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Circadian Rhythms of Carbohydrate Metabolism in Women with Different Types of Obesity

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Blood levels of glucose and immunoreactive insulin were assessed in women with android and gynoid types of obesity or normal body weight (control group) in the dynamics of oral glucose tolerance test in the morning and in the evening. In the control group, the mean concentrations of glucose and immunoreactive insulin were significantly higher in the evening at all test terms (0, 60 and 120 min), which is indicative of physiological insulin resistance in the evening. In the group of women with gynoid obesity, no difference in the levels of glucose and immunoreactive insulin was revealed in the morning and evening tests, but in the evening tests, glucose (60 and 120 min) and immunoreactive insulin (120 min) levels were lower than in the control group. In the group of women with android obesity, the evening glucose level on minutes 60 and 120 of the test was higher than in the morning, but immunoreactive insulin did not vary throughout the day and on minutes 60 and 120 it surpassed the corresponding parameter in the control group by 2-4 times. In case of gynoid obesity, glucose load was followed by hyperinsulinemia and hypoglycemia; in android obesity, it was followed by hyperinsulinemia, hyperglycemia, and insulin resistance irrespective of the time of the day.

Key Words: *women; gynoid and android obesity types; glucose tolerance test; insulin; circadian rhythm*

Growing prevalence of obesity is associated with increasing incidence of cardiovascular diseases, atherosclerosis, diabetes mellitus type 2, and other socially important diseases contributing to disability and mortality in the population [11]. In the pathogenesis of obesity, impaired energy balance with predominance of energy consumption over energy expenditure is usually considered as the main factor [11]. However, there is evidence that impaired circadian rhythm of energy metabolism is one of the causes of obesity [5,8]. It was shown that synchronization of circadian physiological and metabolic processes plays a role in body weight control [6] and prevention of cardiovascular diseases [10,12].

In recent years, the concept of "metabolically healthy obesity", when a person with obese phenotype has almost no specific metabolic disorders, is being actively developed [9]. We have shown that gynoid obesity, specified by lower fat distribution type, has the properties of "metabolically healthy obesity . In contrast, in case of android obesity (upper body fat distribution) marked metabolic disorders, hypoadiponectinemia, and insulin resistance are the risk factors of cardiovascular diseases, diabetes mellitus type 2, *etc*. [2]. The role of cyrcadian rhythms of energy metabolism, in particular carbohydrate metabolism, in different obesity types is unknown. The pathogenesis

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of obesity is based on insulin resistance of peripheral tissues that can be detected at the early stages by oral glucose tolerance test (OGTT).

The aim of this study was to investigate specific features of glucose utilization and pancreatic insular apparatus reactivity in response to glucose load in different obesity type in the morning and in the evening.

MATERIALS AND METHODS

The study included women aged 18-50 years; all examinees signed written informed consent. The study was approved by the Local Bioethics Committee. Anthropometric examination included body weight (kg), height (m), waist and hips circumference (cm) measurements. Body mass index (BMI) was calculated as body weight divided by the square of body height (kg/m^2). According to WHO, BMI≥30 kg/m^2 corresponds to obesity, normal body weight (NBW) is less than 25 kg/m^2. Obesity type was determined by the ratio of waist circumference to hip circumference (WC/HC); women with WC/HC <0.85 were included into group 1 (gynoid obesity; *N*=23) and women with WC/HC≥0.85 comprised group 2 (android obesity; *N*=10) [7]. Group 3 (control) included 22 women with NBW.

Standard OGTT was performed in the morning (08.30-10.30) and in the evening (20.30-22.30). The test was preceded by 12-h fasting. During OGTT, women drunk 75 g of glucose dissolved in 200 ml water acidified with citric acid. Peripheral blood samples were taken from fingertips before the load (0 min) and on minutes 60 and 120. Whole blood glucose was measured by the enzymatic method, serum immunoreactive insulin level was measured by ELISA using commercial kits.

Statistical data processing was carried out using Statistica 6.0 software (StatSoft). Results are presented as mean values and errors of mean (*M*±*m*). Comparative analysis was performed using Kruskal–Wallis test for multiple comparisons and Mann–Whitney test for paired comparisons. The minimum probability of the null hypothesis was accepted at 5% level of significance ($p<0.05$).

RESULTS

Baseline blood glucose determined by OGTT in the morning (Fig. 1) did not differ in all groups. Blood glucose level significantly increased in 60 min after load ($p<0.0001$ in all groups), while in 120 min, this value decreased in comparison with the value observed in 60 min ($p<0.0001$ for all groups). In women with NBW and android obesity, blood glucose level returned to initial level in 120 min and in women with gynoid obesity, it dropped below the baseline values

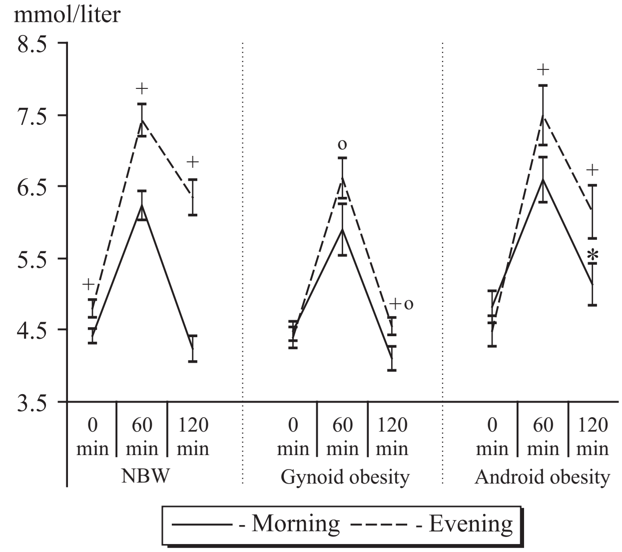

Fig. 1. Blood glucose level in women with different types of obesity in the morning and evening OGTT. Here and in Fig. 2: $p<0.05$ in comparison with NBW group *morning test, °evening test, +between morning and evening tests within the same group.

($p<0.05$). Higher glucose level in women with android obesity at minute 120 of the test type in comparison with women with NBW reflects decelerated glucose utilization in the morning.

In the evening, initial blood glucose level in different groups did not differ, however, in the NBW group, glucose level was higher in the evening than in the morning (Fig. 1). After load (60 min), blood glucose level in all groups also increased ($p<0.0001$ for all groups), but in women with android obesity and NBW it was significantly higher than in women with gynoid obesity ($p<0.05$). It should be noted that glucose level elevation on minute 60 of OGTT in the evening was more expressed than in the morning in all groups. In 120 min, glucose level decreased, but returned to initial values only in women with gynoid obesity. In women with android obesity and NBW, this value significantly surpassed the baseline ($p<0.0005$), which can be interpreted as impaired glucose tolerance in the evening. This is also evidenced by the fact that in women with android obesity type and NBW glucose level on minute 120 of OGTT in the evening was much higher than in the morning. However, glucose tolerance in women with android obesity was impaired in the morning and in the evening, while in women with NBW, it was revealed only in the evening test.

Measurement of serum level of immunoreactive insulin after morning OGTT revealed the highest values in women with android obesity in all three time points, intermediate in women with gynoid obesity, and the lowest values in women with NBW (Fig. 2). Serum immunoreactive insulin was elevated in 60 min after load in all groups ($p<0.0001$). By 120th minute,

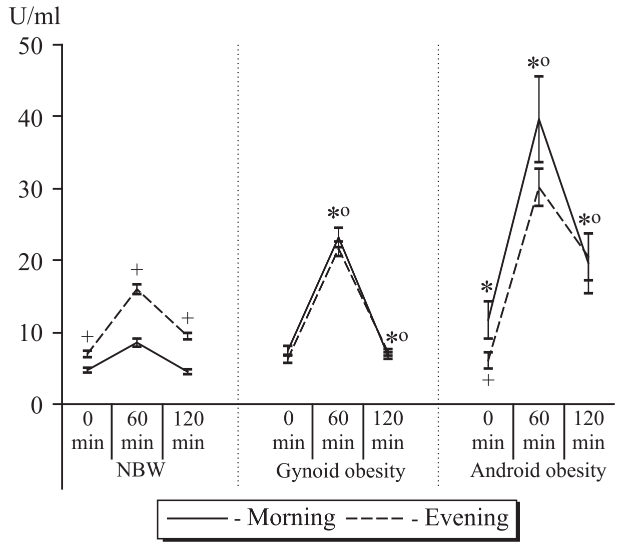

Fig. 2. Morning and evening levels of immunoreactive insulin in the blood of women with different types of obesity according to OGTT data.

this parameter decreased in all groups ($p<0.0001$), but to different extents; in women with android obesity, it remained maximum and significantly surpassed the baseline ($p<0.001$).

After evening OGTT, immunoreactive insulin in women with NBW was significantly higher than in the morning at all time points (Fig. 2). It should be noted that glucose level in the evening test was also higher and the decline of this parameter was decelerated by minute 120 (Fig. 1). The dynamics of serum immunoreactive insulin in women with gynoid obesity was similar in the morning and in the evening. In women with android obesity, this parameter remained above the baseline ($p<0.0001$) on minute 120 of OGTT (Fig. 2).

Obtained results suggest that in women with NBW, carbohydrates are the main energy substrate in the morning, while in the evening, energy metabolism is switched to preferential utilization of lipids, which is the cause of reduced glucose utilization rate in peripheral tissues and can be described as "physiological insulin resistance". This phenomenon can be related to regulation of preferential utilization of one or another substrate in energy metabolism at different time of the day [4,6]. This switching from carbohydrate to preferentially lipid metabolism in the evening is aimed at recovery of the structure and functions of somatic cells and elimination of excessive fat [4].

In women with gynoid obesity, functional hyperinsulinemia was revealed, which under conditions of carbohydrate load provokes hypoglycemia due to increased glucose utilization by insulin-dependent tissues. It can be assumed that due to enhanced utilization of glucose in tissues of women with gynoid obesity, as we have shown previously in the test with food deprivation [1], it becomes not only the dominant substrate of energy metabolism both in daylight and in the dark time of day, but also lipogenesis substrate in the adipose tissue, thus contributing to fat accumulation.

In women with android obesity, OGTT curve in the morning and in the evening little differs from that in women with NBW, but this is achieved via compensatory hyperinsulinemia. Thus, the level of immunoreactive insulin on minute 60 of the morning test was by 4 times higher than that of women with NBW, and in the evening, it was by 2 times higher. The elevated immunoreactive insulin levels in the end of morning and evening tests are indicative of insulin resistance. The fact that in women with android obesity, glucose level in the evening on minute 60 of OGTT was higher and insulin level was lower than in the morning attests to the development of functional exhaustion of the pancreatic insular system [3], which is the risk factor for type 2 diabetes mellitus.

Thus, circadian rhythms of carbohydrate metabolism were impaired in both types of obesity. In case of gynoid obesity, glucose load was followed by hyperinsulinemia and hypoglycemia, while in women with android obesity it led to hyperinsulinemia, insulin resistance, and hyperglycemia irrespective of the time of day, which determines specific features of android and gynoid obesity development.

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