N Engl J Med 1999; 341: 1097-195
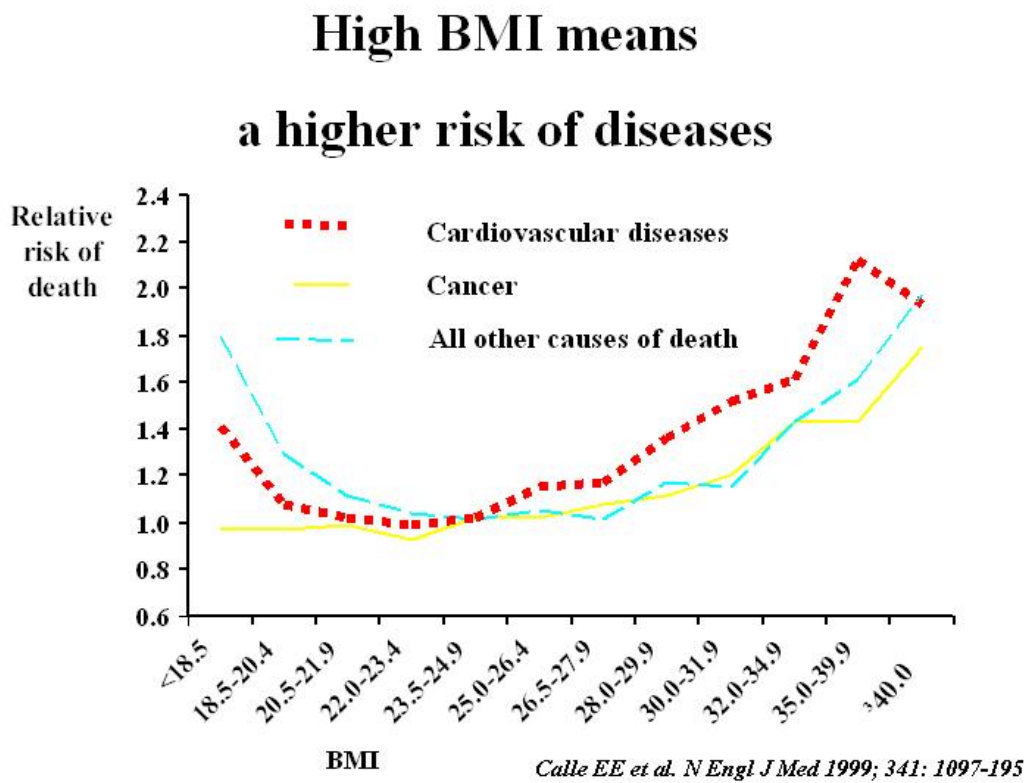
First of all it's the insulin resistance and the risk of diabetes mellitus type 2 in cases where insulin resistance comes together with impaired insulin production. Among other contributing factors, lower levels of adiponectin could play a role in impaired insulin sensitivity.

Figure 3 – Obesity is a risk factor for T2DM

Obesity is a risk factor for T2DM

BMI (kg/m^2)
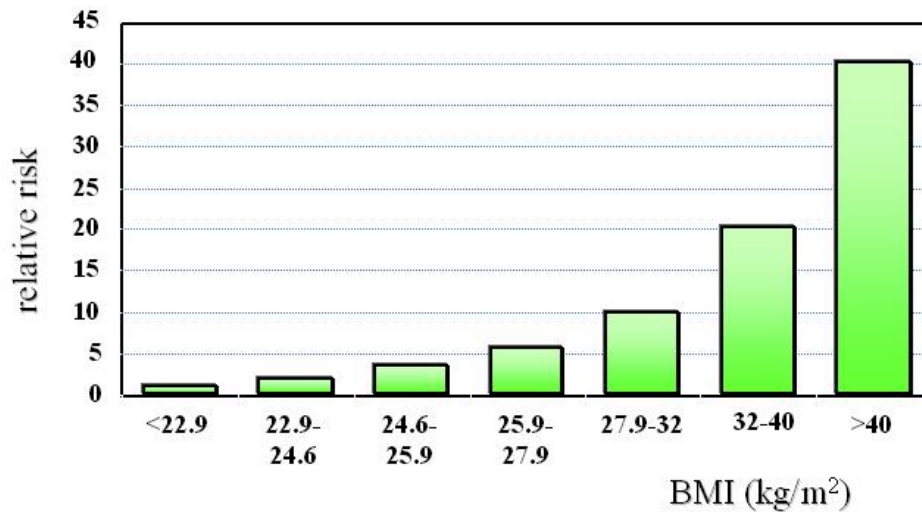
Chan et al, Diabetes Care, 1994

Obesity is associated with polycystic ovarian syndrom in women and some less severe impairments of sexual functions in men. Obesity is a major risk factor for cardiovascular diseases, such as high blood pressure, coronary disease and congestive heart failure.

Figure 4 – Obesity is related with higher incidence of hypertension
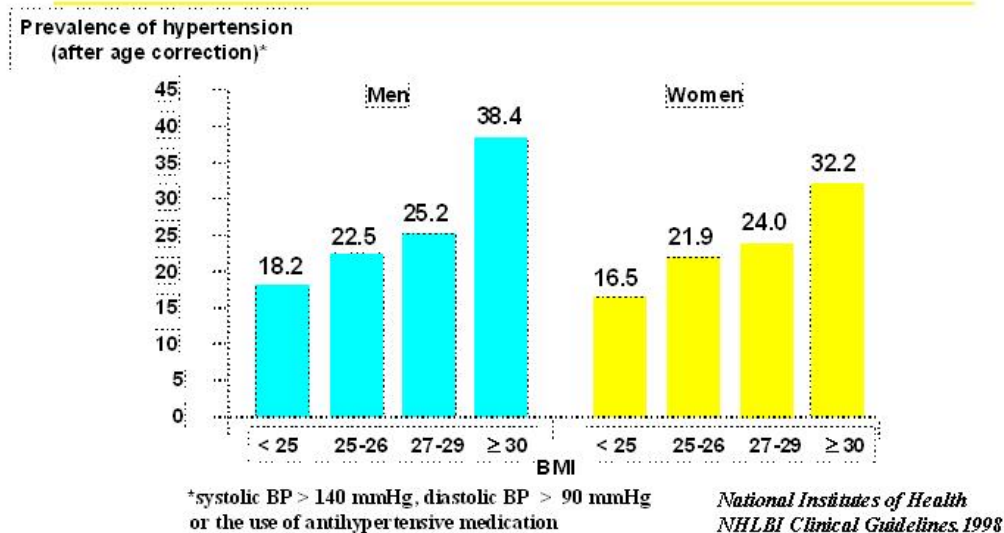
Furthermore the negative influence on respiratory function have been proven. Gallstones are more frequent in obese individuals and also some types of cancer appear more often. Not surprisingly, the joint suffer under an excessive body weight and thus obesity is a risk factor for degenerative diseases of joints.

1.6. Treatment – weight reduction

The treatment of obesity should suit each individual, thus it's neccessary to gather data about each patient. First we assess patients obesity related history. Then we perform a physical examination among other to gain anthropometrical data. It is not sufficient to evaluate obesity only by BMI (see above). Since intraabdominal and abdominal subcutaneous fat is more risky, we try to evaluate it's percentage from total body fat. That's most widely done by waist circumference (measure above iliac crest and considered normal in European population if lower then 94 in men and 80 in women) or waist/ hip ratio (WHR) which is

abnormal if higher than 0,9 in women and 1,0 in men. We also need to check for signs that could indicate that obesity is secondary to another disease), consider comorbid conditions and evaluate physical fitness by a questionnare and treadmill test and assess readiness and resistance to change the lifestyle using the analog scale (e.g. from 1 to 10).

The goal of therapy is set depending on the severity of the findings. But at least 10% weight loss in 6 months is always considered reasonable.

In milder cases we administer lifestyle changes and pharmacology only if comorbidities appeared, in moderate cases we administer both of the above stated methods and we may include even surgical treatment if comorbidities are diagnosed and in most severe cases, i.e. in BMI over 40, we use all 3 of above stated treatment methods.

The lifestyle changes focus on diet, physical therapy and behavioral therapy. Dietary interventions include caloric restriction by 500-1000 kcal/d, replacement of regular meals by meal replacements (such as prepared entrees or canned food) as it makes it easier to control the portion size, proper proportion of macronutrient with the standart set as 45-65% of carbohydrates, 20-35% of fat and 10-35% of proteins. As an alternative could be used a low carbohydrate-high protein diet. Another useful concept introduced a lower caloric density foods, i.e. the same amount of food as eaten usually but lower in caloric content due to higher fiber and/or water content. A very aggressive approach is represented in very low calorie diet with caloric intake lower than 800 kcal/d. It's indicated only in very severe cases where the patient could benefit from a rapid weight loss, is motivated and failed to lose weight using other conservative strategies. Rapid weight loss is associated with higher risk of gall stones formation (due to decreased proportion of biliary acids in bile) which could be prevented by prophylactic administration of ursodeoxycholic acid in a dose of 600mg/d. It's important to make sure that the patient gets a recommended dose of vitamins a micronutriens, fiber and nonsaturated

fatty acids even over the period of above mentioned dietary intervention. It was proved that self monitoring (i.e. regular checking of weight on an own scale at home) is a positive predictor of weight outcome. [2] On the other hand, as a negative predictor is seen a number of previous attempts to loose weight. [2]

Physical therapy is more successful if combined with dietary interventions and it seems to be very useful in weight loss maintenance. Recent recommendation for a general population advice to engage in a moderate physical ectivity for at least 30 min a day most days a week, preferably every day. As a weight loss strategy, the physical activity needs to be implemented gradually as most patient have major difficulties implementing exercise into their life and may need a help from exercise physiologist or personal trainer. A step-meter could be used to assess the physical activity related to activities of daily living which also plays an important role. The support by friends and family was shown to have a positive affect on weight outcome achieved by regular physical activity. [2]

All of the above mentioned strategies are difficult for the patient to implement and keep. Thus a cognitive-behavioral therapy is essential. It should help the patient realize what, when, where a how should be implemented. That should be stated by the patient. He or she should also state the changes in lifestyle he or she is expecting to achieve till the next office visit and on the next visit, this could be checked as a indicator of a successful progress.

Pharmacological treatment is only indicated in BMI over 27 if comorbidities are present of over BMI 30 even without comorbidities. It should be always combined with a lifestyle program that shows how to use these drugs as such a combination improves the results. Medication could be devided in 3 groups – centrally acting monoamine increasing drugs that lower appetite, drugs decreasing absorption of macronutrients from

gastrointestinal tract and drugs suppressing the endocannabinoid system. The only approved centraly acting agent is sibutramin, serotonin and norepinephrine reuptake inhibitor. It could cause mild gastrointestinal problems but also increases blood pressure and heart rate for which uncontrolled hypertension, coronary heart disease, congestive heart failure or history of stroke are contraindication of sibutramin. GI acting drug orlistat is a slowly reversible inhibitor of lipases and causes a 30% decrease in absorption of dietary fat. It has no systemic side effects but can cause gastrointestinal problem such as oily stool or more frequent defecation. These side effect usually decrease with time and could be also treated with administration of psyllium mucilloid together with orlistat. Endocanabinoid system acts via 2 types of receptors, CB1 found in brain and CB2 found on the surface of immune cells. CB1 is increasing appetite both directly and by regulating the action of other substances influencing appetite. CB1 antagonist rimonabant has been proved to be an effective antiobesitic agent. Unfortunatelly has side effects such as depression, anxiety and nausea.

The surgical treatment is approved for the most severe cases, i.e. in BMI over 40 or over 35 in case of severe medical comorbidities. The procedures can be in general divided into 2 groups – restrictive and restrictive-malabsorptive. Into the first group counts laparoscopic adjustable silicone gastric banding. The latter includes Roux-en-Y gastric bypass and biliopancreatic diversion. These treatment methods proved to be the most efficient in cases of severe obesity and reduce weight for over 30% in nearly 60% of patients for longer then 5 years. But restrictive-malabsorptive procedure come with the risk of vitamin and micronutrient deficiency and so the patient needs to have the substances supplemented (B12, iron, folat, calcium, vit D etc...).

Without further discussion, which could be a basis for another thesis, I'd just like to mention the fact observed in the past, that the weight loss does not always have a positive effect on patient's prognosis in terms of mortality and morbidity. [3] This brings an important goal to not just find

an effective way of losing weight but also determining who is supposed to lose weight and who should rather keep it.

Figure 5 – Weight change and mortality

Weight change and mortality: the Nord-Trøndelag health study

Change in BMI[a]	Person-years	Total mortality			CV mortality		
		No. of deaths	Age-adj. RR	Multivariable[c] RR (95% CI)	No. of deaths	Age-adj. RR	Multivariable[c] RR (95% CI)
Males							
Loss	6521	319	1.7	1.6 (1.4–1.8)	150	1.7	1.5 (1.2–1.9)
Stable	37 617	627	1.0	1.0	285	1.0	1.0
Gain	65 124	607	1.0	1.0 (0.9–1.1)	274	1.0	1.0 (0.9–1.2)
Females							
Loss	10 214	327	1.8	1.7 (1.5–2.0)	150	1.8	1.7 (1.3–2.1)
Stable	32 683	357	1.0	1.0	147	1.0	1.0
Gain	84 931	435	0.8	0.9 (0.8–1.0)	178	1.0	1.0 (0.8–1.3)

Droyvold et al. J Intern Med. 2005 Apr;257(4):338-45

1.7. Weight maintenance

So far, most of the projects focusing on weight management were short term, i.e. up to 6 months in duration. In range of these 6 months the effect of dietary or another intervention tends to seem positive.

These short term studies suggested as positive predictive factors of weight maintenance: lower depression score, more years of weight maintenance. On the other hand, weight regain was correlated with decreased dietary restraint, increased susceptibility to overeating, small relapses and inconsistent dietary habits (e.g. different dietary patterns on weekdays and on weekends) [2]

But the problem is, that in a longer run, i.e. 1 year and more, which actually better represents the course of weight management in patient's life, most of the interventions fail to make the patient keep the weight loss he achieved.
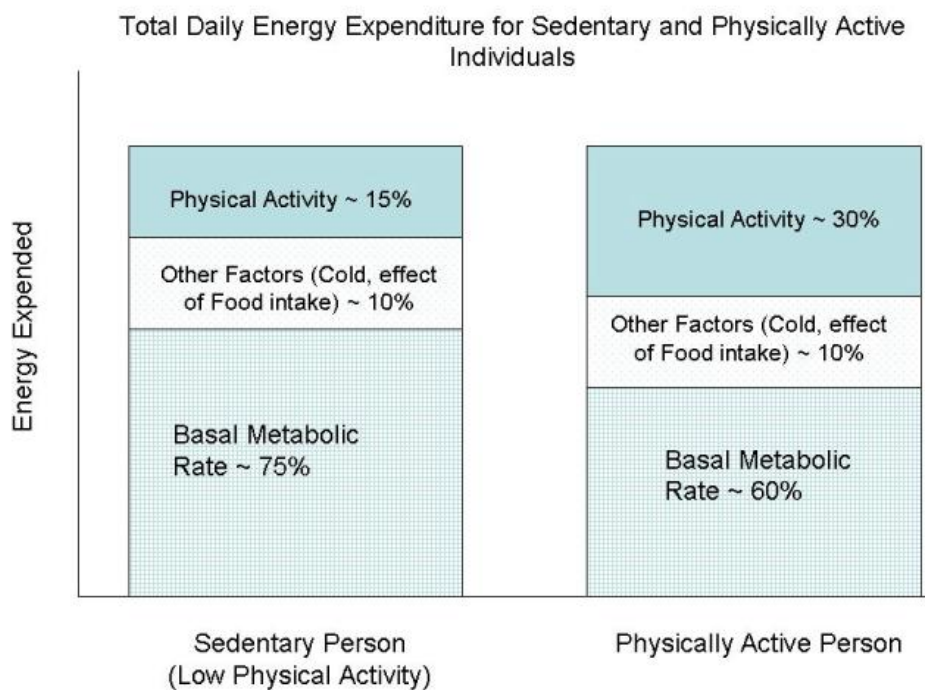
Since so far no predictors of a successfull weight loss and weight maintenance for a longer time span were investigated, we set this as a goal for our study.

2. Weight regulation

2.1. Major regulators

Body weight is regulated by endocrine and neural system. Changes in body weight are a result of the difference between energy intake and expenditure. The intake is regulated by appettite which is a result of action of hormonal, metabolic a neural agents on hypothalamus. Surprisingly, it's not physical activity that influences the energy expenditure most. It's the basal metabolic rate which is from a great extent fixed.

Figure 6 – Energy expenditure

Total Daily Energy Expenditure for Sedentary and Physically Active Individuals

Energy Expended

Physical Activity ~ 15%

Other Factors (Cold, effect of Food intake) ~ 10%

Basal Metabolic Rate ~ 75%

Physical Activity ~ 30%

Other Factors (Cold, effect of Food intake) ~ 10%

Basal Metabolic Rate ~ 60%

Sedentary Person (Low Physical Activity)

Physically Active Person

From: http://gaagdnewsletter.sitesuite.ws/page/nutrition_for_sport.html

Thus, influencing the body weight through changes in energy expenditure is somewhat slow.

Interestingly, obese have higher basal metabolic rate but they also eat more than non-obese people. That may imply that if they didn't eat more, they would not keep their higher weight. But if an obese person starts loosing weight, at the point he or she reaches his or her ideal weight, the

basal metabolic rate is lower then that of the people with the same BMI but without history of obesity. That makes it extremely difficult to maintain lower weight.

In the regulation of body weight, adipose tissue is a very key element. It comprises adipocytes and vascular-stromal compartment. Adipocytes store lipids but also produce many substances influencing body weight balance, blood pressure, coagulation, inflamatory response etc. In the stromal compartment reside among other preadipocytes that evolve into adipocytes under the influence of PPAR gama.

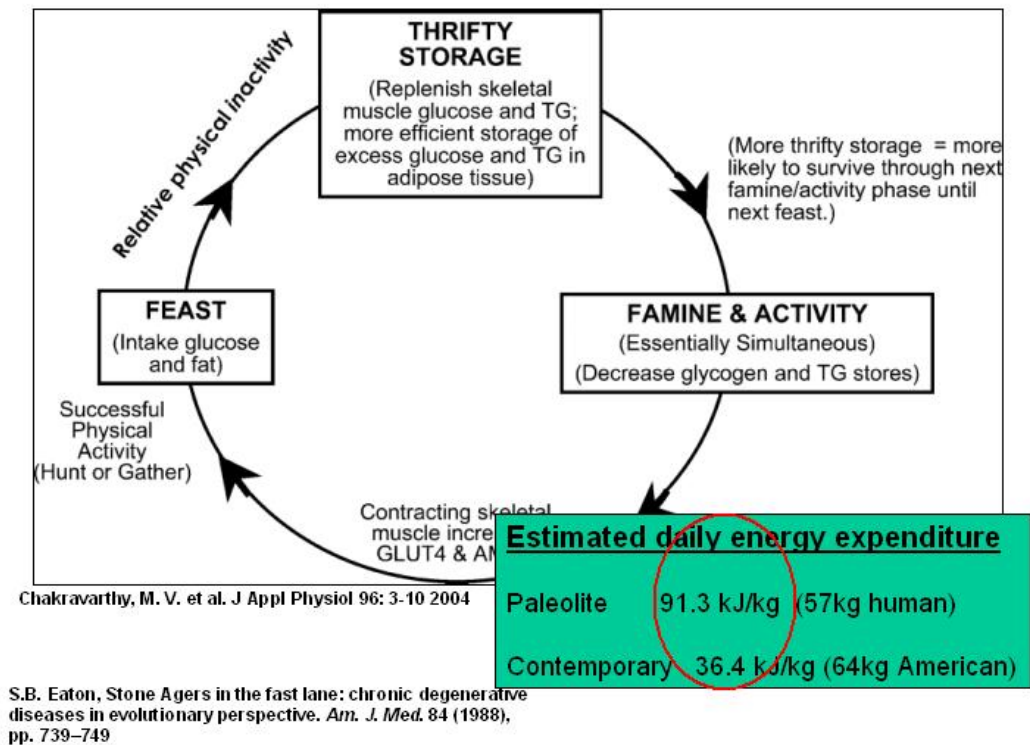Figure 7 - Adipocyte and the substances it produces
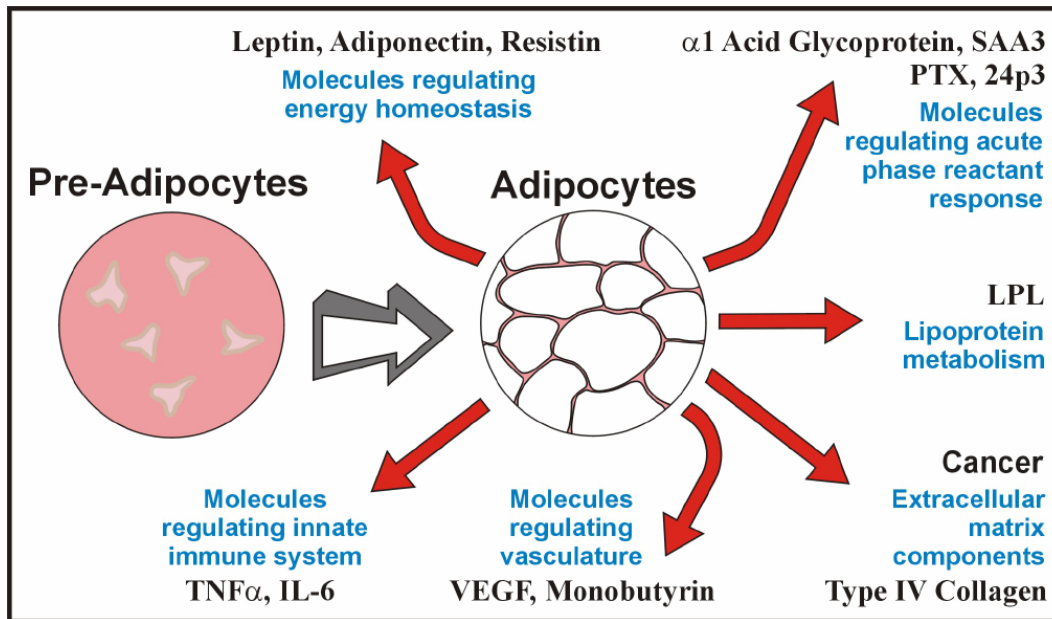
from: Physiol. Res. 54: 133-140, 2005

In and individual, genes seem to be a more important factor in development of obesity than the environment, but if considering the whole population, it seems to be the environment, that changed so much and thus influences our lifestyle in the past decades, that it made the majority of population to be prone to obesity.

Figure 8 - Average energy expenditure change from paleolite

THRIFTY STORAGE
(Replenish skeletal muscle glucose and TG; more efficient storage of excess glucose and TG in adipose tissue)

Relative physical inactivity

(More thrifty storage = more likely to survive through next famine/activity phase until next feast.)

FEAST
(Intake glucose and fat)

FAMINE & ACTIVITY
(Essentially Simultaneous)
(Decrease glycogen and TG stores)

Successful Physical Activity
(Hunt or Gather)

Contracting skeletal muscle incre... GLUT4 & AM...

Chakravarthy, M. V. et al. J Appl Physiol 96: 3-10 2004

Estimated daily energy expenditure

Paleolite 91.3 kJ/kg (57kg human)

Contemporary 36.4 kJ/kg (64kg American)

S.B. Eaton, Stone Agers in the fast lane: chronic degenerative diseases in evolutionary perspective. Am. J. Med. 84 (1988), pp. 739–749

The risk factors in adult include a female gender and lower income. In children the risk increases with time spent watching television. Also lipid rich diet especially when combined with simple carbohydrates contributes to weight gain. Among less known and proved risk factor are sleep deprivation and changes in gut flora.

The gene most important for development of obesity is the one for leptin. Leptin has many effects which include increase of energy expenditure and decreases of appetite by increasing POMC, which then needs to be converted by proconvertase to alfa-MSH that acts on it's receptor and it's effect decreases appetite. Failure of any of these steps or a disorder in one of many others regulatory pathways can cause severe obesity but these disorder are very rare.

Another gene playing a major role is adiponectin. Both adiponectin and leptin will be further discusses in the following text.

2.2. Leptin

2.2.1. General description

Leptin is secreted from adipocytes but also from stomach epithelium and placenta. [4] It's synthesis is influenced by unknown factors. Insulin and glucocorticoids are believed to be two of them. [4] It's levels are also increased by the cell fat content [4] and it's also in a positive correlation with body fat percentage.

Figure 9 – Control of leptin synthesis and secretion

Control of leptin synthesis and secretion

From: http://www.vivo.colostate.edu/hbooks/pathphys/endocrine/bodyweight/leptin.html
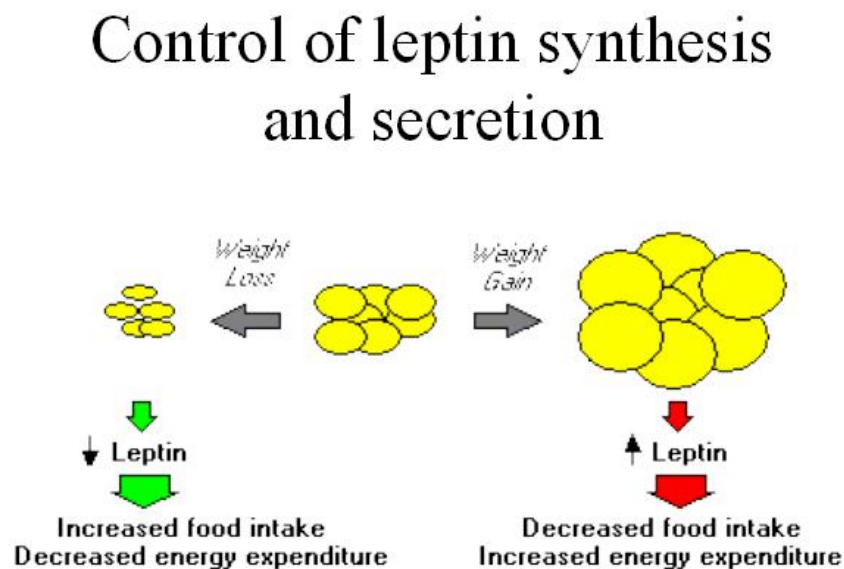
2.2.1. Effects of leptin

Leptin has receptors in brain, where it plays an important role in body weight regulation. Receptor are also found on the surface of T lymphocytes, vascular endothelium. [4]

Leptin is leading to weight loss. On one hand it's lowering energy intake due to the decreased appetite. Leptin lowers the synthesis of stimulators of feeding behavior, NPY, agouti related polypeptide [5] and endocanabinoids [6] and increases production of alfa-MSH [7] which promotes satiety .

On the other hand leptin increases energy expenditure. It elevates body temperature and oxygen consumption and by decreasing the amount of fat tissue it increases energy expenditure neccessary for keeping the body temperature.

Figure 10 – Effects of leptin
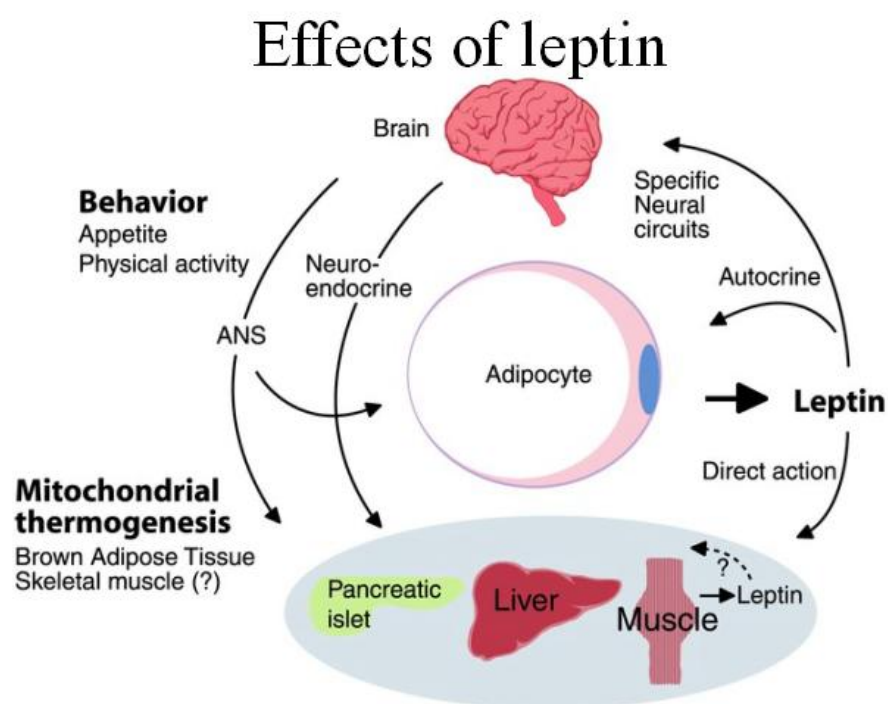
Obesity and insulin resistance, Barbara B. Kahn, Jeffrey S. Flier, *J Clin Invest.* 2000; 106(4):473

Surprisingly, leptin has an affect on reproductive system. It enhances reproductive functions probably by increasing the secretion of gonadotropin RH and LH and FSH

Leptin also enhances immune functions. It's also said to modulate the development of atherosclerosis. That could be exactly due to the different lymphocytes response. [8]

Among the less discussed function is the role in angiogenesis and the regulation of surfactant synthesis in fetal lungs.

2.2.3. Epidemiology

What the epidemiology of leptin variations concerns, it needs to be said, that the mutation in Ob gene (the gene for leptin protein) or leptin receptor are very rare. [9]

It was proved that obese people have higher levels of leptin in their blood.

Figure 11 – Leptin levels in obese
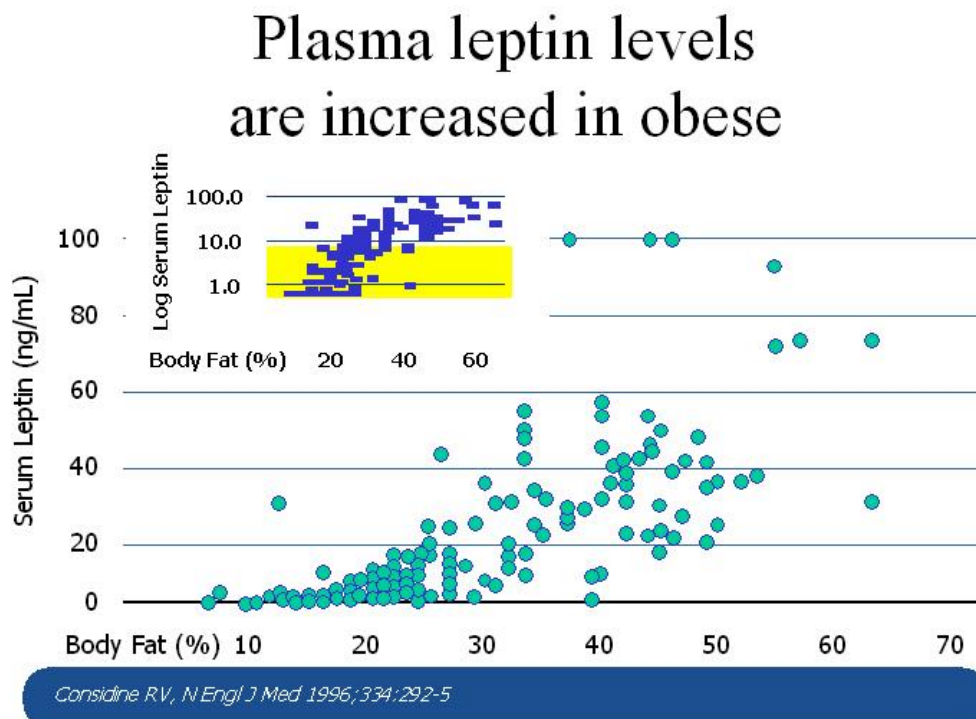
Since they're obese it shows that leptin is not having it's effect and this phenomenon is explained as leptin resistance. Studies suggest that the development of leptin resistance depends on the type of diet, not just on the final body fat percentage. In rodents, leptin resistence reached higher level if fructose was administered at higher ratio in diet. [10]
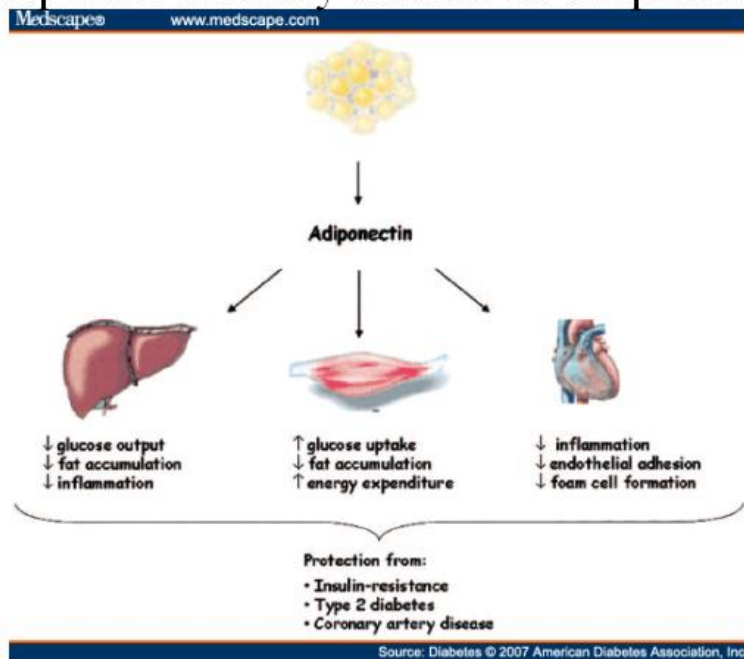
2.3. Adiponectin

2.3.1. General description

Adiponectin is a hormon secreted exclusively from adipose tissue. It has a relatively high plasma levels – 0,01% of all proteins, i.e. 5-10 ug/ml. It's level in blood is negatively corellated with body fat percentage – it's reduced in diabetic subjects and weight reduction increases adiponectin. It's levels are higher in females than in males. [11]

2.3.2. Effects

Adiponectin is supressing metabolic derrangement that can cause diseases such as diabetes type 2, endotelial dysfunction and subsequent atherosclerosis, metabolic syndrome and nonalcoholic fatty liver disease. It does so by influencing metabolic pathways of energy substrates. It influences beta oxidation (thus changing RQ) and triglyceride clearance, gluconeogenesis, glucose uptake. As a result, it's also improving insulin sensitivity. [12] The final result of all above stated changes is weight loss. But adiponectin exerts it's weight reduction potential via brain too. [13]

Figure 12 – Proposed salutary effects of adiponectin

From: http://www.medscape.com/viewarticle/557976

2.3.3. Epidemiology

Variants in the adiponectin gene influence adiponectin levels, adiposity and plasma lipid levels. A haplotype in the adiponectin gene promoter (-11391, -10068 and -4521) was significantly associated with higher plasma adiponectin levels in non-hispanic white. [14]

3. THE NUGENOB STUDY FOLLOW-UP

3.1. The NUGENOB study

We took an advantage of an existing subjects database created in 2003 under the EU-funded project called Nugenob. The name stands for „Nutrient-Gene interactions in human obesity – implications for dietary guidelines“. There where 770 participants of caucasian origin in the study – 607 women and 163 men.

3.2. Methods

They met the criteria which were age between 20 and 50 and BMI of 30 or over. Excluded were those who experienced a weight change of more than 3 kilograms in tha past 3 months, those who had hypertension, diabetes or hyperlipidemia on medication, untreated thyroid gland disease, surgically treated obesity, those who were pregnant at the time of the study, drug users and alcohol drinkers and those who were taking part in any other weight loosing programme or study. In the group of slim individuals, also those who had a history of BMI higher than 25 or history of any medication use except for contraceptive pills and medication for treatment of any chronic illness. Participants were hired through the media or were refered by their general practitioner or any other medical personell or institution in the United Kingdom, Netherlands, France, Spain, Czech republic, Sweden and Denmark. The protocol was approved by an ethical committee and each participant signed the conscent before the study started.

The measurements were taking place in the local centrers in the above stated countries. All the participants underwent a 1-day clinical examination. On the day the study started, they came to the laboratory at 8am after 12 hour fasting and 3 days of a regime that consisted of participant's regularly diet, alcohol restriction and an excessive physical activity. After emptying the urinary bladder the subjects were taken their

anthropometrical measures. The height was taken with no shoes and using a standardized weight measuring device. The weight was measured three times with underwear on only and the average was used in the following calculations. The waist and hip circumference was measure in the underwear that is not any compressive. Thenafter, the body composition was evaluated in the subject in a supine position, using a multi-frequency bioimpedance (Bodystat, Quadscan 4000, Isle of Man, British Isles.) The participants where also taken blood tests for glucose, cortisol, free fatty acids, triglycerids, insulin-like growth factor-I and leptin and 39 randomly selected individuals were also tested for adiponectin levels.

As a part of the study, further assesment was made including questionnaires investigating dietary habits, lifestyle etc. Further measurements including basal metabolic rate, respiratory quotient and energy expenditure and dynamics of various metabolits in the bloodstream in a fasting state and then 3 times in 1 hour intervals after drinking a high fat meal (95% fat from which 60% were saturated fatty acids, 3% carbohydrates and 2% proteins) in the amount counting for one half of the basal metabolic energy expenditure. The drink had to be drunk in less than 10 minutes. But since these details are not our data input source and were prooved to have no statistically significant effect on the measures obtained later in the study, I'm not going to describe the techniques of those measurements any further.

Then the individuals underwent a 10-week hypocaloric dietary programme under which they were randomly divided into 2 group with eather low fat/ high carbohydrate diet (20-25% of energy from fat, 15% of energy from protein, 60-65% of energy from carbohydrate) or high fat/ low carbohydrate diet (40-45%, 15%, 40-45% of energy income in the same respective groups of nutrients). The subjects were tested again 10 weeks after the study started and for most of the collaborating centers that was

the end of data gathering. For adiponectin, only the previously selected 40 individuals were retested.

The Department of Sport Medicine at the 3rd Faculty of Medicine at Charles University in Prague has decided to use their patient database from the Nugenob study and retest the patients 1 year after the intitial study started. Then, the methods were the same as in the initial study, except the adiponectin measurement, which were not performed.

All the data gathered were analysed using SPSS 16.0.0 software. In the analysis we created a new variable, leptin-resistance-index, as a ration between leptin levels and fat mass.

3.3. Results

Figure 13 - Descriptive statistics – mean values and standard deviations

	baseline	10 weeks	1 year
weight [kg]	100,52 ± 16,59	93,65 ± 16,43	95,24 ±15,15
waist [cm]	106,11 ± 12,90	99,55 ± 12,76	102,59 ± 12,46
hip [cm]	119,38 ± 10,33	114,43 ± 10,82	117,01 ± 8,16
FFM [kg]	59,45 ± 11,57	58,00 ± 11,34	57,68 ± 9,56
FM [kg]	41,14 ± 11,76	35,81 ± 11,89	36,82 ± 8,77
leptin [ng/ml]	29,38 ± 15,25	20,08 ± 12,96	
adiponectin [ug/ml]	15,98 ± 11,05	15,57 ± 10,26	

Higher baseline leptin is associated with lower reduction of weight, waist and fat mass between the beginning and the 10th week.

Higher baseline leptin-resistance-index is associated with lower reduction of weight, waist and fat mass between the beginning and the 10th week.

Higher decrease of leptin between the beginning and the 10th week was associated with higher increase of fat mass between the 10th week and 1 year.

Higher decrease of leptin-resistance-index between the beginning and the 10th week was associated with higher increase of fat mass between the 10th week and 1 year.

Nor baseline adiponectin neither it's change between the baseline and the 10th week are associated with any of the investigated anthropometric measures.

Association between baseline leptin and reduction of anthropometric measures between the baseline and the 10th week are: with weight reduction (r = -0,149; p < 0,000), waist reduction (r = -0,144; p < 0,000) and fat mass reduction (r = -0,66; p = 0,98).

Association between baseline leptin-resistance-index and reduction of anthropometric measures between the baseline and the 10th week are: with weight reduction (r = -0,180; p < 0,000), waist reduction (r = -0,150; p < 0,000) and fat mass reduction (r = -0.176; p< 0,000).

Association between decrease in leptin between baseline and 10th week and fat mass increase between the 10th week and 1 year shows r = 0,202, p = 0,64).

Association between decrease in leptin-resistance-index between the baseline and the 10th week and fat mass increase between the 10th week and 1 year shows r = 0,231, p = 0,34.

3.4. Discussion

Leptin is supposed to help in weight and fat mass reduction. But here we can see that the association was negative. That could be explained by the phenomenon of leptin resistance. That means that leptin resistance is higher when there's more leptin on each weight unit of fat mass. In this state, leptin can not excert it's influence in the body. One of the putative effects is an influence on central nervous system through which leptin stimulates sympatoadrenergic system and thus increases energy expenditure and eases weight loss.

The above stated assumption is supported by our results of correlation analyses between anthropometrical changes and leptin-resistance-index, which we counted as a ration of leptin levels and fat mass.

Contrary, decrease in leptin till the 10th week is associated with increase of fat mass till 1 year. One of the possible explanation could be, that after 10 weeks of weight reduction program, leptin resistance disappeared and then, leptin could excert it's full potential. If at this time, leptin was lower, it could not stimulate sympatoadrenergic system so much and that resulted in increase of fat mass. Another possible explanation is, that in subjects, where baseline leptin was higher, it was easier to decrease it. But after the end of weight reduction programme these patients came back to their lifestyle and gained more fat mass as they were getting back to the state from which they started at baseline.

Again, the above stated analyses was consistent with analyses of correlation between leptin-resistance-index and anthropometric changes between 10th week and 1 year.

What adiponectin concerns, the fact that none of the investigated anthropometrical parameters was in a significant correlation with baseline adiponectin level and it's dynamic, could be due to the fact, that in our study of less then 40 patients, adiponectin changes between baseline and the 10th were very minor (average baseline adiponectin level was 15,98 ± 11,05 and at 10 weeks it was 15,57 ± 10,26).

Conclusion

In this study I tried find a tool of recognizing high risk patients at the beginning of a weigh losing programme so that they could receive more care and finally achieve satisfactory results just as the other patients.

I found out, that high leptin at the beginning is a risk factor for worse outcome after 10 weeks (i.e. lower weight loss, lower waist circumference reduction and lower fat mass reduction) and explained this by the existence of leptin resistance, a state in which body is insensitive to the effects of leptin, which is in general known to decrease body weight. Thus, improving leptin effects in secondary prevention, i.e. weigh reduction programmes in obese and overweight patients, could have a positive outcome.

The first possibility would be leptin administration. Even this method could be reasonable just as insulin administration in diabetes type 2 patients who are impossible to be compensated with the use of peroral antidiabetics. But just as in the case of diabetes type 2, more reasonable treatment would be administration of leptin-sensitizing substance. Some studies suggest that amylin, hormon produces by pancreatic beta cells, is an important leptin sensitivity enhancer. [15]

I also found out that decrease in leptin after 10 weeks of low-calorie diet programme was a risk factor for a long term (1 year) outcome. One of the explanations could be, that after fasting period, leptin sensitivity is restored and then, by the time leptin can fully excert it's potential in weight and fat mass losing proceses, it's low levels are associated with worse outcome. Here, after the leptin sensitivity is restored, even leptin administration would be a potential tool of improving the results of secondary prevention, i.e. it would help the previously obese patients who lost some weight to keep it and not regain it.

Unfortunatelly, adiponectin did not prove to be a good predictor of anthropometrical changes in our study. But this could be due to our relatively small patient population for adiponectin testing, which was smaller than 40 individuals. But adiponectin is know to be in a positive correlation with better metabolic state. Thus, increasing adiponectin by medications such as thiazolidinediones (throught PPARγ activation) [16] could be reasonable and have a positive effect in prevention of complications of obesity. The issue of influence of adiponectin deserves more attention and should be further investigated by another study involving more then just 40 patients.

Summary

In prior times, storing fat was an important capability of human body that helped to survive. But these days, human in the developt countries do not lack energy from nutrition any more and fat storing processes lost much of their benefits and are showing their costs in the form of obesity and it's complications.

Obesity is a stage of increased body fat content. It's defined using a ratio betwen body weight and square of body height, so called body mass index, BMI. Obesity is represented by BMI higher then 25.

In the Czech republic, about 20% of women and 16% of men are obese. Similar situation is in the most developed countries, topped by the USA, where about 30% of the population are meeting the obesity criterion.

In the etiology of obesity, we can distinguish internal and external factors. What the internal factors concerns, obesity could be either secondary to another disease, which is relativelly rare or primary. Primary obesity is in a very small number of cases caused by an monogenic inherited pattern. Most often it's caused by a failure of body regulation processes. That means a dysbalance between energy intake and energy expenditure. And it just this balance that's also disrupted by external factors. In tha last decades, energy intake was made very easy for our population but energy expenditure decreased with the change from manual labour to sedentary lifestyle such as administrative work.

Obesity comes with a number of complications. Among them are diabetes mellitus type 2, hypertension, atherosclerosis, depression, reproductive disorders, arthrosis and others. Thus, preventing and treating obesity has a major effect on the health outcome.

In order to achieve better results in weight reduction efforts, the treatment of obesity needs to run on an individual basis. Each patient needs to be first assessed in term of his anthropometrical, biochemical and psychological measures and then an individual goal needs to be established. 10% weight loss in 6 months is seen as a minimum. The treatment always starts with lifestyle changes. If those fail, pharmacological treatment take place. In the most severe cases, also surgical treatment come available.

But even if a considerable weight loss is achieved, weight maintenance is still a major problem. In order to help with this issue, we've decided to try to find predictors of weight maintenance parameters so that patients in higher risk could receive more attention.

The role of adipokines, leptin and adiponectin, is broadly discussed. These two substances were the main object of interest in our study, too.

Leptin is secreted mainly from adipocytes and it's levels increase with cell fat content and body fat percentage. Leptin itself is supposed to lead to weight loss, both by decreasing appetite and increasing energy expenditure. But high levels of leptin can be a signal of so called leptin resistance, a state in which leptin is losing it's effects.

Adiponectin is secreted exclusively from adipose tissue. In obese people, lower levels of adiponectin were proven (i.e. adiponectin levels are in a negative correlation with body fat percentage). That's consistent with the fact that adiponectin is affecting a range of metabolic prosesses and thus improves insulin sensitivity and metabolic profile.

For evaluation, we used a obese patient database (of almost 750 subjects), that was build under the Nugenob study, conducted in 8 cities in

7 european countries (Denmark, the Netherlands, Sweden, Great Britain, Czech republic, France (2 centers) and Spain. The study consisted of many different measurement. What is important for this thesis, all the participants were measured their anthropometric and biochemical parameters at the baseline and 10 weeks later, at the end of a standardized weight reduction program. 1 year after the initial study, approximatelly 100 subjects were retested for their anthropometrical measures in the laboratory of Department of Sport medicine, 3rd Faculty of Medicine, Charles University in Prague. Based on the data gathered, we tried to determine whether leptin and adiponectin levels in the initial study could be used as predictors for the measures taken 1 year later.

I found out, that higher baseline leptin and the ratio between leptin and fat mass (which we named leptin-resistance-index) is a risk factor for worse outcome after 10 weeks of low-calorie diet. This could be explained by the phenomenon of leptin resistance, in which leptin can not excert it's potential and help in weight and fat mass reduction. Thus, I suggest that recognizing patients with high leptin or leptin-resistance-index at the beginning of weigh reduction programmes that are part of secondary prevention of obesity (i.e. prevention of obesity complications), could be useful. These patients may need to receive extra attention and care in order to achieve satisfactory results. A part of this care could be administration of some leptin sensitizing agent (e.g. the putative leptin sensitizer – amylin.) [15]

I also recognized a bigger decrease of leptin and leptin-sensitivity-index between baseline and 10th week as a risk factor for a worse outcome between 10th week and 1 year. Here the worse outcome was represented by higher increase in fat mass. This could be explained as a lack of leptin after leptin sensitivity was restored by 10 weeks of low-calorie diet. Also these patients could receive an extra care as a part of

secondary prevention of obesity, and could be for example administered leptin.

I did not find any significant correlation between adiponectin and the anthropometrical measures investigated. I suggest that this could be due to lower patient database (40 subjects) and the fact that in this patient population, adiponectin changes between baseline and 10th week were not any significant. But since adiponectin is known to have a positive effect on the general metabolic state, it's role in changes of anthropometric measure in obese patient in secondary prevention of obesity should be deeply investigated in a study involving more patients.

Souhrn

Schopnost ukládat energii ve formě tuku byla v minulosti velmi důležitá pro přežití. Ale v dnešní době, kdy již nedostatkem potravy netrpíme, tato schopnost pozbyla mnohé ze svých výhod a naopak se objevily její stinné stránky v podobě obezity a jejích komplikací.

Obezita je spojena s větším obsahem tuku v těle a je obvykle definována pomocí poměru tělesné hmotnosti a druhé mocniny tělesné výšky, tzv. body mass indexu, BMI. Jako obezita je označen stav s BMI nad 25.

V České republice trpí obezitou asi 20% žen a 16% mužů. Podobná situace je ve většině vyspělých zemí. Například v USA je prevalence obezity až 30%.

Na rozvoji obezity se podílí vnitřní a vnější faktory. Mezi vnitřní faktory se řadí například genetická výbava, která může způsobit vzácnou dědičnou primární obezitu, nebo různé nemoci, u kterých se obezita rozvíjí sekundárně . Nejdůležitějšími vnitřními faktory jsou ale parametry systému řídícího rovnováhu příjmu a výdeje energie. Tuto rovnováhu kromě vnitřních faktorů ovlivňuje výrazně i vnější prostředí. To se v posledních desetiletích hodně změnilo ve smyslu přechodu z fyzické práce na prace sedavou, se kterou se pojí nižší energetický výdej.

S obezitou se pojí celá řada komplikací. Patří mezi ně diabetes mellitus 2. typu, hypertenze, ateroskleróza, deprese, poruchy reprodukce, artróza nosných kloubů aj. Prevence a léčba obezity je proto důležitým cílem v péči o zdraví.

K léčbě obezity je nezbytné přistupovat individuálně. Je potřeba nejdříve posoudit fyzické, biochemické i psychické parametry pacienta a

potom stanovit reálný cíl léčby, kterým by měl být vždy úbytek minimálně 10 procent váhy za 6 měsíců. Jako první přichází na řadu režimová terapie a teprve v případě, že selže, dostává se ke slovu farmakoterapie, ev léčba chirurgická.

Obtížné ale není pouze snížení váhy, ale také jejich udržení. Právě na něj jsem se zaměřil ve své práci a snažil se najít klíč k včasnému rozpoznání pacientů, kteří jsou ve větším riziku opětovného příbytku váhy, aby bylo možné těmto pacientům včas pomoci.

Účinky adipokinu, leptinu a adiponektinu, jsou v dnešní době široce diskutovaným tématem. I já jsem se na ně ve své práci zaměřil.

Leptin je hormon sekretovaný hlavně adipocyty. Jeho sekrece se zvyšuje s množstvím tělesného tuku a obsahem tuku v buňkách. Leptin působí pozitivně na úbytek váhy, jak prostřednictvím zvýšení energetického výdeje v periferních tkáních, tak snížením příjmu potravy. Vysoké hladiny leptinu se ale mohou pojit se stavem, který se nazývá leptinovou rezistencí, kdy se účinky leptinu projevují jen v omezené míře.

Adiponektin je produkován výhradně tukovou tkání. Bylo prokázáno, že hladiny leptinu jsou nižší u obézních pacientů v porovnání s normální populací. To může souviset s tím, že adiponektin má pozitivní vliv na mnoho metabolických procesů a zlepšuje inzulínovou citlivost a metabolický profil a proto je jeho nedostatek predispozicí pro metabolická onemocnění.

Pro svou práci jsem využil data získaná v rámci projektu Nugenob, který probíhal v 8 evropských městech a kterého se účastnilo přibližně 750 osob. V rámci studie prošli dobrovolníci desetitýdenní dietní intervencí. Před začátkem intervence a po jejím skončení byli všichni vyšetření. Byla odebrána kompletní antropometrie (výška, váha, obvod pasu, boků), změřeno množství tělesného tuku a netukové hmoty, odebrána krev pro

stanovení řady parametrů včetně hladin leptinu a u 40 osob také adiponektinu. Pro většinu výzkumných center tím studie skončila. Ústav tělovýchovného lékařství ale ve studii pokračoval a pozval si pacienty ze své datábáze na kontrolu výše zmíněných parametrů ještě po 1 roce od ukončení původní studie. Na základě takto získaných dat jsem se snažil odhalit možnost predikce změn antropometrie na základě hladin leptinu a adiponektinu.

Zjistil jsem, že vyšší počáteční hodnota leptinu, stejně tak jako vyšší poměr leptinu a tukové hmoty (coby ukazatel leptinové rezistence) souvisí z menším úbytkem hmotnosti, obvodu pasu a tukové hmoty po desetitýdenní dietní intervenci. Jedním z možných vysvětlení je, že vysoký leptin ukazuje na již zmíněnou leptinovou rezistenci, která snižuje účinek leptinu ve smyslu snížení váhy a tukové hmoty. Vyšší hodnoty leptinu nebo poměru leptinu a tukové hmoty by tedy mohly sloužit jako ukazatel, podle kterého by bylo možné rozpoznat rizikové pacienty v rámci sekundární prevence obezity, tzn. prevence komplikací, a takovým pacientům věnovat zvýšenou pozornost, aby i oni dosáhli požadovaných výsledků. Jednou z možných intervencí je podání látky zlepšující leptinovou rezistenci. Podle některých studií by pro to byl vhodný amylin, látka produkovaná pankreatem. [15]

Jako rizikový se ukázal být větší pokles leptinu nebo poměru leptinu a tukové hmoty mezi začátkem studie a 10. týdnem, který se pojil s větším nárůstem množství tukové tkáně mezi 10. týdnem a 1 rokem. To je možné vysvětlit tím, že po dietní intervenci byla obnovena leptinová rezistence a proto nižší hladiny leptinu opravdu odpovídaly nárůstu množství tukové tkáně. I tuto skutečnost je možné využít v rámci sekundární prevence obezity. Rizikoví pacienti by mohli například absolvovat terapii leptinem.

Mezi hladinou adiponektinu a sledovanými antropometrickými parametry jsem neobjevil žádnou signifikantní korelaci. To si vysvětluji

také malým souborem pacientů (40 osob) a skutečností, že změna hladiny adiponektinu mezi začátkem studie a 10. týdnem nebyla signifikantní. O adiponektinu je nicméně známo, že má pozitivní efekt na metabolický profil. Jeho možnosti v sekundární prevenci obezity by tedy neměly ujít dalšímu zájmu a bylo by vhodné věnovat se této problematice ve studii s větším počtem osob.

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A list of pictures, tables and graphs