

## CLINICAL STUDY

## Predictors of abnormal glucose metabolism in women with polycystic ovary syndrome

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### Abstract

**Objective:** Polycystic ovary syndrome (PCOS) is a risk factor for type 2 diabetes mellitus and screening for abnormal glucose metabolism has been recommended by an oral glucose tolerance test (OGTT). This procedure is time-consuming and inconvenient, limiting its general use. Therefore, an easy method is wanted to separate PCOS women with normal from those with potentially abnormal glucose metabolism.

**Design:** Simple parameters obtained from 101 consecutive PCOS patients were assessed by receiver operating curve analysis for their ability to predict abnormal glucose metabolism.

**Results:** Comparing discriminating parameters at defined sensitivities revealed that, assessed by homeostasis model assessment (HOMA), insulin resistance (HOMA%S) had the highest specificity. At a cut-off point of 73.1%, HOMA%S had a sensitivity of 95.5% and a specificity of 51.9%. Applying this cut-off separated 59 women who had a high probability of abnormal glucose metabolism from 42 women who were at low risk (less than 2.5%). Fasting insulin was the second-best parameter and had a similar specificity. A screening strategy which applies HOMA%S or fasting insulin could almost halve the number of OGTTs by directing them to those PCOS women most likely to be suffering from abnormal glucose metabolism. The negative predictive value of this strategy was 97%. The strategy was tested and confirmed in a second and independent cohort of 264 PCOS women.

**Conclusions:** HOMA%S, or to a lesser extent fasting insulin, appears to allow for stratified metabolic screening of PCOS women with OGTT.

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### Introduction

Polycystic ovary syndrome (PCOS) is one of the most frequent endocrine diseases, affecting about 5–10% of reproductive women (1). PCOS is a heterogeneous disorder characterized by hyperandrogenism, chronic anovulation and infertility (2–5). A significant proportion of PCOS women suffer from insulin resistance, which appears to play a role in the etiology of PCOS, since amelioration of insulin resistance by lifestyle or pharmacological intervention has been shown to improve hyperandrogenism and fertility (6–9).

Insulin resistance is recognized as a major risk factor for the development of type 2 diabetes mellitus (T2DM) and subsequent cardiovascular disease (10–13). Several studies have shown that approximately 20–40%

of adult and adolescent PCOS women, including both lean and obese patients, suffer from abnormal glucose metabolism, i.e. either impaired glucose tolerance (IGT) or T2DM (14–17). Accordingly, PCOS is regarded as a risk factor for the development of T2DM and general screening of PCOS patients for IGT and/or T2DM has been recommended (13, 17–19). Screening appears to be warranted as early lifestyle intervention may prevent the development of T2DM in patients with IGT on the one hand (20, 21) and on the other hand early diagnosis and treatment of T2DM may reduce the burden of diabetes and its complications (19, 22, 23).

As PCOS is common and metabolic screening will be an ongoing process with retesting of individuals at regular intervals, the number of tests that need to be

performed is high. Fasting glucose has been suggested as a screening parameter for abnormal glucose metabolism. However, several studies demonstrate that a substantial proportion of PCOS women with IGT or even T2DM show normal fasting glucose concentrations (14, 16, 17). This indicates that fasting glucose is too insensitive to be used as a screening parameter for abnormal glucose metabolism in PCOS. Instead, it has been suggested that PCOS patients should be screened for abnormal glucose metabolism with an oral glucose tolerance test (OGTT) using the 2 h glucose concentration after a 75 g glucose challenge (13). This procedure, however, is relatively time-consuming, which may limit its use as a general screening instrument in PCOS women. Therefore, a rapid and simple method would be desirable to separate PCOS women with normal from those with potentially abnormal glucose metabolism. Thereby, the need to perform an OGTT could be reduced by reserving OGTTs for those persons who are most likely to be suffering from abnormal glucose metabolism.

In the present study we prospectively evaluated 101 consecutive PCOS women for the presence of abnormal glucose metabolism and we investigated the ability of simple parameters obtained from fasting blood and indices calculated from these parameters to predict normal or abnormal glucose metabolism.

## Subjects and methods

### Subjects

We prospectively studied 101 consecutive women with PCOS, who were referred to our clinic because of hirsutism, oligo-/amenorrhea, or infertility. The patients were included after written informed consent was obtained. The cohort has been published in part previously (24–26). The diagnosis of PCOS was based on (i) the presence of chronic ovulatory dysfunction, i.e. oligomenorrhea (four or fewer cycles in the last 6 months) or amenorrhea (no cycles in the last 6 months), and (ii) clinical signs of hyperandrogenism, i.e. hirsutism or acne, or (iii) laboratory findings, i.e. hyperandrogenemia, defined as serum androgen levels (dehydroepiandrosterone sulfate (DHEAS), 17-OH-progesterone, androstenedione or total testosterone) above the upper limit of normal for the respective assay, and (iv) the exclusion of other disorders such as Cushing's syndrome, late-onset 21-hydroxylase deficiency, thyroid dysfunction, hyperprolactinemia, or androgen-secreting tumors. These diagnostic criteria are consistent with those most commonly used for PCOS, often referred to as the NIH consensus criteria (27). The clinical and endocrine features of the women are given in Table 1. Twelve patients suffered from Hashimoto's thyroiditis. They were euthyroid under thyroid hormone replacement therapy. Three women suffered from arterial hypertension and were under antihypertensive

**Table 1** Clinical and endocrine features of the PCOS cohort ( $n = 101$ ).

Characteristic	Mean $\pm$ S.E.M
Age (years)	28.7 $\pm$ 0.55
BMI (kg/m <sup>2</sup> )	32.0 $\pm$ 0.82
Fasting plasma glucose (mmol/l)	4.6 $\pm$ 0.06
Fasting insulin (pmol/l)	102.1 $\pm$ 6.80
HOMA%S	75.4 $\pm$ 4.63
HOMA-IR	3.09 $\pm$ 0.22
HOMA%B	173.8 $\pm$ 8.16
HOMA-B	318.1 $\pm$ 54.0
Testosterone (nmol/l)	3.37 $\pm$ 0.14
Estradiol (pmol/l)	245.8 $\pm$ 29.22
Progesterone (nmol/l)	4.88 $\pm$ 1.08
LH (U/l)	8.7 $\pm$ 0.54
LH/FSH	1.86 $\pm$ 0.2
DHEAS ( $\mu$ mol/l)	7.53 $\pm$ 0.38
Androstenedione (nmol/l)	8.26 $\pm$ 0.3
SHBG (nmol/l)	52.5 $\pm$ 4.78
17-OH-progesterone (nmol/l)	2.46 $\pm$ 0.15
Overweight or obese subjects (BMI > 25 kg/m <sup>2</sup> )	77 (76.2%)

medication. All other patients were free of other diseases and were taking no medications.

All women were studied within the first 10 days following menstruation in the case of mild oligomenorrhea, or at random if they suffered severe oligo- or amenorrhea. Blood was sampled in the morning after an overnight fast. Glucose was measured immediately, and blood samples were stored at  $-20^{\circ}\text{C}$  until analysis of the other parameters. All women had normal fasting glucose levels ( $<6.1$  mmol/l). A 2 h glucose value during OGTT (75 g) of  $>7.8$  mmol/l was considered as IGT and of  $>11.1$  mmol/l as T2DM according to the WHO definition (28). Insulin resistance (HOMA%S) and  $\beta$ -cell function (HOMA%B) were assessed by homeostasis model assessment (HOMA) using the mean of three fasting glucose and insulin levels. The values were calculated using the computer program HOMA-CIGMA version 2 kindly provided by Dr J C Levy (Radcliffe Infirmary, Oxford, UK) (29, 30). In addition, insulin resistance and  $\beta$ -cell function were calculated as HOMA-IR ((fasting insulin (mU/l)  $\times$  fasting glucose (mmol/l))/22.5) or as HOMA-B ((20  $\times$  fasting insulin (mU/l))/(fasting glucose (mmol/l) – 3.5)). Body mass index (BMI) was calculated as body weight (kg) divided by body height squared (m<sup>2</sup>).

A second cohort of 264 PCOS women with fasting glucose levels below 6.1 mmol/l was used principally to test the screening strategy derived from the cohort above. The patients presented between 1998 and 2002 with menstrual disorders and hyperandrogenism to an outpatient clinic for Gynecological Endocrinology in Berlin, Germany. The diagnosis of PCOS was based on the same criteria as described above. As part of the routine work-up all PCOS women were screened for the presence of insulin resistance (fasting insulin and fasting blood glucose) and abnormal glucose metabolism (OGTT). All measurements were performed under

terms of routine clinical practice. The computerized data of 264 women were used for retrospective analysis. The women were  $28.1 \pm 0.44$  years old, their BMI was  $30.4 \pm 0.41$  kg/m<sup>2</sup>, and 213 patients (80.7%) were overweight or obese (BMI > 25 kg/m<sup>2</sup>). Total testosterone amounted to  $1.58 \pm 0.06$  nmol/l, androstendione was  $10.12 \pm 0.39$  nmol/l, and DHEAS  $8.34 \pm 0.22$   $\mu$ mol/l. Fasting glucose was  $5.20 \pm 0.03$  mmol/l, fasting insulin  $138.3 \pm 10.2$  pmol/l, and HOMA%S  $65.1 \pm 2.3\%$ . According to the WHO definition for the OGTT (28), 222 women had normal glucose tolerance (NGT), 38 had IGT and four had T2DM.

### Assays

Blood glucose was measured by the glucose dehydrogenase method on a Cobas Mira Laboratory System (Roche, Mannheim, Germany). Plasma insulin was measured by a commercial RIA (Pharmacia, Freiburg, Germany) with a lower limit of sensitivity of < 14 pmol/l and an intra- and interassay coefficient of variation (CV) values of 5.8 and 5.8%. Proinsulin was measured by a two-site IRMA, using a monoclonal proinsulin antibody (Clone 7F8, mouse anti-human proinsulin; Biotrend, Cologne, Germany) as the catcher antibody, and a radioiodinated monoclonal C-peptide antibody (clone PEP-001, mouse anti-human C-peptide; Novo-Nordisk, Bagsvaerd, Denmark) for detection. Proinsulin serum concentrations were calculated using recombinant human proinsulin (Sigma, Munich, Germany) as the standard. The intra- and interassay CV values were < 5% and < 8%, and the lower limit of sensitivity was < 1 pmol/l. There was no crossreactivity with insulin (up to 1650 pmol/l) or with C-peptide (up to 9.9 nmol/l). Serum 17-OH-progesterone was determined by RIA (Schering, Berlin, Germany) with a lower level of sensitivity of 0.06 nmol/l and intra- and interassay CV values of 3.2 and 3.9%. Serum luteinizing hormone (LH), and progesterone were measured by chemiluminescence immunoassays using kits obtained from Bayer Diagnostics (Fernwald, Germany). The intra- and interassay CV values were 2.9 and 2.4% for LH and 3.7 and 3.9% for progesterone, and the minimal detectable concentrations were 0.07 mU/ml (LH) and 0.48 nmol/l (progesterone). DHEAS was assayed by a chemiluminescence immunoassay purchased from Nichols Institute Diagnostics (Bad Nauheim, Germany) with a lower sensitivity of 0.027  $\mu$ mol/l and intra- and interassay CV values of 7.1 and 9.0% respectively. Serum follicle-stimulating hormone (FSH) was determined by an IRMA (BioChem ImmunoSystems, Freiburg, Germany) with a lower sensitivity of 0.3 mU/ml and inter- and intraassay CV values of 2 and 3.1% respectively. Estradiol, testosterone, and sex hormone-binding globulin (SHBG) were measured by RIA obtained from DSL (Sinsheim, Germany) with lower detectable concentrations of 8.1 pmol/l, 0.28 nmol/l and 5 nmol/l respectively. The respective inter- and intraassay CV values

were 2.2 and 2.7% for estradiol, 9.6 and 8.6% for testosterone, and 2.2 and 4.4% for SHBG. Androstenedione was determined by RIA (Coulter Immunotech, Marseille, France) with a lower sensitivity of 0.06 nmol/l and inter- and intraassay CV values of 5.6 and 6%. The upper limits of normal were 3.4 nmol/l, 12.0  $\mu$ mol/l and 10.5 nmol/l for testosterone, DHEAS and androstenedione respectively.

In the second cohort, glucose was measured with the Synchron CX Delta-System (Beckman-Coulter, Krefeld, Germany), insulin by RIA (BioChem ImmunoSystems) with a lower limit of sensitivity of 7 pmol/l and intra- and interassay CV values of 6.8 and 6.8%, testosterone with a chemiluminescence immunoassay purchased from Bayer Diagnostics with a lower sensitivity of 0.35 nmol/l and intra- and interassay CV values of 2.9 and 4.3%, androstendione by RIA (DSL) with a lower sensitivity of 0.07 nmol/l and intra- and interassay CV values of 4.3 and 5.9%, and DHEAS by a chemiluminescence immunoassay (Immulite; DPC Biermann, Bad Nauheim, Germany) with a lower sensitivity of 0.41  $\mu$ mol/l and intra- and interassay CV values of 8.2 and 11.6%. The upper limits of normal were 1.75 nmol/l, 8.1  $\mu$ mol/l and 9.8 nmol/l for testosterone, DHEAS and androstenedione respectively.

### Statistics

Statistical analyses were performed with SPSS software (version 10.0; SPSS Inc., Chicago, IL, USA). Mean values are reported  $\pm$  s.e.m. if not stated otherwise. Significance was considered at two-tailed  $\alpha < 0.05$ . The nonparametric Mann-Whitney U test was used to analyze for differences in skewed continuous variables, while differences in normally distributed continuous variables were compared by unpaired Student's *t*-test. Normal distribution was tested by the Kolmogorov-Smirnov test. Differences in frequencies were tested by the chi-square test. The area under the receiver operating characteristic (ROC) curve (AUC) was used to define variables discriminating between normal and abnormal glucose metabolism (IGT and T2DM). The 95% confidence interval (95% CI) for the ROC AUC was used to test the hypothesis that the theoretical area is 0.5. If the CI did not include the 0.5 value, the laboratory test was considered to have an ability to distinguish between PCOS women with and without impaired glucose metabolism. The optimal cut-off point for each variable was selected by calculating Youden's index ( $J = \text{sensitivity} + \text{specificity} - 1$ ). The ROC curves were tested for significant differences by the method of Hanley & McNeil (31) using MedCalc Software (MedCalc, Mariakerke, Belgium). MedCalc Software was used to calculate specificities, positive predictive values and negative predictive values (NPVs) at defined sensitivities of 95.5 and 90.9%.

## Results

The clinical and endocrine characteristics of the cohort are depicted in Table 1. Seventy-six (75.2%) of the PCOS patients were overweight or obese. All 101 subjects had normal fasting glucose levels (<6.1 mmol/l). Twenty-two (21.8%) subjects had an abnormal 2 h glucose concentration (>7.8 mmol/l) in the OGTT, 18 with IGT (17.8%) and four with T2DM (4%). The women with abnormal glucose metabolism differed from those with NGT in that they were more obese, were more insulin-resistant (higher fasting insulin levels, lower HOMA%S, higher HOMA-IR), had higher testosterone, and lower SHBG levels, while there were no differences in fasting glucose or the family history of diabetes (Table 2).

In our cohort, 78.2% of the OGTTs performed showed NGT. This means that a large number of OGTTs performed for screening for abnormal glucose metabolism in PCOS women could be saved, if there were fasting values that could direct an OGTT to those women most likely to be suffering from impaired glucose metabolism. All women had normal fasting glucose levels. Therefore, the 22 women with either IGT or T2DM would have been missed without performing an OGTT.

We examined whether other parameters could be useful predictors for the state of glucose metabolism in PCOS women with normal fasting glucose. ROC curves were used to analyze the characteristics of various continuous parameters and indices and their suitability as potential screening tests. The ROC AUC was calculated as a quantitative measure of their comparative test utility. The ROC curve of a discriminating

parameter should be different from the bisecting line, which means that the AUC needs to be significantly different from 0.5 (Table 3). The AUC for age, fasting glucose and  $\beta$ -cell function assessed either by the computer program (HOMA%B) or by the approximative formula (HOMA-B) were not significantly different from 0.5 (Table 3). The ROC curves for all the other parameters shown in Table 3, however, were significantly different from the bisecting line, but the AUCs for these parameters were not significantly different from each other. For these parameters cut-off points were calculated by Youden's index and are listed in Table 3 together with the respective sensitivities and specificities, which varied over a wide range.

To further evaluate the individual parameters we compared them at given sensitivities that are in the range of typical screening tests (32). HOMA%S had the highest specificity at a sensitivity of both 90.9 and 95.5% as shown in Table 4. Applying the 95.5% cut-off point of HOMA%S at 73.1% to the metabolic screening of PCOS women allowed separation of a subgroup of 59 PCOS women (58% of the entire cohort) who probably suffered from abnormal glucose metabolism from a group of 42 women (42% of the entire cohort) who were at low risk for abnormal glucose metabolism (less than 2.5%). Accepting a lower sensitivity (90.9%) at a cut-off point of 71.3% for HOMA%S increased the low-risk subgroup to 46 PCOS women (46% of the cohort) at the expense of a slightly higher risk for abnormal glucose metabolism (less than 5%). Fasting insulin, a more simple parameter which does not require any calculations, was the second-best parameter according to Table 4. At a cut-off of 70 pmol/l (sensitivity 95.5%) 38 PCOS

**Table 2** Characteristics of the PCOS women with and without abnormal glucose metabolism (IGT/T2DM). Means  $\pm$  S.E.M.

Characteristic	Normal glucose metabolism (n = 79)	Abnormal glucose metabolism (n = 22)	P-value for difference
Age (years)	28.27 $\pm$ 0.65	30.14 $\pm$ 0.87	0.28
BMI (kg/m <sup>2</sup> )*	31.08 $\pm$ 0.94	35.32 $\pm$ 1.50	0.03
Fasting plasma glucose (mmol/l)*	4.6 $\pm$ 0.07	4.8 $\pm$ 0.13	0.14
Fasting insulin (pmol/l)	93.72 $\pm$ 7.56	132.27 $\pm$ 13.89	0.001
Fasting proinsulin (pmol/l)	11.7 $\pm$ 2.0	22.9 $\pm$ 5.62	<0.001
HOMA%S	82.97 $\pm$ 5.54	48.24 $\pm$ 3.85	0.001
HOMA-IR	2.79 $\pm$ 0.23	4.16 $\pm$ 0.51	0.001
HOMA%B	168.02 $\pm$ 9.38	194.45 $\pm$ 15.93	0.058
HOMA-B	255.7 $\pm$ 46.5	539.6 $\pm$ 177.3	0.12
Testosterone (nmol/l)	3.19 $\pm$ 0.17	3.99 $\pm$ 0.28	0.004
Estradiol (pmol/l)	258.5 $\pm$ 36.8	198.8 $\pm$ 18.7	0.54
Progesterone (nmol/l)	5.07 $\pm$ 1.3	4.15 $\pm$ 1.62	0.83
LH (U/l)*	9.05 $\pm$ 0.63	7.36 $\pm$ 0.92	0.2
LH/FSH	1.94 $\pm$ 0.25	1.57 $\pm$ 0.21	0.64
DHEAS ( $\mu$ mol/l)*	7.23 $\pm$ 0.43	8.62 $\pm$ 0.74	0.13
Androstenedione (nmol/l)*	8.0 $\pm$ 0.32	9.22 $\pm$ 0.74	0.10
SHBG (nmol/l)	58.9 $\pm$ 5.77	28.96 $\pm$ 3.96	0.005
17-OH-progesterone (nmol/l)	2.49 $\pm$ 0.19	2.34 $\pm$ 0.23	0.94
Overweight or obese subjects (BMI > 25 kg/m <sup>2</sup> )	55 (70.5%)	22 (100%)	0.003
Family history of T2DM	32 (40.5%)	12 (54.5%)	0.47

\* Normally distributed variable.

**Table 3** Areas under the ROC curves for different parameters for separating normal and abnormal (IGT/T2DM) glucose metabolism. Sensitivity and specificity to detect IGT/T2DM at the given cut-offs.

Variable	Area under ROC (95%-CI)	P-value (asymptotic significance for area=0.5)	Cut-off (calculated by Youden's index)	Sensitivity (at cut-off)	Specificity (at cut-off)
Age	0.576 (0.454–0.697)	0.28	—	—	—
Fasting plasma glucose	0.608 (0.476–0.741)	0.12	—	—	—
HOMA-B	0.615 (0.500–0.730)	0.10	—	—	—
HOMA%B	0.633 (0.517–0.748)	0.058	—	—	—
BMI (kg/m <sup>2</sup> )	0.652 (0.540–0.763)	0.031	27.25	0.95	0.38
Proinsulin/insulin	0.693 (0.569–0.818)	0.006	0.1115	0.77	0.64
Glucose/insulin (mmol/pmol)	0.704 (0.600–0.809)	0.003	0.064	0.91	0.48
Testosterone (nmol/l)	0.707 (0.595–0.819)	0.004	2.95	0.85	0.54
SHBG (nmol/l)	0.712 (0.597–0.828)	0.005	29.5	0.74	0.69
Fasting insulin (pmol/l)	0.728 (0.629–0.828)	0.001	74.0	0.91	0.54
HOMA%S	0.734 (0.635–0.832)	0.001	73.25	0.95	0.52
HOMA-IR	0.735 (0.637–0.833)	0.001	2.3252	0.86	0.58
Glucose/proinsulin (mmol/pmol)	0.747 (0.636–0.859)	<0.001	0.496	0.82	0.61
Fasting proinsulin (pmol/l)	0.762 (0.657–0.868)	<0.001	10.5	0.77	0.68

women and at a cut-off of 73 pmol/l (sensitivity 90.9%) 45 women were assigned to the low-risk groups. For both parameters all the women with T2DM were correctly assigned to the high-risk group. The NPV of a strategy using the 95.5% sensitivity cut-off points of HOMA%S or insulin was around 97%, which means that only very few women with abnormal glucose metabolism were missed.

The ability of HOMA%S and fasting insulin to discriminate PCOS women with normal from those with abnormal glucose metabolism was tested in a second cohort of 264 PCOS women. These women were

investigated in an outpatient center for endocrinological gynecology and 42 of these women had abnormal glucose metabolism defined by an OGTT. Again, according to the ROC curve analysis, HOMA%S and fasting insulin were suitable for discrimination between normal and abnormal glucose metabolism (AUC for HOMA%S 0.701, *P* < 0.001, AUC for fasting insulin 0.7, *P* < 0.001). Applying the above calculated 95.5% sensitivity cut-off values for HOMA%S (73.1%) and fasting insulin (70 pmol/l) to this second cohort separated 34% or 32% of the women to the low-risk group with an NPV of 93%, indicating that again only very few PCOS women with impaired glucose metabolism are missed. All four women with diabetes were assigned to the high-risk group. Again, HOMA%S was slightly superior.

Thus, using HOMA%S, or to a lesser extent fasting insulin, in the metabolic screening of PCOS women reduces the number of OGTTs by reserving an OGTT to those persons who most probably suffer from abnormal glucose metabolism.

**Table 4** Cut-points and specificities of different parameters at 95.5% and 90.9% sensitivity.

Variable	Cut-point Specificity PPV NPV			
95.5% sensitivity				
HOMA%S	73.1	51.9	35.6	97.6
Fasting insulin (pmol/l)	70	46.8	33.3	97.4
HOMA-IR	2.05	46.8	33.3	97.4
Glucose/insulin (mmol/pmol)	0.069	43.0	31.8	97.1
BMI (kg/m <sup>2</sup> )	27.1	38.5	30.4	96.8
SHBG (nmol/l)	61	35.7	28.6	96.2
Fasting proinsulin (pmol/l)	5	35.4	29.2	96.6
Glucose/proinsulin (mmol/pmol)	0.87	31.6	28.0	96.2
Testosterone (nmol/l)	2.26	30.3	26.6	96.9
Proinsulin/insulin	0.05	11.5	23.3	90.0
90.9% sensitivity				
HOMA%S	71.3	55.7	36.4	95.7
Fasting insulin (pmol/l)	73	54.4	35.7	95.6
HOMA-IR	2.12	48.1	32.8	95.0
Glucose/insulin (mmol/pmol)	0.063	48.1	32.8	95.0
Testosterone (nmol/l)	2.43	39.5	28.1	93.7
Fasting proinsulin (pmol/l)	5.5	38.6	29.2	94.2
BMI (kg/m <sup>2</sup> )	27.4	38.5	29.4	93.8
SHBG (nmol/l)	58	37.1	27.9	92.9
Glucose/proinsulin (mmol/pmol)	0.86	32.9	27.4	92.9
Proinsulin/insulin	0.06	20.5	24.4	88.9

PPV, positive predictive value; NPV, negative predictive value.

### Discussion

In our cohort of 101 consecutive PCOS women with a mean age of just under 29 years almost 22% had either IGT or T2DM. This prevalence rate is similar to the ones reported from cohorts of Mediterranean, North American and Asian PCOS women of comparable age (15, 16, 33), and is significantly higher than that of the general population (34). Screening of PCOS women for abnormal glucose metabolism has been recommended (13, 17, 19). In our group, fasting glucose, the screening parameter suggested by the American Diabetes Association (19), was almost identical in women with and without IGT or T2DM, which is consistent with previous reports (14, 16, 17). Furthermore,

based on the ROC analysis, fasting glucose was no discriminating parameter. Thus, an OGTT appears to be the only way to reliably detect abnormal glucose metabolism in PCOS. Since this procedure is relatively time-consuming and inconvenient to the patient, we investigated whether there is a rapid and simple method that could direct the need to perform an OGTT to those women who are the most likely to be suffering from abnormal glucose metabolism.

PCOS women with abnormal glucose tolerance were on average more obese and insulin-resistant, showed higher insulin, proinsulin and testosterone levels, and had lower SHBG levels than the patients with NGT. Obesity is a well known risk factor for insulin resistance and accounts for about 30% of the degree of insulin resistance in PCOS women (24). The higher androgen and lower SHBG levels may in turn be a result of their more pronounced insulin resistance, since there is good evidence that compensatory hyperinsulinemia amplifies androgen production in PCOS (35) and inhibits SHBG synthesis by the liver (36).

We tested these and other parameters for their ability to distinguish between PCOS women with and without abnormal glucose metabolism. To define the best discriminating parameters between normal and abnormal glucose metabolism ROC curves were calculated and analyzed, which is a method often used to investigate such an issue (37). Age, fasting glucose and the HOMA indices calculated to assess  $\beta$ -cell function were found inappropriate for discrimination. With respect to the AUC under the ROC curve, a variety of other parameters were found significantly different from the bisecting line. Therefore, all these parameters were in principle useful for discrimination, especially since the AUCs for these parameters were not significantly different from each other. At the respective cut-off points calculated by the Youden's index a wide range of sensitivities and specificities for the individual parameters was observed, which made a direct comparison difficult. For that reason we compared the parameters at defined sensitivities, which were in the range of other screening tests like the fecal occult blood test that is recommended for the screening of colorectal carcinoma (32). In this context HOMA%S had the highest specificity. At a cut-off point of 73.1% HOMA%S had a specificity of 51.9% and a sensitivity of 95.5%. Clearly, the specificity of HOMA%S is relatively low. In our opinion, however, this limitation can be tolerated, as the consequence of a positive screening result is an OGTT, which otherwise would have been performed in any case, and which is a safe test.

Applying a cut-off of 73.1% HOMA%S to the metabolic screening of PCOS women allowed us to classify a high-risk (58% of the cohort) and a low-risk group (42% of the cohort) for the presence of abnormal glucose metabolism. HOMA%S needs to be calculated from fasting glucose and insulin. This appears to be feasible in daily practice by using freely available computer software. Fasting insulin, a more simple

parameter, however, which does not require any calculations, was the second-best parameter and might be sufficiently accurate for risk stratification in routine clinical settings. A screening strategy which applies HOMA%S or fasting insulin could therefore markedly reduce the numbers of OGTTs needed for the screening of abnormal glucose metabolism in PCOS women. The NPV of this strategy is high (97%), indicating that only very few women with abnormal glucose metabolism are missed. The ability of HOMA%S and fasting insulin to separate PCOS women with low risk from those with high risk for abnormal glucose metabolism could, in principle, be confirmed in a second large cohort of PCOS women.

Thus, HOMA%S or fasting insulin appear to allow for stratified metabolic screening of PCOS women with OGTT. The cut-off points for both HOMA%S and fasting insulin are expected to vary depending on the insulin assay used and therefore assay-specific adjustment might be needed (38). Future testing in other cohorts will show whether such a screening procedure is useful and beneficial in daily clinical practice.

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