Clomiphene fails to revert hypogonadism in most male patients with conventionally treated nonfunctioning pituitary adenomas

Clomifeno não reverte o hipogonadismo na maioria dos homens com adenomas pituitários não funcionais tratados de forma convencional

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ABSTRACT

Objective: To evaluate the effect of clomiphene in men with hypogonadism and conventionally treated nonfunctioning pituitary adenomas (NFPA). **Patients and methods:** Open label, single-arm, prospective trial. Nine hypogonadal men (testosterone < 300 ng/dL and low/normal LH) with previously treated NFPA. Clomiphene (50 mg/day orally) for 12 weeks. Testosterone, estradiol, LH, FSH, prolactin and erectile function were evaluated before and after 10 days, 4, 8 and 12 weeks of clomiphene treatment. **Results:** After clomiphene treatment, testosterone and erectile function improved in only one patient. In the remaining eight patients, testosterone levels decreased whereas LH, FSH, and estradiol remained unchanged. Insulin sensitivity increased in unresponsive patients. **Conclusions:** Compared with hypogonadal men with prolactinomas under dopaminergic therapy, clomiphene treatment failed to restore normal testosterone levels in most patients with conventionally treated NFPA. Arq Bras Endocrinol Metab. 2011;55(4):266-71

Keywords

Nonfunctioning pituitary adenoma; hypogonadotropic hypogonadism; clomiphene; testosterone

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RESUMO

Objetivo: Avaliar o efeito do clomifeno em homens com hipogonadismo e adenoma hipofisário não funcionante (NFPA) previamente tratados. **Pacientes e métodos:** Aberto, braço único, prospectivo. Nove homens hipogonádicos (testosterona < 300 ng/dL e LH normal/baixo) com NFPA previamente tratados. Clomifeno (50 mg/dia oral) por 12 semanas. Testosterona, estradiol, LH, FSH, prolactina e função erétil foram avaliados antes e após 10 dias, 4, 8 e 12 semanas de clomifeno. **Resultados:** Após clomifeno, a testosterona e a função erétil melhoraram em um paciente. Em outros oito pacientes, os níveis de testosterona reduziram enquanto os níveis de LH, FSH, e estradiol permaneceram inalterados. A sensibilidade à insulina aumentou nos não respondedores. **Conclusões:** Em contraste com homens hipogonádicos com prolactinomas tratados com agonistas dopaminérgicos, a maioria dos hipogonádicos com NFPA falha em restaurar os níveis de testosterona durante o uso de clomifeno. Arq Bras Endocrinol Metab. 2011;55(4):266-71

Descritores

Adenoma hipofisário não funcionante; hipogonadismo hipogonadotrófico; clomifeno; testosterona

INTRODUCTION

Clinically nonfunctioning pituitary adenomas (NFPA) are usually diagnosed due to compressive symptoms, most typically visual field abnormalities, headache and hypopituitarism. Hypogonadotropic hypogonadism in patients with NFPA is highly prevalent both at diagnosis and after conventional treatment with pituitary surgery and/or radiotherapy (1-5). Untreated hypogonadism decreases patient quality of life and fertility and increases cardiovascular risk (6-8).

Male hypogonadal patients are usually treated with testosterone replacement, most often with intramus-

cular injections that require frequent applications and induce large fluctuations in serum testosterone levels, with corresponding fluctuations in patient energy, libido, sexual performance and mood. In addition, testosterone replacement has an inhibitory effect on spermatogenesis and fertility, which is undesirable in patients who want to have children (7,8).

Clomiphene is a well-known selective estrogen receptor modulator that increases gonadotropin secretion via hypothalamic-pituitary action (9). Clomiphene has been extensively used in the evaluation of the gonadotropic axis and in the induction of ovulation. Clomiphene has also been shown to revert hypogonadotropic hypogonadism in several conditions, such as falciform anemia, uremia, alcohol and steroid abuse, and in the stimulation of gonadotropin secretion in patients with sulpiride-induced hyperprolactinemia and gonadotropin suppression (10-14). More recently, we have shown that clomiphene led to the recovery of gonadal function in most male patients with prolactinomas and persistent hypogonadism under dopamine agonist treatment, irrespective of prolactin levels (15). In this study, we evaluated the therapeutic potential of clomiphene in reverting hypogonadism in patients with conventionally treated NFPA.

PATIENTS AND METHODS

Patients

Twelve consecutive adult male patients with NFPA, previously treated with surgery with or without radiotherapy, seen during one year at the Neuroendocrine Unit of the Endocrinology Division, Escola Paulista de Medicina, Universidade Federal de São Paulo, presented hypogonadotropic hypogonadism and were considered to be included the protocol. Hypogonadotropic hypogonadism was defined by total testosterone levels below 300 ng/dL (10.5 nmol/L), and normal/low LH levels after at least two months of surgery. Testosterone replacement was discontinued for at least two months before evaluation. Three patients were excluded from the study due to prostate disease, limitations in schedule, or cognitive deficit. Nine patients completed the study.

Study design

This study was an interventional, open label, uncontrolled, single-arm, prospective trial with oral clomiphene citrate (Clomid®, Medley, kindly provided by Dr. Enrico Repetto), 50 mg/day for 12 weeks, designed to assess the effects of the drug on serum testosterone levels of male patients with NFPA previously treated with surgery. The secondary aims of the study were to evaluate the effects of clomiphene on sexual function, body composition, lipid and glucose metabolism, and quality of life.

Physical examination, erectile function and quality of life questionnaires, serum hormone measurements (testosterone, SHBG, estradiol, prolactin, LH and FSH), serum PSA, lipids (total, LDL and HDL cholesterol, and triglycerides), glucose and insulin (before and after oral glucose), and body composition were all analyzed before and at the end of 12 weeks of clomiphene treatment. In addition, hormonal evaluations were performed at 10 days and at 4 and 8 weeks of treatment, and after discontinuing clomiphene treatment for 12 weeks. The study protocol was approved by the ethics committee of the institution, and a written informed consent was obtained from all participants.

Assays

Serum testosterone, SHBG, estradiol, prolactin, LH, FSH, insulin and PSA were measured in fasting serum by automated immunochemoluminescent assays (ADVIA Centaur, Bayer). Free testosterone values were calculated from total testosterone and SHBG results using a fixed albumin level, as previously described (16). Glucose was measured in plasma using an enzymatic method (Hexokinase II). Total cholesterol and triglycerides were measured in plasma by a peroxidase colorimetric immunoenzymatic method. HDL cholesterol was measured in serum using an immunoenzimatic method, and LDL and VLDL were calculated.

Body composition

Body composition [lean body mass, fat mass and bone mineral content (BMC)] was determined by dual-energy x-ray absorptiometry (QDR 2000-Plus, Hologic), according to the ICSD guidelines (17).

Questionnaires

Erectile function was evaluated using a 15-question survey (IIEF, international index of erectile function), and quality of life was evaluated using a 26-question survey (WHOQOL-BREF, World Health Organization – Quality of Life). Both questionnaires were previously validated in Portuguese (18,19).

Statistical analysis

Patients were classified as responsive and unresponsive according to their testosterone levels during treatment (> 300 ng/dL or 10.5 nmol/L), in at least 2 of 3 measurements between 4 and 12 weeks of clomiphene treatment. Statistical analyses were carried out using ANOVA or Friedman's test according to data distribution, followed by Bonferroni's or Dunn's post-tests, respectively. Comparisons within the same group were performed by paired *t* test or Wilcoxon signed-rank test, according to data distribution. Comparisons between two independent groups were performed by unpaired *t* test or Mann-Whitney test, as appropriate. Correlations between two variables were calculated by Pearson coefficient (r). Statistical significance was set at $P < 0.05$. Data were analyzed using GraphPad Prism 4.03 and GraphPad StatMate 2. Data are presented as mean ± standard error (SE), or as median when indicated.

RESULTS

Baseline characteristics

As shown in table 1, all nine patients that concluded the study had large macroadenomas at diagnosis, and had been previously treated at least once with pituitary surgery. Six patients had residual tumors after their last surgery. Five patients received conventional radiotherapy. All patients were on stable replacement doses of thyroxine with normal serum FT4 levels, and five were on glucocorticoid replacement therapy with prednisone (2.5-5.0 mg/d P.O.). Four patients had low levels of IGF-1, and two patients had low levels of prolactin.

Gonadotropins, total and free testosterone and estradiol levels during clomiphene treatment

As shown in table 2, eight patients did not respond to treatment. In this group, testosterone levels not only failed to increase, but also decreased significantly during clomiphene treatment, whereas FSH, LH, and estradiol levels showed no significant changes. SHBG increased significantly from 18.2 ± 4 nmol/L to 31 ± 5 nmol/L ($P < 0.05$), and free calculated testosterone decreased significantly from 3.9 ± 0.6 ng/dL to 1.9 ± 0.6 ng/dL (0.13 ± 0.02 nmol/L to 0.06 ± 0.02, $P < 0.01$) after 12 weeks of clomiphene treatment.

Only one of nine patients (patient #1) responded to clomiphene treatment, with increases in testosterone and estradiol levels. In this patient, serum SHBG and free testosterone levels, evaluated at baseline and after 12 weeks of clomiphene treatment, also increased from 32 nmol/L to 83 nmol/L and from 2.2 ng/dL (0.07 nmol/L) to 9.4 ng/dL (0.32 nmol/L), respectively. After 12 weeks of clomiphene withdraw, total testosterone decreased to pretreatment levels [270 ng/dL (9.45 nmol/L)].

Table 1. Baseline characteristics of nine male patients with NFPA and persistent hypogonadism

Patient	1	2	3	4	5	6	7	8	9
Clinical presentation at diagnosis									
Age	46	55	60	40	38	49	62	37	37
Initial complaint* (years)	H (1.5)	H (1)	VL (1.5)	H (2)	VL (0.5)	VL (1)	LL (1.5)	VL (8)	VL (3)
Hypogonadism (years)	0.6	4	nr	2	2	0.5	1.5	nr	3
Visual impairment**	4	6	4	4	6	4	2	6	7
Tumor size*** (cm)	2	4.5	3.9	MA	2	3.8	4	3	MA
Previous treatment									
TS	2	1	1	1	3	2	1	2	2
TC	N	N	N	N	2	N	N	1	N
RT	N	N	N	Y	Y	Y	N	Y	Y
Previous hormone replacement									
Testosterone	Y	N	Y	Y	Y	Y	Y	Y	Y
Thyroxine	Y	Y	Y	Y	Y	Y	Y	Y	Y
Corticosteroid	N	Y	N	N	N	Y	Y	Y	Y
Baseline prolactin****	13.9	7.3	1	14.3	6.7	9.1	1.3	3.5	8

* Duration of major symptom until diagnosis; ** Number of impaired quadrants of the visual field; *** Largest diameter at diagnosis; **** Unit ng/mL; normal range: 2,1-17,7 ng/mL.
H: headache; VL: visual loss; LL: loss of libido; MA: macroadenoma (tumor size not available); TS: transsphenoidal; TC: transcranial; RT: radiotherapy; nr: not reported; y: yes, n: no.

Table 2. Results for serum testosterone, estradiol, LH and FSH levels in male patients with NFPA and persistent hypogonadis

Patient	Total testosterone (ng/dl)					Estradiol (pg/ml)				
	basal	on clomiphene			off	basal	on clomiphene			off
	0	4 wks	8 wks	12 wks	24 wks	0	4 wks	8 wks	12 wks	24 wks
1	224	631	486	810	270	57.3	38.4	51.1	83.8	29.2
2	82.3	53.1	59.5	58.5	68.5	16.5	15.5	10	10	68.5
3	152.3	117.8	94.2	74.9	89.3	10	13.8	10	19.7	89.3
4	52.5	31	13.8	25.8	42.3	10	11.6	11.2	26.1	42.3
5	206	201.8	169.4	171.6	167.8	12	17.6	10	27.2	167.8
6	246.5	71	65	48.1	114.8	16.7	26.7	15	11.7	114.8
7	157.7	86.9	83.9	81.9	435.6	10	16.5	10.9	10	435.6
8	87.1	61.3	38.4	26.3	18.5	13	18.8	10	10	18.5
9	249.3	285.1	262.5	187.1	242.7	42.8	40.1	21.5	42.9	242.7
mean ± se	154 ± 27	113 ± 31	97 ± 29	84 ± 22	147 ± 48	16 ± 4	20 ± 3	12 ± 1	19 ± 4	17 ± 2.4
P value	0.002				0.03*	0.2				0.8
Patient	**LH (mU/ml)**					**FSH (mU/ml)**				
	basal	**on clomiphene**			**off**	**basal**	**on clomiphene**			**off**
	0	4 wks	8 wks	12 wks	24 wks	0	4 wks	8 wks	12 wks	24 wks
1	3.1	7	6.2	5.8	3	4.3	4.2	6.9	5.7	4
2	0.9	0.9	0.5	0.4	1.4	1.6	0.9	1.1	0.3	1.6
3	0.1	0.6	1.3	0.7	1.2	0.2	0.3	0.5	1.9	3.2
4	0.5	0.3	0.4	0.2	0.3	0.3	0.3	0.3	0.4	0.9
5	1	1.4	1.5	1.9	1.4	0.3	0.3	0.7	2.4	1.7
6	1.2	0.9	1	1.1	1.3	9.3	13.9	11	13.2	17.5
7	1.2	1	0.8	0.7	6.8	1.7	1.6	1.1	1.9	2
8	0.4	0.2	0.3	0.3	0.1	0.3	0.3	0.3	1.3	0.4
9	1.3	3	3.1	3.4	2.8	2.1	2.4	1.5	1.5	3.1
mean ± se	0.8 ± 0.2	1 ± 0.3	1.1 ± 0.3	1.1 ± 0.4	1.9 ± 0.7	1.9 ± 1	2.6 ± 1.6	2.4 ± 1.5	2.8 ± 1.5	3.8 ± 1.9
P value	0.7				0.54	0.3				0.1

Body composition, serum glucose, insulin, cholesterol and triglycerides during clomiphene treatment

In the eight unresponsive patients, BMI tended to increase from 27.5 ± 0.6 to 27.7 ± 0.6 kg/m^2 (P = 0.05). Fat mass increased significantly, whereas lean body mass, waist circumference and bone mineral density remained unchanged after clomiphene treatment. Fasting serum glucose tended to decrease from 92 ± 2 to 85 ± 3 mg/dl (P = 0.07), fasting insulin decreased from 18 ± 4 to 12 ± 3 µU/mL (P < 0.05), HOMA-IR decreased from 4 ± 0.8 to 2.5 ± 0.6 (P < 0.05), and total cholesterol decreased from 214 ± 12 to 193 ± 10 mg/dL after clomiphene treatment in unresponsive patients, but no significant changes were observed in serum LDL, HDL and triglycerides, or in glucose and insulin levels at 120 minutes after the oral glucose tolerance test (OGTT).

DISCUSSION

In this study, we have shown that chronic administration of clomiphene citrate failed to recover gonadal function in most male patients with conventionally treated NFPA and persistent hypogonadotropic hypogonadism. This finding is in sharp contrast to the high response rate (71%) we have recently observed in male hypogonadal patients with prolactinomas, treated with clomiphene using the same study protocol (15). Clomiphene failure to restore gonadal function in most patients with treated NFPA is likely to reflect a much lower functional reserve of gonadotrophic cells and/or GnRH neurons in these patients, as compared with prolactinoma patients (20). In fact, because of the poor efficacy of currently available medical treatment for NFPA, as opposed to the high efficacy of dopamine agonists in prolactinomas, NFPA patients are frequen-

Table 3. Results for glucose, insulin, body composition, cholesterol and triglycerides in nine male patients with NFPA and persistent hypogonadism

Patient	Fasting glucose (mg/dl)		OGTT 120' glucose (mg/dl)		Fasting insulin (µU/ml)		OGTT 120' insulin (µU/ml)		IR HOMA		ISI Matsuda	
	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC
1	121	94	108	155	99.0	25.0	110.0	163.0	29.8	5.8	0.7	1.7
2	99	85	123	151	11.7	7.8	60.0	80.3	2.9	1.6	4.1	4.1
3	93	75	80	64	27.0	10.2	42.4	10.8	6.2	1.9	2.7	8.5
4	93	100	86	82	7.6	4.4	40.8	40.1	1.7	1.1	4.5	5.9
5	94	81	129	106	36.5	30.9	285.2	65.3	8.5	6.2	1.0	3.5
6	94	90	143	102	6.7	7.5	85.3	64.6	1.6	1.7	5.0	5.5
7	96	81	193	130	13.9	15.4	176.0	106.4	3.3	3.1	1.9	2.1
8	93	86	105	109	16.2	9.9	46.2	44.6	3.7	2.1	3.3	3.6
9	78	84	74	80	21.3	12.0	59.2	65.2	4.1	2.5	2.8	5.1
mean (se)*	92 ± 2	85 ± 3	116 ± 14	103 ± 10	18 ± 3.6	12 ± 2.9	99 ± 31	60 ± 10	4 ± 0.8	2.5 ± 0.6	3.1 ± 0.5	4.8 ± 0.7
P value	0.07		0.2		0.03		0.2		0.02		0.05	

Patient	Fat mass		Muscle mass		Total cholesterol (mg/dl)		LDL cholesterol (mg/dl)		HDL cholesterol (mg/dl)		Triglycerides (mg/dl)	
	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC	baseline	after CC
1	26.5	30.1	65.9	71.8	219.0	155.0	133.0	80.0	36.0	34.0	250.0	207.0
2	14.2	16.0	56.3	57.4	224.0	233.0	144.0	144.0	40.0	43.0	198.0	228.0
3	20.1	22.1	73.1	68.9	145.0	136.0	93.0	81.0	35.0	41.0	84.0	68.0
4	26.9	29.7	56.8	55.0	214.0	179.0	130.0	113.0	45.0	41.0	196.0	123.0
5	25.0	27.7	59.8	60.6	200.0	177.0	117.0	101.0	58.0	53.0	126.0	117.0
6	21.7	21.1	63.9	63.5	255.0	219.0	**	**	52.0	51.0	494.0	508.0
7	18.7	20.2	54.4	53.9	226.0	197.0	**	92.0	37.0	30.0	597.0	376.0
8	25.8	27.6	61.0	59.7	204.0	204.0	131.0	120.0	33.0	51.0	199.0	208.0
9	20.9	20.6	55.5	55.4	250.0	203.0	133.0	80.0	62.0	64.0	135.0	96.0
mean (se)*	22 ± 1	23 ± 2	60 ± 2	59 ± 2	214 ± 12	193 ± 10	124 ± 7	107 ± 8	45 ± 4	46 ± 4	254 ± 66	215 ± 54
P value	0.02		0.25		0.02		0.06		0.9		0.2	

CC: clomiphene; IR: insulin resistance index; ISI: insulin sensitivity index; OGTT: oral glucose tolerance test.

tly submitted to multiple surgeries and radiotherapy (1-5), increasing the risk of irreversible pituitary damage.

In unresponsive patients, clomiphene not only failed to increase, but also significantly decreased testosterone levels. This reduction may be due the inhibitory effect of clomiphene on LH pulsatility and/or testicular androgen biosynthesis, since there is no variation in LH levels (21). Clomiphene is an estrogen receptor agonist/antagonist, but its ultimate *in vivo* effects are dependent on the dose, target tissue and endogenous estrogenic activity (9). In the hypothalamus-pituitary axis, at low doses (25 to 50 mg/day), clomiphene is reportedly ineffective in prepubertal boys, but stimulates gonadotropin secretion in pubertal and adult males. At high doses (500 mg/day), clomiphene suppresses gonadotropin secretion even in normal adult males (22,23).

Decreased testosterone in unresponsive patients, *per se*, should impair insulin sensitivity. However, improved insulin sensitivity was observed during clomiphene treatment. This observation taken together with the decrease in testosterone and increase in SHBG levels, reinforce the predominance of clomiphene estrogenic activity in these patients. In fact, estrogen increases insulin sensitivity in males with aromatase deficiency (24,25). Improved insulin sensitivity in persistent hypogonadal males during clomiphene therapy is further evidence that, in males, testosterone mediates insulin sensitivity by means of estrogen receptors.

In the only responsive patient, gonadotropin levels increased and testosterone levels reached the normal range 10 days after clomiphene therapy started, and remained stable thereafter. This response was similar to the one previously described in male patients with

prolactinomas under dopamine agonist treatment (15). Although this patient did not receive radiotherapy, he had been submitted to pituitary surgery twice. Nonetheless, a critical hypothalamic-pituitary reserve was able to respond to clomiphene treatment. Considering the low response rate and the size of the sample we could not identify predictive factors related to the response to clomiphene.

In conclusion, most patients with conventionally treated NFPA did not have their normal testosterone levels restored during clomiphene therapy. Additional studies with larger number of patients may identify factors that predict the response to clomiphene in patients with NFPA and hypogonadism. In addition, clomiphene improved insulin sensitivity even with the decrease in testosterone levels.

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