METFORMIN EFFECTS ON CLOMIFENE-INDUCED OVULATION IN THE POLYCYSTIC OVARY SYNDROME

Ben Ayed B.*, Dammak dit Mlik S. **, Ben Arab H*., Trabelssi H.*, Chahtour H.*, Mathlouthi N.*, Dhuib M.*, Kassis M. ***, Saiidane D. **, Trabelssi K .*,Guermazi M.*

* *Department of Obstetrics & Gynaecology, CHU Hedi Chaker-Sfax. - ** Animal physiology and cellular biology laboratory, faculty of Pharmacy- Monastir. *** Preventive medicine laboratory, faculty of Medicine- Sfax.*

Ben Ayed B., Dammak dit Mlik S., Ben Arab H., Trabelssi H., Chahtour H., Mathlouthi N., Dhuib M., Kassis M., Saiidane D., Trabelssi K.,Guermazi M.

APPORT DE LA METFORMINE LORS DE LA STIMULATION OVARIENNE DANS LE SYNDROME DES OVAIRES POLYKYSTIQUES.

LA TUNISIE MEDICALE - 2009 ; Vol 87 (n°01) : 43 - 48

Ben Ayed B., Dammak dit Mlik S., Ben Arab H., Trabelssi H., Chahtour H., Mathlouthi N., Dhuib M., Kassis M., Saiidane D., Trabelssi K.,Guermazi M.

METFORMIN EFFECTS ON CLOMIFENE-INDUCED OVULATION IN THE POLYCYSTIC OVARY SYNDROME

LA TUNISIE MEDICALE - 2009 ; Vol 87 (n°01) : 43 - 48

RÉSUMÉ

Introduction : Le syndrome des ovaires polykystiques (SOPK), encore appelé l'hyperandrogénie ovarienne fonctionnelle ou l'anovulation hyperandrogénique, est l'un des plus complexe et fréquent désordre endocrinologique rencontré chez la femme en âge de procréation. Diverses anomalies font le motif de consultation pour les femmes en atteintes. La dys-anovulation, donnant généralement une infertilité, figure en tête d'affiche et semble être aggravée par l'insulinorésistance caractérisant le syndrome métabolique associée fréquemment au SOPK.
L'atténuation de cette insulinorésistance, par des drogues insulinsensibilisantes tel la metformine, fut l'objet de plusieurs études récentes mettant au point particulièrement l'apport de ces drogues lors de la stimulation ovarienne par le citrate de clomifène chez des femmes à ovaires polykystiques.
But:L'objectif de notre étude prospective est de comparer l'efficacité de l'association de la metformine au citrate de clomifene par apport à l'utilisation de ce dernier seul chez des femmes atteintes du SOPK.
Méthodes: Du 24 février jusqu'au 29 septembre 2007, on s'est mis à la sélection des femmes atteintes du SOPK parmi ceux se présentant à la consultation de stérilité (service de gynéco obstétrique du CHU Hedi Chaker de Sfax), et ce, selon les critères du diagnostic de Rotterdam 2003. Les femmes en atteintes ont été réparties aléatoirement en deux groupes pour lesquels on a prescrit le citrate de clomifène associé au placebo (groupe «CC+placebo») ou à la metformine,850mg 2 fois par jours pendant tout le cycle ovulatoire (groupe«CC+metformine»). Le suivi de ces patients était fait par un monitorage échographique et biologique (dosage d'oestradiol E2) les jours 7, 11 et 13 du cycle.
Résultats : Au bout des 7 mois d'étude, 32 femmes sont présumées atteintes du SOPK. Les deux groupes en formés, de 16 patientes chacun, s'avèrent comparables au niveau des différentes caractéristiques (âge ; paramètres anthropométriques ; données cliniques, hormonales et échographiques…).
L'induction de l'ovulation est jugée réussie devant la présence d'au moins un follicule à l'état mature (taille >=16mm) à J13, un taux d'E2 correspondant à 150-250pg/follicule mature et accessoirement une épaisseur de l'endomètre dépassant 8 mm. Dans l'ensemble, une ovulation était induite chez 62.5% dans le groupe «CC+metformine» par apport à 37.5% pour le groupe «CC+placebo». Cette différence, bien que statistiquement non significative (vu la petite taille de la population), est bien en faveur d'un apport bénéfique de la metformine dans le protocole de traitement, et ce, via l'augmentation de l'oestradiolémie et du taux des follicules matures et la réduction du nombre total des follicules. Loin d'agir sur l'obésité et/ou l'hyperandrogénie, l'effet de cette molécule semble bien lié à son effet insulinosensibilisateur qui parait le déterminant principal de son efficacité avec, éventuellement, une modulation des dysrégulations de la thèque et de la granulosa tant accentuées par l'hyperinsulinémie.
Conclusion : La prescription de la metformine chez les femmes à ovaires polykystiques permet d'améliorer la qualité de la réponse ovulatoire au citrate de clomifene.

MOTS-CLÉS

Syndrome des ovaires polykystiques, Stimulation ovarienne, Metformine

SUMMARY

Background: Polycystic ovary syndrome (PCOS) is a common, complex endocrine disorder for women on reproductive age. A high incidence of ovulation failure is observed in PCO women and perhaps linked to insulin resistance related to metabolic features
In the last few years some studies assessed hyperinsulinimea and insulin resistance attenuation effects, by insulin sensitizing agents such as metformin, in PCOS women suggesting potential scope for these drugs in CC ovulation induction quality improvement.
Aim : Our prospective study aim is to compare the effectiveness of clomifene citrate plus metformin and clomifene citrate plus placebo in women with newly diagnosed polycystic ovary syndrome.
Methods: From February 24 to September 29 (2007), PCOS was explored on women attending the Department of Obstetrics & Gynaecology sterility consultation unit (CHU Hedi Chaker-Sfax) according to the Rotterdam 2003 diagnostic criteria. PCOS patients were randomized to receive, in addition to clomifene citrate treatment, placebo or metformin 850 mg two times a day all ovulatory cycle for three trials maximum. Ovulation detection was done by the E2 serum measurements and ovarian transvaginal ultrasonography' evolution controlling on 7th, 11th and 13th day of the cycle.
Results: Within 7 months, 32 PCOS women were recruited in the study and equally allocated to the two groups. Baseline characteristics were similar in metformin group and placebo one. Ovulation was characterized by the presence of at least one mature follicle (>16mm), a circulating estradiol concentration in the edge of 150-250pg and accessory an endometrial depth >8mm. The ovulation rate in the metformin group was 62.5% compared with 37.5% in the placebo group, a non-statistically significant (small study population) but important difference (1.66 times). Analyses show a higher mature follicle number and estradiol concentration in metformin group than in the placebo one. Metformin effect was, in our study, his only insulinosensitizer property consequence far away a 'making thinner' or Hyperandrogenism reducing ones.
Conclusion: The ovulatory response to clomifene can be increased in polycystic ovary syndrome women by decreasing insulin secretion with metformin.

KEY-WORDS

Polycystic ovary syndrome, incuded ovulation, Metformine

إضافة الميتفورمين في التنشيط البويضي باستعمال الكلوميفين سترات لدى النساء المصابات بمتلازمة التكيس المبيضي

الباحثون : بن عياد . ب - الدماق دي ميلك . س - بن عرب . هـ - طربلسي . هـ - شحطور . هـ - المثلوثي . ن - ذويب . م - القسيس . م - سعيدان .د - طربلسي. ك - القرمزي . م.

المقدمة:ستعد متلازمة التكيس المبيضي من أهم الاضطرابات الإفرازية للمرأة في مرحلة الخصوبة و أكثرها انتشارا. يشمل هذا الاضطراب عديد العوارض لعل أهمها اختلال (أو انعدام)الأباضة المنجر عنه في أغلب الأحيان العقم والمعزز بما يعرف بحالة' المقاومة الوظيفية للأنسيلين 'المصاحبة عادة لمتلازمة التكيس المبيضي. مثل استغلال بعض المواد المخفضة لهذه الحالة،مثل الميتفورمين،و الإضافة الناجمة عنه في أطار التنشيط البويضي باستعمال الكلوميفين سيترات لدى النساء المصابات بمتلازمة التكيس المبيضي موضوع بحث للعديد من الدراسات في الآونة الأخيرة. في نفس هذا الإطار،نحاول في دراستنا مقارنة فاعلية استعمال الكلوميفين على حدة مقابل جمعه بالميتفورمين لدى النساء المصابات بمتلازمة التكيس المبيضي. الطّرق :خلال الفترة الممتدة من 24 فيفري إلى 29 سبتمبر 2007 تم انتقاء النساء المصابات بمتلازمة التكيس المبيضي من جملة اللواتي توجهن للمعاينة لدى وحدة العقم بقسم النساء و التوليد بمستشفى الهادي شاكر بصفا قس ،و ذلك حسب المعايير المحددة لتشخيص هذه المتلازمة بمؤتمر روتردام 2003. بصفة عشوائية ،وزعت النسوة المصابات إلى فريقين وصف لكل منهما الكلوميفين بالإضافة للعلاج البديل" فريق البلاسيبو "أو للميتفورمين بمعدل 850 مغ مرتين يوميا على طوال الشهر المبيضي " فريق الميتفورمين ." لمتابعة مدى فاعلية العلاجين ،تم الالتجاء للتصوير بالصدى للبويضات و تقدير معدل هرمن الأوستراديول في الدم ،و ذلك ،في الأيام 11 ،7 و 13 من الشهر المبيضي . النتائج :في غضون السبعة أشهر ،تم تشخيص متلازمة التكيس المبيضي لدى 32 امرأة معاينة قسمن بالتساوي إلى فريقين متشابهين على معظم الأصعدة و الخصائص(العمر ،القياسات ،الخاصيات الهرمونية ،التصويرية.)... في دراستنا حدد مفهوم نجاح التنشيط البويضي كالآتي :وجود مسام يانع 16=<(مم)واحد على الأقل ،تقدير معدل الأوستراديول في الدم ب 250-150 بيكوغرام/مسام و ثانويا ،قياس 8 مم كأقل تقدير لعمق بطانة الرحم باليوم 13. توفرت هذه المعايير ،و أعتبر العلاج فعالا آذا في التنشيط البويضي ،لدى ٪ 62.5 من جملة النساء في " فريق الميتفورمين "مقابل ٪37.5 ل" فريق البلاسيبو."تعد هذه النتيجة، بالرغم من كونها بلا د لالة من الناحية الإحصائية(نظرا للعدد الغير كافي للنسوة)، هامة على الصعيد التطبيقي باعتبار أن استخدام الميتفورمين قد مكّن من تحسينات معتبرة في النتائج 1.66 (مرة،)و ذلك ،من خلال زيادة في معدل الأوستراديول في الدم و نسبة المسام اليانعة. بعيدا عن أي أثر على الوزن أو على نسبة الهرمونات الذكرية ،اتضح ،من خلال الدراسة ،أن فعالية الميتفورمين ،في التنشيط البويضي لدى المصابات بمتلازمة التكيس المبيضي ،تكمن أساسا في خاصيته المخفضة لحالة' المقاومة الوظيفية للأنسيلين .'

الخلاصة :يساهم استخدام مادة الميتفورمين من تحسين الاستجابة البويضية للتنشيط البويضي باستعمال الكلوميفي سيترات لدى النساء المصابات بمتلازمة التكيس المبيضي.

Polycystic ovary syndrome (PCOS) is a common, complex and heterogeneous endocrine disorder for women on reproductive age. Using the more recent Rotterdam consensus (1), PCOS diagnosis include two of the following three criteria: oligo- or anovulation, clinical and/or biochemical signs of hyperandrogenism, polycystic ovaries and exclusion of other aetiologies. PCOS pathogenesis is still largely unknown. Women with the syndrome are frequently insulin resistant, independent of obesity (2). In this regard, several data support the hypothesis that insulin resistance and the associated hyperinsulinemia play a pathogenetic role in PCOS (2). Insulin concentrations are proported to hyperandrogenism because of increased ovarian androgen production and decreased synthesis of sex hormone-binding globulin (SHBG). PCOS management depends largely on the cause of the women consulting. In the case of the anovulatory sterility, which the PCOS is responsible for 40% of, the first choice drug is the antioestrogen clomifene citrate. It enhances release of pituitary gonadotrophins, resulting in follicular recruitment. Three quarters of women with polycystic ovary syndrome will ovulate with clomifene citrate. Complications of treatment are rare and usually mild. Patients who do not ovulate on the maximum dose of 150 mg are considered to be clomifene citrate resistant (3). In the latter we frequently go for the use of gonadotrophins or the laparoscopic ovarian surgery.

As hyperinsulinaemia plays a significant role for anovulation in PCOS women, clinical improvements can be anticipated following serum insulin concentrations reduction (4).

Weight loss in obese women with PCOS leads to a reduction of hyperinsulinaemia and so an amelioration of the symptoms especially ovulation and pregnancy rate (5).

Insulin-sensitizing agents, not yet having the AMM for this indication, have been tried in PCOS patients' management (6). Metformin, recommended extensively for the type II diabetes treatment, is now the most widely used insulin sensitizer for induction of ovulation in women with polycystic ovary syndrome. This drug improves insulin sensitivity by different mechanisms, thus determining a subsequent reduction in plasma insulin levels (7, 8). Benefit results are observed on ovulation rate, hyperandrogenism and obesity (BMI<40kg/m^2) (3). This has led to the recommendation to use metformin alone or in combination with clomifene citrate as first line treatment in infertile women with polycystic ovary syndrome.

The aim of this prospective study was to determine whether reducing hyperinsulinemia with metformin would increase the ovulatory response to clomifene in women with the polycystic ovary syndrome.

METHODS

Patients

From 24 February to 29 September 2007, patients attending the sterility consultation unit of the Department of Obstetrics & Gynaecology, CHU Hedi Chaker-Sfax, were recruited for our study since they fulfilled the Rotterdam 2003 PCOS' diagnosis criteria. Diagnosis of PCOS was based on the presence of ovulation abnormalities, clinical and/or biochemical signs of hyperandrogenism, polycystic ovaries (table 1), after exclusion of Cushing's syndrome, late-onset 21-hydroxylase deficiency, thyroid dysfunction, hyperprolactinemia or androgen secreting tumors …

Table 1: PCO diagnostic criteria researchable in the Department of Obstetrics & Gynaecology sterility consultation unit, CHU Hedi Chaker-Sfax (UP).

Ovulation abnormalities	Hyperandrogenism signs	polycystic ovaries (Echography)
Oligomenorrhea / amenorrhea	Hirsutism	Ovary's surface > 12 cm2
LH/FSH ≥ 1	Severe acne	follicles number>10 follicles of
	Testosterone	2–10 mm/2 ovaries
		ovarian stroma density

Experimental Protocol

Through the interrogatory and clinical exams, carried out in the first consultation, a primary selection for the suspected patients that suffer from PCOS was done.

Hormonal analyses (FSH, LH, estradiol, testosterone, prolactine) were systematically recommended for these patients in the ovulatory cycle' third day.

Transvaginal ultrasonography was accomplished generally in the day 12 of the cycle. Taking in consideration these exams results and the Rotterdam consensus, PCOS was diagnosed for 32 women.Those patients were randomly allocated to clomifene citrate plus metformin (metformin group, 16 patients) or

clomifene citrate plus placebo (placebo group, 16 patients).
From the third day to the seventh day after menstruation, women of the two groups took 100 mg clomifene citrate a day (Serpafar ®, tablet containing 50 mg of clomifene).
The patients of the merformin groups received, in addition to clomifene, 1700mg of metformin (Glucophage ® retard, tablet containing 850 mg of metformin) all over the cycle.
Obese patients were asked to be on a healthy diet and practice more physical exercise in order to lose weight and improve their lives quality.
Ovarian activity was assessed by E2 serum measurements and ovarian transvaginal ultrasonography on day 7, 11 and 13 of the cycle. Women who didn't ovulate on the first cycle of treatment were asked to go for a second eventually third time as maximal number of trial before classifying them 'no responder'.
Taking in consideration the short time of the study and some women' lack of comprehension and adherence, we couldn't ensure the six cycles recommended in the literature to definite a CC-resistant (9).

Statistical analysis
The principal outcome measure was ovulation. We describe the populations and compared the cumulative ovulation rates as well as other rates and proportions among groups using student and Chideux (with a 5% significance level) test statistics by means of SPSS 11.5.1.

RESULTS

Patients
Applying the Rotterdam 2003 consensus, PCOS was diagnosis with 32 women.

Table 2: Baseline characteristics of women in study according to allocation to clomifene citrate plus metformin or clomifene citrate plus placebo. (UP).

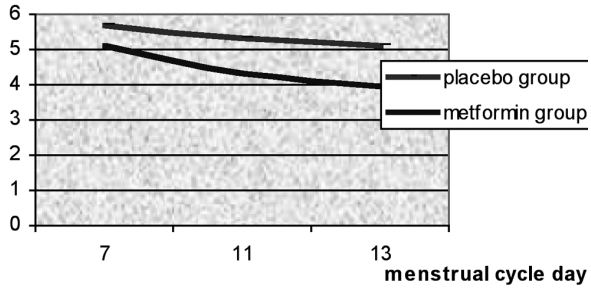
Parameters		Population (n=32)	Placebo group (n=16)	Metformin group(n=16)
Mean age (years)		31.09 (22-42)	32.81 (22-35)	29,38 (25-42)
Primary sterility		22	13	9
Smoking habit		1	1	0
hypertension		1	1	0
BMI (kg/m²)	**Mean BMI**	28.43 (21.83- 33.51)	28.01	28.45
	<20	0	0	0
	20< <25	7	3 (18.8%)	4 (25%)
	25< <30	10	5 (31.25%)	5 (31.25%)
	>30	15	8 (50%)	7 (43.75%)
waist:hip ratio>0.9		25	12	13
Ovulatory signs	**oligomenorrhea**	20	8	12
	Amenorrhea	3	2	1
	Irregular cycle	18	7	10
Hirsutism		9 (28,13%)	5 (31.25%)	4 (25%)
Acne		17 (53,13%)	6 (37.5%)	11 (68.75%)
Acanthosis nigricans		2 (6,25%)	0	2 (12.5%)
Hormonal	**Mean FSH (mUI/ml)**	7,04 (2.6-13.7)	7,48	6,61
assays	**Mean LH (mUI/ml)**	8,58 (1.4-25.8)	7,06	10,09
	LH/FSH mean	1,32	1,08	1.57
	>1	18 (56.25%)	7 (43.75%)	11 (68.75%)
	Mean Estradiol (pg/ml)	63,92 (2.84-227.3)	61,33	67,38
	Mean Prolactine (ng/ml)	23,71(5.9-86)	28,58	17,71
	Mean Testostérone (ng/ml)	1,14(0.18-7)	0,86	1,53
Polycystic ovary (echography)				
	Ovary surface>12cm²	25 (78.13%)	14 (87.5%)	11 (68.75%)
	ovarian stroma hyperdensity	6 (18.75%)	1 (6.25%)	5 (31.25%)
	follicles number>6 follicles	28 (87.5%)	14 (87.5%)	14 (87.5%)
	<8 mm / ovary			

23 patients had oligoamenorrhea (fewer than six menstrual periods in the preceding year). Clinical signs of hyperandrogenemia (acne, hirsutism, …) and/or biochemical one (elevated serum free testosterone concentrations) were found with 19 women, however 28 one had signs ultrasonography of the ovaries consistent with the diagnosis of the polycystic ovary syndrome (ovary's surface>12cm^2, peripheral follicles number>10 follicles of 2–10 mm in diameter and increased density of ovarian stroma …).
Patients with the whole three criteria were found to be 13.
The median BMI (binding mass index, defined on the first consultation) in this study was 28,43 kg/m2, ranging from 21.83 to 33.51 kg/m2. 15 women (47% of the population) were described as obese with a BMI>30 kg/m^2 .
Baseline characteristics were similar in the two groups (table2). Age, hyperandrogenism and obesity analysis of those groups shows the absence of a statistical significant difference.
Among the 25 obese and extra weight women, asked to lose some mass before taking medication, only 10 were on the diet. But no detectable difference in weight was observed.
None had abandoned the study but some women were not to the treatment adherent (forgetfulness, misunderstanding of the protocol…). Consequently, other cycle of experience was conducted for these patients.
Metformin gastrointestinal side effects (10) were reclaimed by almost all the patients without, although, giving up the study.
In the other side, we defined one case of ovarian hyperstimulation high risk (one mature follicle (16mm), many small immature follicles (>10/ovary) and 2998,4 pg/ml of E2 on day 13 of the cycle) . The patient, belonging to the metformin group, was at her third trial. The treatment was so interrupted and the case was classified as 'no responder'.

Ovulation induction
In our study, ovulation was characterized by the presence of at least one mature follicle (>16mm), a circulating estradiol concentration in the edge of 150-250pg and accessory an endometrial depth >8mm.
The failure of ovulation induction was defined by:
- A high risk of ovarian hyperstimulation (E2>3000pg with many small immature follicle).
- Insufficient ovulatory response within three cycle of treatment (mature follicle absence and low E2 concentrations).
In the whole population 16 patients (50%) ovulated, 10 belonged to the metformin group (62.5%) compared to 6 in the placebo one (37.5%).
Cumulative rates of ovulation show to be much higher in the metformin group (1.66 time the placebo group rate), but the difference was, statistically, not significant (p = 15.72% > 5%).

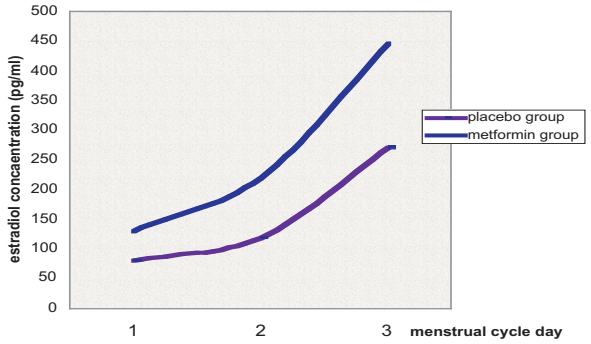
Follicle number
As shows figure 1, in day 13 the follicle number counted via echographical monitoring for metformin group women is much lower than placebo group one (23.17% of reduction).
Mature follicle percentage of the total follicle pool is estimated to be 35% in metformin group, while the same percentage is 15% in placebo group.

Figure 1 : Total follicles number evolution on day 7, 11 and 13 (UP).



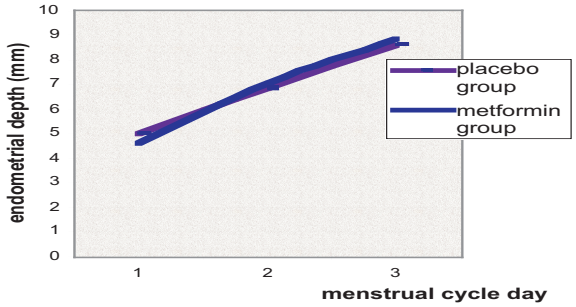
Estradiol
The estradiol concentration evolution is illustrated by figure 2

Figure 2 : estradiol concentration evolution on day 7, 11 and 13(UP).



Endometrial depth
The endometrial depth evolution is illustrated by figure 3

Figure 3 : endometrial depth evolution on day 7, 11 and 13(UP).



Obesity
Ovulation rate in metformin group obese women was 3/7 (42.85%) and 2/8 (25%) for the placebo one.
Subdividing the whole population into obese (BMI>30kg/m^2) (n=15) and no-obese women (n=17), ovulation rate (two protocols included) was 33.33% in the first group and 64.7% in the second one…

From statistical point of view, no significant difference in ovulation rates between obese and no obese women of the whole population, neither between metformin and placebo group was observed.

Hyperandrogenism

Ovulation rate in hirsute women of two groups were 4/8 (50%) in placebo group versus 6/11 (54.54%) in metformin one.

Among the 19 hirsute women of the whole population, 10 ovulate (47.36%). For the no hirsute (13), only 6 ovulate (46.15%). Statistical analysis show no significant difference in ovulation rates between hirsute and no hirsute women of the whole population, in a hand, and of metformin and placebo group in the other one.

Age: (figure 4)

Ovulation study in step with the different age sections (figure5) in the entire population shows highest rates with sections <30 and 30-35years (61.53% and 87.5 % respectively).

Apart from sections <30 and >40 years, ovulation rates seems to be much higher for women belonging to metformin group compared with placebo ones (figure 4).

Figure 4 : ovulation rate in step with age(UP).

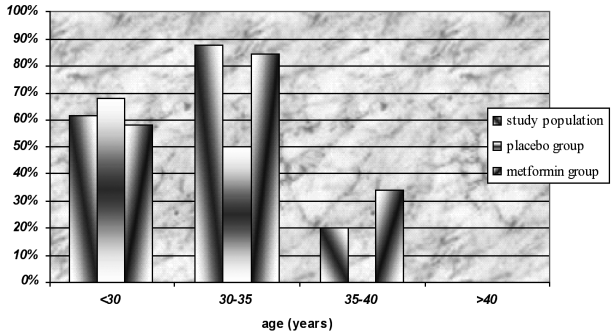
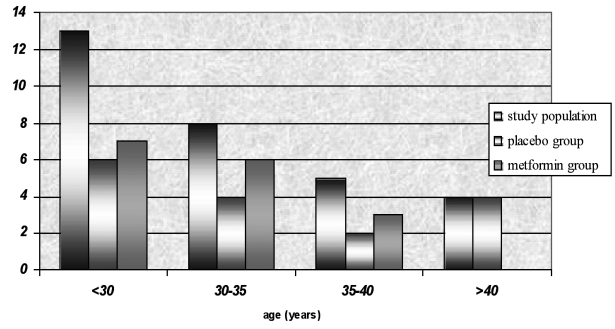


Figure 5 : patients repartition in step with age. (UP).



DISCUSSION

Most of the studies addressing the use of metformin in patients with PCO have been observational and focused on the menstrual pattern and hormonal profile.

The effect of such an insulinsensitizer added to ovulation induction protocol, especially using the clomifene citrate, has rarely been documented.

There have been only eight randomized placebo-controlled studies published in the literature but their results are conflicting (table 3).

Table 3 : Summary of published studies on the use of metformin in CC treated PCOS' women (UP).

Study	N	Mean BMI	Ovulation rate «metformin group»	Ovulation rate «placebo group»
Nestler and al. (1998)	61	32.3	90%	8%
El-Biely and Habba (2001)	90	28	80%	65%
Vandermolen and al.(2001)	27	38	75%	27%
Hun Yu Ng and al.(2001)	18	24	5%	21% (NS)
Kocak and al. (2002)	56	31	77%	14%
Malkawi and Qublan (2002)	28	-	68.75%	25%
Sturrock and al. (2002)	26	33.3	42%	29% (NS)
Moll and al. (2006)	162	28.14	64%	72% (NS)

Vandermolen and al. (2001) (11), Kocak and al. (2002) (12) and Malkawi and Qublan (2002) (13) reported significant improvements in ovulation from CC treatment after taking metformin in obese women with CC-resistant PCO.

Similarly, Nestler and al. (1998) (14) and El-Biely and Habba (2001) (15) demonstrated that in obese PCOS women the use of metformin significantly improved the ovulation rate and increased the SHBG concentration when compared with the control.

In contrast, some studies show no improvement in the number of women having evidence of ovulation and in the ovulation rate in PCO women after metformin added to the CC protocol, when compared to the placebo. In this way, Moll and al. (2006) (16) and Hun Yu Ng and al. (2001) (17) demonstrated a higher ovulation rate taking place in placebo group comparatively to metformin one.In the first, moll and all included women with polycystic ovary syndrome defined according to the 2003 consensus without taking in consideration any other criteria for the selection. The heterogeneity of the initial population can be behind the result especially if added to the high number of dropouts in the metformin group (63 droppers out). In the other hand, Hun Yu Ng and al, studying 18 thin Chinese PCOS women, demonstrated also a little superiority of the ovulation rate occurring in placebo group. Attention should be attracted, although, to the low percentage in two groups (5% and 21%). This no improvement in the ovulation rate, contrasting a significant reduction of body mass index, serum testosterone and fasting leptin concentrations in the metformin group, may be the little number of the participants and/or the unusual selection criteria (thickness, Asiatic origin…) consequence. The smallness of the population is described also as the reason for the insignificant difference found with the Sturrock and

al.(2002)' study (18) demonstrating, although, an improvement in the number of women having evidence of ovulation and in the ovulation rate in 26 obese women with CC-resistant PCO after metformin added to the CC protocol to the placebo.
In this study, metformin contributed to multiply the ovulation rate CC-induced by a 1.66 factor. This result contrasts a significant statistical difference absence.
This result can be justified by 2 reasons:
- The study population smallness (32 patients). Moreover, the same percentages lead to a statistical significant difference if related to a 64 patients' population.
- The selection of patients on the Rotterdam consensus criteria' only base: obesity, CC-resistance, age…weren't included on the inclusion or exclusion criteria; so that, our study group reflects the largest group of women with polycystic ovary syndrome a fertility clinic will see and treat. Obesity factor elimination (to avoid its effects on CC or metformin resistance) was unsuccessful and the study was accomplished on that heterogenic population. The metformin benefit contribution on the CC ovulation induction seems to be related to its effects on estradiol concentration (spectacular climb on the day 13) and mature follicle number. Metformin action on the endometrial depth couldn't be characterized in the study since most of the women were taken an estrogenic treatment avoiding the cervical CC side effects (9). Contraception during the use of metformin is no longer recommended. In fact,Recent data provide some reassurance about the safety of metformin in respect of lack of teratogenicity when taken in early pregnancy, although no long-term follow-up data are available(19, 20, 21). Limited data are available about the pharmacokinetics of metformin during pregnancy. In one small study of seven women, the clearance of metformin increased with gestation and the associated increased renal elimination. More data are required to clarify the possible need for dose adjustment as pregnancy proceeds.
Moreover, some authors recommended the use of the molecule at pregnancy time for reducing either gestational diabetes risk effect (22) or spontaneous abortion rate (23). In our study we were limited to ovulation detecting without looking forward at the pregnancy possibilities. Generally, we recommend the progressive ascension of metformin dose, so that we reach within 2 weeks the 1700mg fixed as necessary for the ovulation induction treatment. This protocol seems to avoid the intestinal molecule side effects (24). Latter, observed with all metformin group patients, were totally justified since we operated directly with 1700mg dose in order to get right away the optimal molecule effect. Ovarian hyperstimulation as well as multiple pregnancies are the most described CC side effects (9). Metformin can help reducing those problems risk by decreasing gonadotrophin and androgen producing when added to CC (25, 26). This fact is not yet confirmed by a specific study (27). One case of high ovarian hyperstimulation was detected in our study. Belonging to the metformin group, the patient was on her third trial of treatment which may amplify the CC effect. On the other hand, metformin, in our study, seems to lessen ovarian hyperstimulation probability since it decrease total follicle number and at the same time increase the mature follicle number observed with metformin group comparatively to the placebo one. This lead to reduce, in metformin treated PCO patients, the immature follicle number wchich is positively proportional to the ovarian hyperstimulation risk (28).
Obesity-ovulation induction relation is largely discussed in literature. It seems to have 2 faces. Some authors describe an important role of overweight in induction ovulation drugs resistance such as CC (9) and metformin (29, 30).
In our study the evaluation of the ovulation rate in obese and thin women groups demonstrate a higher one in the second group. The considerable difference (almost double), in spite of being statistically insignificant (small population), may verify the literature in this way.In the other hand, metformin is accused to have a 'making thinner' effect, not yet confirmed (31, 32). Some study consider that reducing weight (by strict diet only or combined to healthy life style) may have a benefit impact on spontaneous and induced ovulation as well as other PCO' symptoms (metabolic syndrome, hyperandrogenism, hyperinsulinimea…) (3). Considering those information, the improvement on ovulation rate induced by metformin can be the consequences of the weight reduce that it make and which influence directly the ovulation. For our study, the rate ovulation difference for the obese in two groups was statistically insignificant which infirm a metformin action by means of the 'making thinner' effect. Hyperandrogenism is also described in literature as an ovulation induction resistance factor (9). Lack of an important significant difference in ovulation rate between hirsute group and no hirsute one in our study contrast this fact.Metformin reducing hyperandrogenism action stills to be a discussion topic. In fact, some uncontrolled studies demonstrate an ovarian androgen production reduction with metformin treated PCO women (33, 34). Accomplished on small and obese population, those research were largely contrasted by other studies such as Crave and al. (1995) (35) who demonstrate a reducing hyperandrogenism loosing weight' affect but not a metformin one and Ehrmann and al. (1997) (36) who didn't found a real reduction of circulating androgen concentration using metformin with PCO women.
Knowing the important contribution of ovarian androgen on the PCO pathogenesis, metformin can effect ovulation via reducing androgen concentration. In our study, the difference in ovulation rate between the metformin and placebo groups can't be owed to the metformin androgen production reduction effect since there is no valuable significant difference in the ovulation rate for hirsute of two groups. The ovulation rate decreases, in the placebo group of our study, as getting on in age. This fact, although not yet confirmed by specific research, can be justified by the natural follicle number decline (37) and some observations such as gonadotrophin doses increasing necessity in step with age one (38).In the same way, none of the preceding study had focused on the age-metformin relation. In ours, this latter seems to be efficient when the CC efficacy decreases. In fact, without an additional effect on the <30years section, 'metformin+CC' shows higher ovulation rate on 30-35 and 35-40 years sections than the CC alone. Mechanism and justification still to be discovered. For the >40 years section, all the patients belonged to placebo group. Eliminating, in our

study, an impact of reducing weight or androgen concentration on its benefit effect on CC ovulation induced, metformin seems to act via the only insulinosensitizer property. The resultant decrease in insulin concentration may soften the amplificatory impact of insulin on thécal deregulation or of the granulosa one, tissular dysfunction described to be behind the PCO pathogeneses. Metformin is the most known insulinosensitizer for the PCO women treatment, but not the only. Other drugs such as D- chiro-inositol and naltrexone are being evaluated for this indication and even used mainly the thiazolidinedione family (3).

CONCLUSION

Based on the results of this trial, metformin seems to improve PCO women CC response.

The important rate ovulation difference observed when adding metformin to the CC protocol, even though not statistically significant, may result of a mature follicle number and estradiol concentration metformin induced. The mechanism of this achievement seems to be explained by the only insulinosensitizer property. The CC resistance is for now an increasing obstacle for PCO women ovulation induction wchich explains the huge recent resort to gonadotrophin, what is so, the addition of metformin on its use protocol?

RÉFÉRENCES

1. Fauser B, Tarlatzis B, Chang J, Azziz R, Legro R, Dewailly D, et al. The Rotterdam ESHRE/ASRM-sponsored PCOS consensus workshop group. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS). Hum Reprod 2004;19:41-7
2. Dunaif A. Insulin resistance and the polycystic ovary syndrome: mechanism and implications for pathogenesis. Endocr Rev.1997; 18:774–800.
3. Robert K., Bogusaw O., Anna G.D., Zbigniew S.H.: Update on the management of polycystic ovary syndrome. Pharmacological Reports, 2006, 58, 614-625
4. Pasquali, R. and Filicori, M.Insulin sensitizing agents and polycystic ovary syndrome. Eur. J. Endocrinology, 1998 ; 138, 253–254
5. Moran LJ, Brinkworth G, Noakes M, Norman RJ: Effects of lifestyle modification in polycystic ovarian syndrome. Reprod Biomed Online, 2006; 12: 569–578.
6. Dewailly D.: What's new? PCO?. Reproduction humaine et hormones, 1999; 8:658-665
7. Baillargeon JP, Iuorno MJ, Nestler JE: Insulin sensitizers for polycystic ovary syndrome. Clin Obstet Gynecol, 2003; 46: 325–340.
8. Glueck CJ, Goldenberg N, Streicher P, Wang P: Metformin and gestational diabetes. Curr Diab Rep, 2003; 3: 303–312.
9. Homburg R: Clomiphene citrate – end of an era? Hum Reprod, 2005, 20, 2043–2051
10. Baillargeon JP: Use of insulin sensitizers in polycystic ovarian syndrome. Curr Opin Investig Drugs, 2005; 6: 1012–1022.
11. Vandermolen DT, Ratts VS, Evans WS, Stovall DW, Kauma SW, Nestler JE. Metformin increases the ovulatory rate and pregnancy rate from clomiphene citrate in patients with polycystic ovary syndrome who are resistant to clomiphene citrate alone. Fertil Steril 2001;75: 310-5
12. Kocak M, Caliskan E, Simsir C, Haberal A.: Metformin therapy improves ovulatory rates, cervical scores, and pregnancy rates in clomiphene citrate-resistant women with polycystic ovary syndrome. Fertil Steril 2002;77: 101-6
13. Malkawi HY, Qublan HS. : The effect of metformin plus clomiphene citrate on ovulation and pregnancy rates in clomiphene-resistant women with polycystic ovary syndrome. Saudi Med J 2002; 23: 663-6.
14. Nestler JE, Jakubowicz DJ, Evans WS, Pasquali R.: Effects of metformin on spontaneous and clomiphene-induced ovulation in the polycystic ovary syndrome. N Engl J Med 1998;338: 1876-80.
15. El-Biely MM and Habba M.: The use of metformin to augment the induction of ovulation in obese infertile patients with polycystic ovary syndrome. Middle East Fertil Soc J 2001; 6: 43-9
16. Moll E, Bossuyt PM, Korevaar JC, Lambalk CB, van der Veen F:. Effect of clomiphene citrate plus metformin and clomiphene citrate plus placebo on induction of ovulation in women with newly diagnosed polycystic ovary syndrome: randomised double blind clinical trial. BMJ 2006; 332:1485
17. Hung Yu Ng E., Ming Sun Wat N., Chung Ho P.: Effects of metformin on ovulation rate, hormonal and metabolic profiles in women with clomiphene-resistant polycystic ovaries: A randomized, double-blinded placebo-controlled trial. Hum Reprod 2001; 16: 1625-31
18. Sturrock ND, Lannon B, Fay TN.: Metformin does not enhance ovulation induction in clomiphene resistant polycystic ovary syndrome in clinical practice. Br J Clin Pharmacol 2002; 53: 469-7343-9.
19. Glueck CJ, Streicher P, Wang P: Treatment of polycystic ovary syndrome with insulin-lowering agents. Expert Opin Pharmacother, 2002; 3: 1177–1189.
20. Heard MJ, Pierce A, Carson SA, Buster JE.: Pregnancies following use of metformin for ovulation induction in patients with polycystic ovary syndrome. Fertil Steril, 2002; 77: 669-73
21. Jakubowicz DJ, Iuorno MJ, Jakubowicz S, Roberts KA, Nestler JE. : Effects of metformin on early pregnancy loss in the polycystic ovary syndrome. J Clin Endocrinol Metab 2002; 87: 524-9
22. Essah PA, Nestler JE: The metabolic syndrome in polycystic ovary syndrome. J Endocrinol Invest, 2006; 29: 270–280.
23. Simmons D, Walters BN, Rowan JA, McIntyre HD: Metformin therapy and diabetes in pregnancy. Med J Aust, 2004; 180: 462–464
24. Goodarzi MO and Korenman SG: The importance of insulinresistance in polycystic ovary syndrome. Fertil Steril, 2003; 80: 255–258.
25. Diamanti-Kandarakis E, Papailiou J, Palimeri S: Hyperandrogenemia: pathophysiology and its role in ovulatory dysfunction in PCOS. Pediatr Endocrinol Rev, 2006; 3 (Suppl 1): 198–204
26. Seli E, Duleba AJ: Treatment of PCOS with metformin and other insulin-sensitizing agents. Curr Diab Rep, 2004; 4: 69–75.
27. Pritts EA: Treatment of the infertile patient with polycystic ovarian syndrome. Obstet Gynecol Surv, 2002; 57: 587–597.
28. Germond M., Gaillard M.C., Senn A. : Syndrome d'hyperstimulation ovarienne. Archives of gynecology and obstetrics, 2005; 246: 53
29. Maciel GAR, Junior JMS, Motta ELA, Haida MA, Lima GR and Baracat EC: Nonobese women with polycystic ovary syndrome respond better than obese women to treatment with metformin. Fertil Steril . 2004; 81:355–360
30. Tang T., Glanville J., Hayden C.J., White D., Barth J.H., Balen A.H.: Combined lifestyle modification and metformin in obese patients with polycystic ovary syndrome. A randomized, placebo-controlled, double-blind multicentre study. Human Reproduction, 2006; 21:80-89
31. Wong LL and Wong TC : Metformin induced anorexia and weight loss. Hawaii Med J, 2003; 62:104–5
32. Siraj ES : Is there a role for metformin or acarbose as a weight-loss agent in the absence of diabetes? Cleve Clin J Med, 2003; 70:702–704
33. Kolodziejczyk B., Duleba, A.J., Spaczynski, R.Z.: Metformin therapy decreases hyperandrogenism and hyperinsulinemia in women with polycystic ovary syndrome. Fertil. Steril.,2000; 73: 1149–1154
34. Nestler JE, Jakubowicz DJ.: Decreases in ovarian cytochrome P450c17activity and serum free testosterone after reduction in insulin secretion in polycystic ovary syndrome. N Engl J Med. 1996;335:617–623
35. Crave J.C, Fimbel S., Lejeune H., Cugnardey N., Dechaud H., Pugeat M.: effects of diet and metformin administration on sex hormone-binding-globulin, androgens and insulin in hirsute and obese women. J Clin Endocrinol Metab, 1995;80 : 2057-62.
36. Ehrmann DA, Cavaghan MK, Imperial J, Sturis J, Rosenfield RL, Polonsky KS.: Effects of metformin on insulin secretion, insulin action and ovarian steroidogenesis in women with polycystic ovary syndrome. J Clin Endocrinol Metab.1997; 82:524–530
37. Faddy MI, Gosden RG, Gougeon A, Richardson SJ, Nelson JF.: Accelerated disappearance of ovarian follicles in midlife: implications for forecasting menopause. Hum Reprod, 1992; 7: 1342 - 1346.
38. FIVNAT : Procréations médicalement assistées à 40 ans. Contracept Fertil Sex, 1990;18: 595