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THE FREQUENCY OF INSULIN RESISTANCE CALCULATED UPON THE BASIS OF A FASTING GLUCOSE TO INSULIN RATIO AND CHARACTERISTICS OF INSULIN RESISTANT WOMEN WITH POLYCYSTIC OVARY SYNDROME

Z. Popovska-Dimova, B. Krstevska

Endocrinology, Diabetes and Metabolic Disorders Clinic,
Medical Faculty, Ss. Cyril and Methodius University, Skopje, R. Macedonia

Abstract: *Background.* The Polycystic Ovary Syndrome (PCOS) is a common endocrine and metabolic disorder of women of reproductive age, characterized by hyperandrogenism and chronic anovulation. Peripheral insulin resistance (IR) has a crucial role in the pathogenesis of this disorder. A fasting glucose to insulin (G/I) ratio is a simple, reliable, sensitive and specific measurement of insulin sensitivity and is a useful test for the identification of IR in women with PCOS, who have to be treated with insulin sensitizers.

Objective. To calculate the frequency of IR among patients with PCOS, using the fasting G/I ratio, and to compare the clinical and biochemical parameters between insulin-resistant and insulin-sensitive groups.

Patients and methods. The study comprised 62 patients of reproductive age (18–40) with PCOS. The diagnosis PCOS was determined according to the diagnostic criteria from Rotterdam 2003, i.e. the presence of two of the following three criteria: oligo-ovulation and/or anovulation; clinical and/or biochemical hyperandrogenism; and polycystic ovaries. All other states associated with hyperandrogenism had been previously excluded. An inclusion criterion for the study was the body mass index (BMI) ≥ 25 (kg/m^2). After calculating the fasting glucose(mg/dl) / insulin(μU/ml) ratio, the patients were separated in two groups, i.e. an insulin-resistant group with a fasting G/I ratio of $< 4.5(mg/10^{-4}U)$ and an insulin-sensitive group with a fasting G/I ratio of $\geq 4.5(mg/10^{-4}U)$.

Results. In our study, 58.06% of women with PCOS and BMI ≥ 25 (kg/m^2) were insulin-resistant. There was no significant difference between the two groups in age or

BMI. Sex hormone binding globulin (SHBG) levels were significantly lower in the insulin-resistant group (17.83 ± 8.38 vs. 42.66 ± 27.65 (nmol/l); p = 0.0036) and they had a higher free testosterone index (19.38 ± 8.91 vs. 9.55 ± 3.51 (%); p = 0.001) in comparison with the insulin-sensitive group. Progesterone levels were significantly lower in the IR group (2.41 ± 2.50 vs. 7.05 ± 8.04 (ng/ml); p = 0.034).

Conclusion. A fasting glucose to insulin ratio is a simple and useful test for identifying insulin-resistant obese women with PCOS. SHBG and progesterone (oligo-ovulation/anovulation) can be useful markers for long-term health risks in women with PCOS (diabetes mellitus type 2 and cardiovascular diseases). In this way we could identify the group of PCOS subjects and its subgroups, which would benefit from therapy with insulin sensitizers, with respect to the reduction of risks of chronic complications.

Key words: Polycystic ovary syndrome (PCOS); insulin resistance (IR); glucose to insulin (G/I); body mass index (BMI); sex hormone binding globulin (SHBG).

Introduction

The Polycystic Ovary Syndrome (PCOS) is a common endocrine and metabolic disorder, characterized by hyperandrogenism and chronic anovulation and affects 6–10% of women of reproductive age [1, 2].

A high percentage of women with PCOS have peripheral insulin resistance [3], this resulting in hyperinsulinemia, and having a crucial role in the pathogenesis of this disorder [2]. The hyperinsulinemia leads to hyperandrogenism [4], mainly through: the stimulation of androgen production and a decrease in SHBG levels (as a result of suppression of hepatic production) with an attendant elevation of the free androgen index.

Women with PCOS have long-term health risks, [5, 6] such as: impaired glucose tolerance, diabetes mellitus type 2, hypertension and cardiovascular diseases. General health risks for women with PCOS are most probably affected by the level of their IR.

Abnormalities in the insulin action are poorly detected by a measurement of either insulin or glucose levels. There are a lot of complicated dynamic tests for the evaluation of insulin sensitivity and adequate detection of insulin resistance. The fasting G/I ratio is a simple, reliable measurement of the insulin sensitivity in obese, white women with PCOS, which is in good correlation with the other the more complicated tests for IR [7], and it has a sensitivity of 95% and a specificity of 84%. It is a useful test for the identification of women with PCOS with IR [7], who need therapeutic treatment by means of insulin sensitizers (biguanides and thiazolidinediones), which decrease the insulin resistance and reduce circulating insulin levels.

The objective of our study was to calculate the frequency of IR among patients with PCOS, using the fasting G/I ratio, and to compare the clinical and biochemical parameters between the insulin-resistant and the insulin-sensitive groups.

Patients and methods

Patients

The study comprised 62 women with PCOS, aged from 18 to 40. The study was carried out in the Endocrinology, Diabetes and Metabolic Disorders Clinic at the Skopje Medical Faculty.

The diagnosis of PCOS was determined according to the diagnostic criteria from Rotterdam 2003 [6], i.e. the presence of two of the following three criteria: oligo-ovulation and/or anovulation; clinical and/or biochemical signs of hyperandrogenism (biochemical: – increased testosterone or dehydroepiandrosterone (DHEA-S) and clinical: hirsutism, acne or alopecia); and polycystic ovaries – present with 12 or more follicles in each ovary measuring 2–9 mm. in diameter, and/or increased ovarian volume (> 10 ml). The other states associated with hyperandrogenism, such as non-classic adrenal hyperplasia (21-hydroxilase-deficient), Cushing's Syndrome, androgen-secreting tumours, thyroid diseases and diseases of hypophysis had been previously excluded.

An inclusion criterion for the study was the body mass index (BMI) of ≥ 25 (kg/m^2). After calculating the fasting glucose(mg/dl) / insulin(μU/ml) ratio, the patients were separated into two groups, i.e. an insulin resistant group with a fasting G/I ratio of $< 4.5(mg/10^{-4}U)$ and an insulin-sensitive group with a fasting G/I ratio of ≥ 4.5 ($mg/10^{-4}U$).

Patients with diabetes mellitus, liver and kidney diseases had been excluded, as well as those taking oral contraceptive pills, less than three months before starting the study.

Study protocol

History; physical examination; Ferriman-Gallway score for hirsutism; anthropology measurements (body weight, body height, calculating BMI, waist circumference (WC)); ultrasonograph examination of ovaries; hormonal analyses (LH, FSH, estradiol, progesterone, testosterone, DHEA-S, SHBG and free testosterone index (FTI)) and measurement of fasting glucose and insulin.

All parameters were performed in the follicular phase of the menstrual cycle, between the second and the ninth day, and progesterone on the 21st day of the cycle. For patients with amenorrhoea for more then two months, all para-

meters were performed each time. For all examinations, venous blood was taken between 08.00 a.m. and 10.00 p.m. Blood serum was used for all analyses except for glucose measurement – blood plasma.

Biochemical methods of analysis

FSH, LH, estradiol, testosterone and DHEA-S were performed by the ECLIA method using a ROCHE ELECSYS 1010 automatic analyzer, insulin by the MEIA method (electrochemiluminiscence immunoassay) using an IMX ABBOTT semiautomatic analyzer, SHBG by the ELISA method (enzyme-linked immunosorbent assay) using an IMX ABBOTT semiautomatic analyzer, glucose with the Glukose-B reagent usinf the glucose-oxidase method with a BECMAN glucose analyzer. Progesterone was performed by the EIA method (enzyme immunoassay), with an automatic analyzer.

Data analysis

We calculated the percentage of insulin-resistant women from the total number of women with PCOS. Further statistical data-processing was performed using the T-test for independent samples, using the Statistics for Windows programme.

Results

Of the total number of 62 women with PCOS, 36 had a fasting G/I ratio $< 4.5(mg/10^{-4}U)$, i.e. 58.06% were insulin-resistant. Then the patients were divided in two groups: an insulin-resistant (IR) group (total 36) and an insulin-sensitive (IS) group (total 26).

The clinical and biochemical features of both groups are summarized in Table 1.

There were no significant differences between the two groups in age, BMI or waist circumference (WC). There were no significant differences in total testosterone levels, but SHBG levels were significantly lower in the insulin-resistant group (17.83 ± 8.38 vs. 42.66 ± 27.65 (nmol/l); $p = 0.0036$) and adequately free testosterone index levels were significantly higher (19.38 ± 8.91 vs. 9.55 ± 3.51(%); $p = 0.001$) in comparison with the insulin sensitive group. Progesterone levels were significantly lower in the IR group (2.41 ± 2.50 vs. 7.05 ± 8.04 (ng/ml); $p = 0.034$).There were no significant differences in FSH, LH, Estradiol and DHEA-S levels.

The fasting insulin levels were significantly higher in the insulin-resistant group (26.26 ± 7.94 vs. 12.72 ± 4.08 (μU/ml); $p < 0.000001$) and fasting glucose levels were significantly higher (4.87 ± 0.63 vs. 4.43 ± 0.65 (mmol/l); $p < 0.01$). Fasting G/I ratios were significantly lower in the IR group than in the IS group (3.54 ± 0.81 vs. 6.92 ± 2.58 (mg/10^{-4}U); $p < 0.000001$).

Table 1 – Табела 1

*Clinical and biochemical features of IR and IS groups**
*Клинички и биохемиски мерења кај ИР и ИС група**

	IR		IS		T-test
parameter	**mean**	**± SD**	**mean**	**± SD**	**p**
age	26.26	6.21	24.85	5.13	NS
BMI (kg/m^2)	33.65	4.78	32.09	3.97	NS
WC (sm)	98.5	9.28	92.29	11.06	NS
FSH(mU/ml)	5.25	1.92	5.91	2.47	NS
LH(mU/ml)	7.8	5.83	8.51	4.73	NS
E2(pg/ml)	62.77	53.94	53.86	39.37	NS
Testosterone (nmol/l)	3.09	1	2.73	0.98	NS
SHBG(nmol/l)	17.83	8.38	42.66	27.65	**p = 0.0036**
FTI (%)	19.38	8.9	9.55	3.51	**p = 0.001**
DHEA-S(μmol/l)	9.78	4.8	8.44	3.63	NS
Progesterone (ng/ml)	2.41	2.5	7.05	8.04	**p = 0.034**
Fasting Insulin (μU/ml)	26.26	7.94	12.72	4.08	**p < 0.000001**
Fasting Glucose (mmol/l)	4.87	0.63	4.43	0.65	**p < 0.01**
Fasting G/I ratio (mg/10^{-4}U)	3.54	0.81	6.92	2.58	**p < 0.000001**

**NS – not significant; НС – нема сигнификантност.*

Discussion

Using the fasting G/I ratios, our results indicate that 58.06 % of women with PCOS and BMI ≥ 25 (kg/m^2), are insulin-resistant. Lego *et al*. [7] declared 53 % insulin-resistant among obese, white women with PCOS. Fasting hyperinsulinemia can also be used as a marker for IR, which is secondary to the profoundly insulin-resistant. But we chose the fasting G/I ratio primarily because of its high sensitivity and specificity, and the possibility of application in everyday clinical work.

SHBG levels in our study were significantly lower in the insulin-resistant group in comparison with the insulin-sensitive group, with a correspondingly higher free testosterone index. Reaven [8], comparing women with and without PCOS, points out the fact that SHBG is lower in PCOS subjects, witha future lowering among the subjects with IR. The hepatic production of SHBG is suppressed by hyperinsulinemia, this connecting it with the lower levels in the IR group. According to Jayagopal *et al*. [9], SHBG is a marker for insulin resistance, which could be used for the identification of insulin-resistant individuals.

Women with PCOS have long-term health risks. Insulin resistance has a central role in these disorders. While the risk of cardiovascular diseases is in the fifth decade of their life (> 45 years), 40% have impaired glucose tolerance, and 10% have diabetes mellitus type 2 in the fourth decade of their life.

Gambineri A. *et al*. [10] found in their study that SHBG is lower in obese women with PCOS and impaired glucose tolerance or diabetes mellitus type 2, compared with that in obese PCOS subjects with normal carbohydrate tolerance. According to Sutton-Tyrrel *et al*. [11], the low SHBG and high FTI are related to a higher risk of cardiovascular disorders. Ekrem *et al*. [12] concluded that diabetes mellitus type 2 is more frequently in patients with PCOS than in the general population of reproductive age and that in subjects with diabetes mellitus type 2, SHBG is lower than in those without diabetes.

If we estimate ovulation at the progesterone levels on the 21st day of the menstrual cycle, it is significantly lower in the insulin-resistant group, thus resulting in a more frequent presence of oligo-ovulation/anovulation (whose clinical presentation is oligomenorrhoea/amenorrhoea) in the group of PCOS women with IR. Solomon et al. [13, 14], in two separate studies, concluded that the women with an irregular menstrual cycle have a significantly higher risk of getting diabetes mellitus type 2 and cardiovascular diseases. They did not evaluate progesterone levels in their studies, and the conclusions were reached on the basis of the duration of the menstrual cycle.

The insulin sensitizer agents [15] biguanides and thiazolidinediones increase peripheral insulin sensitivity, thus decreasing the degree of insulin resistance and circulating insulin levels. The biguanid metformin is the most often

used insulin-sensitizer in the treatment of PCOS. Through the reduction of hyperinsulinemia it causes a significant increase in circulating SHBG levels, thus leading to a decrease of free androgens, stimulating ovulation and establishing a normal menstrual cycle. In this way it reduces risks of developing diabetes mellitus type 2 and cardiovascular diseases.

Conclusion

A fasting glucose to insulin ratio is a simple, reliable, sensitive and specific measurement of insulin sensitivity in obese women and is a useful test for identification of IR in women with PCOS in daily clinical work. SHBG and progesterone (oligo-ovulation/anovulation) can be useful markers for long-term health risks in women with PCOS (diabetes mellitus type 2 and cardiovascular diseases). Using the fasting G/I ratio, we can identify the group of PCOS subjects and, with measurement of SHBG and progesterone, its subgroups who would benefit from therapy with insulin sensitizers, with respect to the reduction of the risks of chronic complications.

REFERENCES

1. Franks S. (1995): Polycystic ovary syndrome. *N Engl J Med*.; 333: 853–861.

2. Bloomgarden Z. T. (2004): Definition of insulin resistance syndrome. The 1st World congress on the insulin resistance syndrome. *Diabetes Care;* 27: 824–830.

3. Dunaif A., (2001): Thomas A. Current concept in the polycystic ovary syndrome. *Annu Rev Med.;* 52: 401–419.

4. Poretsky L., Cataldo N. A., Rosenwaks Z., Giudice L. C. (1999): The insulin-related ovarian regulatory system in health and disease. *Endocr Rev.;* 20: 235–582.

5. Ehrmann D. A. (2005): Polycystic ovary syndrome. *N Engl J Med.;* 352: 1223–1236.

6. The Rotterdam ESHRE/ASRM-sponsored PCOS consensus workshop group. Revised 2003 consensus on diagnostic criteria and long term risks related to polycystic ovary syndrome (PCOS). *Hum Reprod.;* 2004: 19: 41–47.

7. Legro R. S., Finegood D., Dunaif A. (1998): A fasting glucose to insulin is a useful measure of insulin sensitivity in women with polycystic ovary syndrome. *J Clin Endocrinol Metab.;* 83: 2694–2698.

8. Bloomgarden Z. T. (2005): Second World congress on the insulin resistance syndrome. Insulin resistance syndrome and nonalcoholic fatty liver disease. *Diabetes Care;* 28: 824–830.

9. Jayagoral V., Kilpatrick E. S., Jennings P. E., Hepburn D. A., Atkin S. L. (2003): The biological variation of testosterone and sex hormone-binding globulin in

polycystic ovary syndrome: implications for SHBG as a surrogate marker of insulin resistance. *J Clin Endocrinol Metab.;* 88: 1528–1533.

10. Gambineri A., Pelusi C., Manicardi M. *et al*. (2004): Glucose intolerance in a large cohort of Mediterranean women with polycystic ovary syndrome. *Diabetes;* 53: 2353–2358.

11. Sutton-Tyrrel K., Wildman R. P., Matthews KA *et al*. (2005): Sex hormone binding-globulin and the free androgen index are related to cardiovascular risk factors in multiethnic premenopausal and perimenopausal women enrolled in the study of women across the nation (SWAN). *Circulation;* 111: 1242–1249.

12. Tok E. C., Ertunc D., Evruke C., Dilek S. (2004): The androgenic profile of women with non-insulin-dependent diabetes mellitus. *J Reprod Med.;* 49: 746–752.

13. Solomon C. G., Hu F. B., Dunaif A. *et. al.* (2001): Long or highly irregular menstrual cycles as a marker for risk of type 2 diabetes mellitus. *JAMA,* 286: 2421–242.

14. Solomon C. G., Hu F. B., Dunaif A. *et.al*. (2002): Menstrual cycle irregularity and risk for future cardiovascular disease. *J Clin Endocrinol Metab.;* 87: 2013–2017.

15. De Leo V., Marca A., Petraglia F. (2003): Insulin lowering agents in the management of polycystic ovary syndrome. *Endocr Rev.;* 24: 633–667.

Резиме

ФРЕКВЕНЦИЈАТА НА ИНСУЛИНСКАТА РЕЗИСТЕНЦИЈА ПРЕСМЕТАНА ПРЕКУ ОДНОСОТ ГЛИКОЗА/ИНСУЛИН НА ГЛАДНО И КАРАКТЕРИСТИКИТЕ НА ИНСУЛИН РЕЗИСТЕНТНИТЕ ЖЕНИ СО ПОЛИЦИСТИЧЕН ОВАРИЈАЛЕН СИНДРОМ

З. Поповска-Димова, Б. Крстевска

Клиника за ендокринологија и болести на метаболизмот, Медицински факултет, Универзитет „Св. Кирил и Методиј“, Скопје, Р. Македонија

Полицистичен оваријален синдром е најчесто ендокринолошко нарушување кај жените во репродуктивна возраст, кое се карактеризира со хиперандрогенемија и хронична ановулација. Периферната инсулинска резистенција има круцијална улога во патогенезата на ова нарушување. Гликоза/инсулин односот на гладно е едноставно, сигурно, сензитивно и специфично мерење на инсулинската сензитивност и е корисен тест за идентификирање на ИР кај обезни жени со ПЦОС, кои треба да бидат третирани со инсулински сензитајзери.

Нашата студија беше дизајнирана да се пресмета фреквенцијата на ИР помеѓу пациентките со ПЦОС, со користење на односот гликоза/инсу-

лин на гладно и да се компарираат клиничките и биохемиските параметри, помеѓу инсулин резистентната и исулин сензитивната група. Во студијата беа вклучени 62 пациентки со ПЦОС. Дијагнозата ПЦОС е поставена според дијагностичките критериуми од Rotterdam 2003, и тоа со присуство на два од следните три критериуми: олиго и/или ановулација; клинички и/или лабораториски знаци за хиперандрогенизам; полицистични овариуми. Критериум за вклучување во студијата беше ИТМ $\geq 25(kg/m^2)$. По пресметување на индексот гликоза(mg/dl) / инсулин(μU/ml), пациентките се поделени на две групи, и тоа: инсулин резистентна група со Г/И однос на гладно < $4.5(mg/10^{-4}U)$ и инсулин сензитивна група со Г/И, однос $\geq 4.5(mg/10^{-4}U)$.

Нашите резултати покажаа дека 58,06% од жените со ПЦОС и ИТМ $\geq 25(kg/m^2)$, користејќи го односот гликоза/инсулин, се инсулин резистентни. Не постоеше сигнификантна разлика помеѓу двете групи по однос на возраста и ИТМ. СХВГ беше сигнификантно понизок кај инсулин резистентната група и соодветно индексот на слободен тестостерон беше сигнификантно повисок, во однос на инсулин сензитивната група. Прогестеронот беше сигнификантно понизок во ИР групата, што сугерира дека во групата на ПЦОС жени со ИР олиго/ановулација (која клинички се презентира со олиго/аменореја) е почесто застапена.

Односот гликоза/инсулин на гладно е едноставен и корисен тест за идентификација на инсулин резистентните обезни жени со ПЦОС, во секојдневната клиничка работа. Субјектите со низок СБВГ и низок прогестерон имаат зголемен ризик од развој на дијабетес мелитус тип 2 и кардиоваскуларни заболувања. Следува дека СХБГ и прогестеронот (олиго/ановулацијата) можат да бидат корисни маркери за долгорочните ризици по здравјето на жените со ПЦОС.

Со користење на Г/И индексот би се идентифицирала групата на ПЦОС субјекти, а со одредување на СХВГ и прогестеронот и подгрупите, кои би имале бенефит од терапијата со инсулински сензитајзери, пред сè во однос на намалување на ризиците за развивање на хронични компликации.

Клучни зборови: Полицистичен оваријален синдром (ПЦОС); инсулинска резистенција (ИР); гликоза/инсулин (Г/И); индекс на телесна маса (ИТМ); секс хормон врзувачки глобулин (СХВГ).

Corresponding Author:

Z. Popovska-Dimova,
Endocrinology, Diabetes and Metabolic Disorders Clinic,
Medical Faculty,
Ss. Cyril and Methodius University,
Skopje, R. Macedonia