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Mestrado e Doutorado

AVALIAÇÃO DA FASE LÚTEA E DA SECREÇÃO DE PROLACTINA E DO HORMÔNIO DO CRESCIMENTO APÓS TESTES DE ESTÍMULO COM O HORMÔNIO LIBERADOR DA TIREOTROPINA (TRH) E METOCLOPRAMIDA EM PACIENTES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA E LEVE

DR. JOÃO SABINO LAHORGUE DA CUNHA FILHO

Orientador: Prof. Dr. Jorge Luiz Gross

Co-orientador: Prof. Dr. Eduardo Pandolfi Passos

Porto Alegre, novembro de 2000

João Sabino Lahorgue da Cunha Filho

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2000

Dedico este trabalho

ao meu pai, mãe e irmão

por me ensinarem a amar e,

à Luciana que eu amo muito.

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SUMÁRIO

LISTA DE ABREVIATURAS

°C	graus Celsius
IMC	índice de massa corporal
m^2	metroquadrado
ml	mililitro
mUI	miliUnidades internacionais
µl	microlitro
µUI	microUnidades internacionais
ng	nanograma
pg	picograma
rpm	rotações por minuto
kg	quilograma
FSH	hormônio folículo estimulante
LH	hormônio luteinizante
TSH	hormônio tireotrófico
GH	hormônio do crescimento
PRL	prolactina

RESUMO

Endometriose está relacionada com infertilidade e, nas formas leves ou mínimas, sem dano tubário, a causa da incapacidade reprodutiva ainda não está esclarecida. Vários mecanismos são propostos para este grupo de mulheres, tais como alterações imunológicas, peritoneais e hormonais que podem acarretar em disfunções foliculares e oocitárias.

Distúrbios na fase lútea e na secreção de prolactina podem associar-se a infertilidade e endometriose, porém com resultados conflitantes. Já o hormônio do crescimento associa-se com a síntese de esteróides e pode ter uma ação co-gonadotrófica, sua secreção pode relacionar-se com o futuro reprodutivo em algumas mulheres.

O presente estudo teve como objetivo o estudo da fase lútea e da secreção de prolactina e hormônio do crescimento após estímulo com *TRH* e metoclopramida em mulheres inférteis com endometriose mínima e leve se comparadas as mulheres férteis com e sem endometriose.

Pacientes com endometriose e infertilidade apresentaram níveis séricos de estradiol inferiores com um aumento na prevalência de hiperprolactinemia e insuficiência lútea, diagnosticada por biópsia endometrial ou progesterona sérica. A presença de endometriose relacionou-se com aumento nos níveis séricos basais de prolactina, 15 e 30 minutos após infusão de *TRH*. Após a administração de metoclopramida, os níveis séricos do hormônio do crescimento e da prolactina não foram diferentes entre os grupos.

Mulheres férteis com endometriose apresentaram níveis séricos do hormônio do crescimento superiores se comparados com mulheres férteis sem endometriose 15 minutos após infusão de *TRH*.

Estes resultados indicam uma alteração na secreção de prolactina e estrógenos em mulheres com infertilidade e endometriose, provavelmente repercutindo em uma alteração na fase lútea comprovada por biópsia endometrial e progesterona sérica. Estas alterações hormonais relacionam-se e podem explicar a causa da infertilidade neste grupo de mulheres sem dano tubário.

INTRODUÇÃO

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Aspectos gerais

Em 1927, SAMPSON foi o primeiro a caracterizar a endometriose como uma entidade patológica responsável por alterações na pelve feminina. Esta afecção foi definida como a presença de tecido glandular e estroma endometriais fora da cavidade uterina (OLIVE & SCHWARTZ, 1993; KONINCKX, 1994; NISOLLE & DONNEZ, 1997).

Estima-se que cerca de 10% a 15% da população feminina apresente esta doença (MUSE & WILSON, 1982; BANCROFT, WILLIAMS, ELSTEIN, 1989; OLIVE & SCHWARTZ, 1993). É freqüente a associação entre endometriose e infertilidade. Trinta a sessenta por cento das pacientes com endometriose apresentam infertilidade associada (OLIVE & SCHWARTZ, 1993; BANCROFT, WILLIAMS, ELSTEIN, 1989; KONINCKX, 1994).

No Serviço de Ginecologia e Obstetrícia do Hospital de Clínicas de Porto Alegre, endometriose foi o principal achado laparoscópico em pacientes com dor pélvica e o segundo diagnóstico entre as com infertilidade (PALMA DIAS et al., 1995).

Vários mecanismos têm sido propostos para explicar a infertilidade em mulheres com endometriose, os distúrbios ovulatórios foram estudados por vários autores (TUMMON et al., 1988; GARCIA-VELASCO & ARICI, 1999; CAHILL & HULL, 2000; GARRIDO et al., 2000) que encontraram uma prevalência de alterações ovulatórias em mulheres com endometriose mínima semelhante às pacientes com infertilidade de causa desconhecida. Em 1992, BANCROFT, WILLIAMS, ELSTEIN (1992) descreveram alterações no eixo pituitária-ovariano em 82% das pacientes com endometriose e infertilidade, destacando as alterações de secreção de *LH,* com prováveis conseqüências na alteração da maturidade oocitária.

Alterações na foliculogênese e maturação oocitária são evidentes em diversos estudos que analisaram a secreção de hormônios pituitários, esteróides ou a atividade das células da granulosa em mulheres inférteis com endometriose mínima ou leve (CAHILL et al., 1995; HARLOW et al., 1996; TOYA et al., 2000; CAHILL & HULL, 2000; GARRIDO et al., 2000). Estes últimos autores também discutem a possibilidade de existir alguma interferência da endometriose sobre o prognóstico reprodutivo em mulheres submetidas a fertilização *in vitro*.

Entretanto, o grupo controle utilizado nestes estudos é bastante heterogêneo, carecendo de uma comparação com pacientes férteis com endometriose, e os achados explicariam parcialmente a infertilidade em pacientes sem dano anatômico importante (endometriose mínima ou leve), uma vez que

para as formas mais graves desta doença, segundo a *American Society for Reproductive Medicine,* o dano tubário impede a gestação (TOYA et al., 2000; CAHILL & HULL, 2000).

Endometriose e insuficiência lútea

A insuficiência lútea foi descrita pela primeira vez em 1949 por JONES, e até hoje seu diagnóstico e tratamento são muito discutidos. Sabe-se que a insuficiência lútea associa-se com alterações da fase folicular e lútea, tendo sua expressão final um suporte inadequado de progesterona para a segunda fase do ciclo. A disfunção lútea pode dever-se a alterações de pulsatilidade de gonadotrofinas, diminuição dos níveis de *FSH*, secreção de inibina, defeito de células da granulosa, hiperprolactinemia, pico anômalo de *LH* e defeitos endometriais, entre outros (GINSBURG, 1992).

Pacientes com insuficiência lútea teriam uma prevalência aumentada de aborto habitual e infertilidade, porém nenhum estudo com grupo controle conseguiu esclarecer a real prevalência desses eventos na população. Além disto, não há estudo bem delineado que constate a redução da incidência de aborto ou infertilidade das pacientes com diagnóstico de insuficiência lútea tratadas de forma adequada (GINSBURG, 1992).

A medida sérica de progesterona só tem validade se for realizada em diversas coletas (mínimo de 3 medidas) e fornece informações quanto à secreção, não informando sobre a ação periférica desse hormônio. FILICORI, BUTLER, CROWLEY (1984) mostraram também que a dosagem de progesterona pode variar em até 10 vezes no intervalo de 2 a 3 horas.

A biópsia de endométrio ainda é o teste mais usado e considerado padrão ouro, apesar de não ser ideal (STRAUSS III & COURTIFARIS, 1998; SPEROFF et al., 1999) para o diagnóstico de insuficiência lútea. O diagnóstico é feito quando há uma discrepância de 2 a 3 dias entre o achado histológico e o ciclo menstrual da paciente. A biópsia deverá ser feita no fundo uterino (zona do endométrio de maior atividade endócrina) com uma sonda de aspiração ou cureta de Novak, sempre após o 23º dia do ciclo (PASSOS et al., 1997).

BATISTA et al. (1993) e JORDAN et al. (1994) criticaram severamente o uso da biópsia de endométrio. Para o último grupo de autores, uma medida de progesterona inferior a 10 ng/ml ou a soma de 3 medidas abaixo de 30ng/ml seria uma maneira mais correta de fazer o diagnóstico de fase lútea inadequada, mas recomenda o uso da biópsia para o acompanhamento e tratamento subseqüente.

HARGROVE & ABRAHAM (1980) tentaram associar endometriose com síndrome pré-menstrual, mostrando que ambas têm o perfil hormonal semelhante: aumento da prevalência de hiperprolactinemia e insuficiência lútea. BALLACH &

VANRELL (1985) refutaram essa idéia e, estudando pacientes com infertilidade e endometriose leve e pacientes férteis e inférteis sem endometriose, não encontraram diferenças significativas nas comparações das medidas de *PRL*, progesterona, estradiol e avaliação da fase lútea por biópsia endometrial. Apenas 18,5% das pacientes com endometriose e infertilidade tiveram o diagnóstico de insuficiência lútea.

PITTAWAY et al. (1983), MOLOEK & MOEGNY (1993) e MATORRAS et al. (1996) usando biópsia de endométrio, não encontraram diferença significativa entre as pacientes com e sem endometriose para o diagnóstico de insuficiência lútea usando como grupo controle mulheres inférteis de causa desconhecida.

KUSUHARA (1992) comparou 24 pacientes inférteis com endometriose e 20 com infertilidade de causa desconhecida. A fase lútea foi avaliada por meio de medida de progesterona, não sendo observada diferença significativa. AYERS, BIRENBAUM, MENON (1987) concluíram que existe uma evidência que aponta para uma alteração na segunda fase do ciclo menstrual em pacientes com endometriose mínima ou leve, podendo explicar a infertilidade. A alteração dá-se na secreção de progesterona e estradiol, com provável alteração na modulação de *LH* e *FSH* e na receptividade desses hormônios em nível de membrana celular. As alterações não se restringem apenas à formação do corpo lúteo, mas, também, a sua atresia e relação com o início de um novo ciclo menstrual.

Vários outros autores, WILLIAMS, OAK, ELSTEIN (1986), AYERS, BIRENBAUM, MENON (1987), TUMMON et al. (1988) e BANCROFT, WILLIAMS, ELSTEIN (1992), investigaram a secreção de progesterona em mulheres inférteis com endometriose, destacando um padrão anômalo com provável alteração de fase lútea. CHEESMAN et al. (1982) e CHEESMAN et al. (1983) já haviam destacado uma provável alteração na 2ª fase do ciclo neste grupo de mulheres, a importância destes achados foi enfatizada por GARCIA-VELASCO E ARICI (1999) relatando a importância clínica e dificuldade, epidemiológica e metodológica, no estudo da função lútea de mulheres inférteis com endometriose.

Endometriose e secreção de prolactina

Desde os trabalhos de SEPPALA, HIRVONEN, RANTA (1976) e DELPOZO et al. (1979), está bem estabelecida a relação entre hiperprolactinemia com insuficiência lútea e infertilidade.

HIRSCHOVITZ, SOLER, WORTSMAN (1978) foram os primeiros a descrever a síndrome endometriose-galactorréia em 8 pacientes, das quais apenas uma apresentava níveis de prolactina (*PRL)* alterados. MUSE, WILSON, JAWAD (1982) aventaram a possibilidade de a hiperprolactinemia ser a responsável ou, pelo menos, ter algum efeito na infertilidade das pacientes com endometriose. Outros autores (CORENBLUM & TAYLOR, 1980) demonstraram que algumas pacientes com galactorréia e irregularidade menstrual poderiam ter

níveis de *PRL* normais, porém, quando estimuladas com *TRH*, apresentavam uma resposta anômala. Corroborando com estes últimos, vários autores acreditam que a secreção anômala de *PRL* em pacientes com infertilidade e endometriose possa ser um achado importante (MUSE, WILSON, JAWAD, 1982; ACIÉN, LLORET, GRAELLS, 1989). Esses achados são contestados por (MATALLIOTAKIS et al., 1996; MACHIDA, TAGA, MINAGUCHI, 1997) que não encontraram associação entre a secreção anômala de *PRL* em mulheres com endometriose mínima ou leve, mesmo após estímulo com *TRH* neste grupo de mulheres.

O *TRH* exerce um aumento na secreção de *PRL* por uma ativação direta de receptores de membrana celular, ativando a proteína-quinase-c com liberação de cálcio intra-celular e secreção de *PRL* do retículo endoplasmático (YEN, 1998) (Figura 1).

HE (1993) registrou prevalência de hiperprolactinemia estatisticamente superior em pacientes com infertilidade e endometriose (61,5%). Em estudo com 41 mulheres (férteis com e sem endometriose e inférteis com endometriose), CUNHA-FILHO et al., 2000 relatam uma prevalência aumentada de hiperprolactinemia (30%) e insuficiência lútea (78,9%) em mulheres inférteis com endometriose. Os autores destacam que, em um modelo de regressão logística múltipla, apenas a presença de endometriose e infertilidade associou-se a hiperprolactinemia e alterações lúteas. Outros autores encontraram uma medida

basal de prolactina duas vezes maior nessas pacientes, porém sem significância estatística (MUSE, WILSON, JAWAD, 1982; ACIÉN, LLORET, GRAELLS, 1989).

Recentemente, GREGORIOU et al., 1999 também estudaram a associação de endometriose e níveis séricos de prolactina após estímulo com *TRH* em pacientes inférteis. Estes autores destacam que mulheres com endometriose e infertilidade tiveram uma resposta ao *TRH* superior se comparadas as férteis sem endometriose. Embora concluindo que 31% das mulheres com endometriose, de todos os estádios, possuam uma alteração na secreção de *PRL* após *TRH,* HAYASHI, TAKETANI, MIZUNO, 1989 não encontraram diferença em relação ao grupo controle formado por mulheres inférteis sem endometriose, talvez pela utilização de mulheres inférteis como grupo-controle, uma vez que a prevalência de hiperprolactinemia oculta também foi elevada neste grupo de mulheres.

Em 1993, ASUKAI, UEMURA, MINAGUCHI já haviam descrito a hiperprolactinemia oculta como a presença de uma alteração hormonal caracterizada pela presença de níveis séricos basais de *PRL* normais porém, após estímulo com *TRH*, estas pacientes apresentaram uma resposta secretora acentuada. Esta alteração na secreção da *PRL* pode levar a distúrbios de foliculogênese por mecanismos centrais que modulam a liberação de *LH*, estradiol e testosterona (KOSTAL & TOSNER, 1997), provocando infertilidade e insuficiência lútea. Testes de estímulo com *TRH* podem ser utilizados para o diagnóstico de alterações hormonais mais sutis e selecionar aquelas pacientes

com alterações de liberação de *PRL* para tratamento com um análogo dopaminérgico, como propôs STEINBERGER et al., 1990.

A pulsatilidade noturna da *PRL* foi estudada por RADWANSKA, HENIG, DMOWSKI, 1987 em mulheres inférteis com endometriose comparadas a outras mulheres com infertilidade. Pacientes com infertilidade e endometriose de diferentes graus tiveram a secreção de *PRL* superior ao grupo controle.

A metoclopramida pode ser utilizada também para a indução da secreção de *PRL*, porém, ao invés de estimular a proteína-quinase c do lactotrófo, a sua ação é pela inibição da ação da dopamina (ALBIBI et al., 1983). Estudando sete mulheres normais do 2° até o 22° dias de seus ciclos menstruais (KAUPPILA et al., 1982) e administrando metoclopramida durante a fase folicular precoce, os autores concluíram existir uma série de alterações na maturação folicular e segunda fase do ciclo decorrentes da hiperprolactinemia provocada pela metoclopramida. Outros autores, estudando a liberação de *PRL* após 2,5mg de metoclopramida pela via oral em mulheres com hiperprolactinemia, concluíram existir um bloqueio na secreção de *PRL* provavelmente secundário a algum distúrbio regulatório central da dopamina (VASQUEZ-MATUTE et al., 1979; SERRI et al., 1988).

Na caracterização da hiperprolactinemia oculta, AISAKA et al., 1987 utilizaram o teste com metoclopramida e com *TRH* e estabeleceram uma relação

entre a secreção de *PRL* após a inibição dopaminérgica central pela metoclopramida com os níveis séricos de prolactina após administração de *TRH*. Geralmente, a resposta após a metoclopramida é 4x superior a resposta após a infusão de *TRH,* mostrando a importância da modulação central da dopamina na liberação da *PRL*.

Endometriose e hormônio do crescimento

O hormônio do crescimento (*GH*) é secretado pela hipófise anterior e constitui cerca de 35-45% de sua massa em adultos, porém, apesar desta abundância, sua função após a puberdade é bastante discutida (KATZ, RICCIARELLI, ADASHI, 1993).

Este hormônio atuaria como co-gonadotrófico, aumentando a ação gonadotrófica, sendo incapaz de atuar sozinho. MASON et al., 1990 demonstraram que as células da granulosa, quando incubadas com *GH* apresentaram um aumento de secreção de estradiol, achados confirmados por BARRECA et al., 1993.

Outros autores (MENASHE et al., 1990) observaram existir uma associação entre a resposta exacerbada ao teste de estímulo com clonidina para secreção de *GH* e o futuro reprodutivo destas pacientes, OVENSEN et al., 1994 também concluíram que o *GH* estimula a esteroidogênese e atua como importante

modulador da função ovariana, associando infertilidade e anovulação com distúrbios na secreção do *GH*. Recentemente, ROSSATO et al., 2000 estudaram pacientes anovulatórias que não responderam ao tratamento com citrato de clomifeno e apresentaram uma diminuição da resposta do *GH* após administração de *TRH* neste grupo de mulheres.

O controle da secreção do *GH* é complexo e envolve várias substâncias neuroendócrinas, sua secreção está ligada aos hormônios pituitários e hipotalâmicos edistúrbios na sua secreção podem associar-se a função reprodutiva (DEVESA et al., 1992; KATZ, RICCIARELLI, ADASHI, 1993).

A modulação da secreção do *GH* é feita por diversos mecanismos (colinérgico, serotoninérgico e dopaminérgico), além da ação do hormônio liberador de *GH* (*GHRH*) e seu inibidor, a somatostatina (*SS*), ambos peptídeos liberados pelo hipotálamo (ARCE et al., 1991; KATZ, RICCIARELLI, ADASHI, 1993) (figura 2).

A metoclopramida pode ser utilizada como teste de estímulo para a secreção do *GH*, tendo ação anti-dopaminérgica central e, segundo MASSARA et al., 1985, também pode modular a secreção do *GH* pelos eixos colinérgicos e serotoninérgicos.

Alguns autores, estudando a secreção do *GH* após infusão de metoclopramida (COHEN et al., 1979; CHIODERA et al., 1982) concluíram que havia um aumento na liberação de *GH*, provavelmente secundária a uma ativação dopaminérgica ou inibição da *SS*, teoria aventada por ARCE et al., 1991.

A dopamina teria uma ação inibitória sobre a *SS* e, consequentemente ativaria a secreção de *GH* (MASSARA et al., 1985 ; VANCE et al., 1987), podemos especular que uma alteração na modulação dopaminérgica poderia estar associada a um distúrbio na secreção basal do *GH* ou após estímulo.

Entretanto, MASALA et al., 1977 e JORDAN et al., 1986 refutam esta hipótese e não demonstraram nenhuma alteração na secreção do *GH* após metoclopramida, estes dois últimos autores estudaram apenas homens com idade adulta e, MASALA et al., 1977 administrou metoclopramida pela via oral, dificultando a comparação com os demais estudos.

Existindo uma alteração na secreção de *PRL* e um distúrbio no eixo pituitário-ovariano (CAHILL & HULL, 2000) em mulheres inférteis com endometriose, podemos especular que o eixo dopaminérgico-somatotrófico poderia estar alterado neste grupo de mulheres inférteis, alterando, também a secreção do *GH*, com conseqüente distúrbio ovulatório.

O *TRH* teria um efeito ativatório na secreção do *GH* apenas em condições clínicas especiais tais como anorexia nervosa, depressão, acromegalia ou doença hepática (PENERAI et al., 1977; ISHIBASHI & YAMAJI, 1978; KALTSAS et al., 1999). Entretanto, outros autores associaram a Síndrome dos Ovários Policísticos (SOP) a um distúrbio no eixo somatotrófico, com uma disfunção na liberação do *GH* após estimulo com *TRH* (ANAPLIOTOU et al., 1989; KALTSAS et al., 1999). PIADIDIS et al., 1995 estudando a secreção de *GH* após infusão de dopamina em pacientes com SOP relacionou uma alteração na secreção deste hormônio com a SOP. Estas alterações descritas entre a SOP e a secreção de *GH* podem contribuir para a infertilidade destas pacientes por alterar os mecanismos de regulação do eixo hipotálamo-hipófise-ovário.

A modulação da secreção do *GH* após infusão de *TRH* pode ser devido a um estímulo direto nas células pituitárias (somatotrófos) ou por uma modulação hipotalâmica, uma vez que o *TRH* é secretado pelo núcleo paraventricular, mesma região anatômica que produz e libera a *SS*.

É evidente a relação entre endometriose e infertilidade, porém não há indícios, na literatura médica, das razões pelas quais pacientes com endometriose mínima ou leve, sem alteração anatômica, não consigam gestar. Uma série de alterações na foliculogênese, maturação oocitária, fertilização, implantação embrionária e suporte lúteo são descritas com resultados discordantes. Como a fase lútea, a *PRL* e o *GH* estão intimamente relacionados ao processo reprodutivo

feminino e as alterações de suas secreções podem nos ajudar a explicar a infertilidade em mulheres com endometriose sem dano anatômico, resolvemos estudar a fase lútea e a liberação da *PRL* e do *GH* em pacientes inférteis com endometriose mínima e leve.

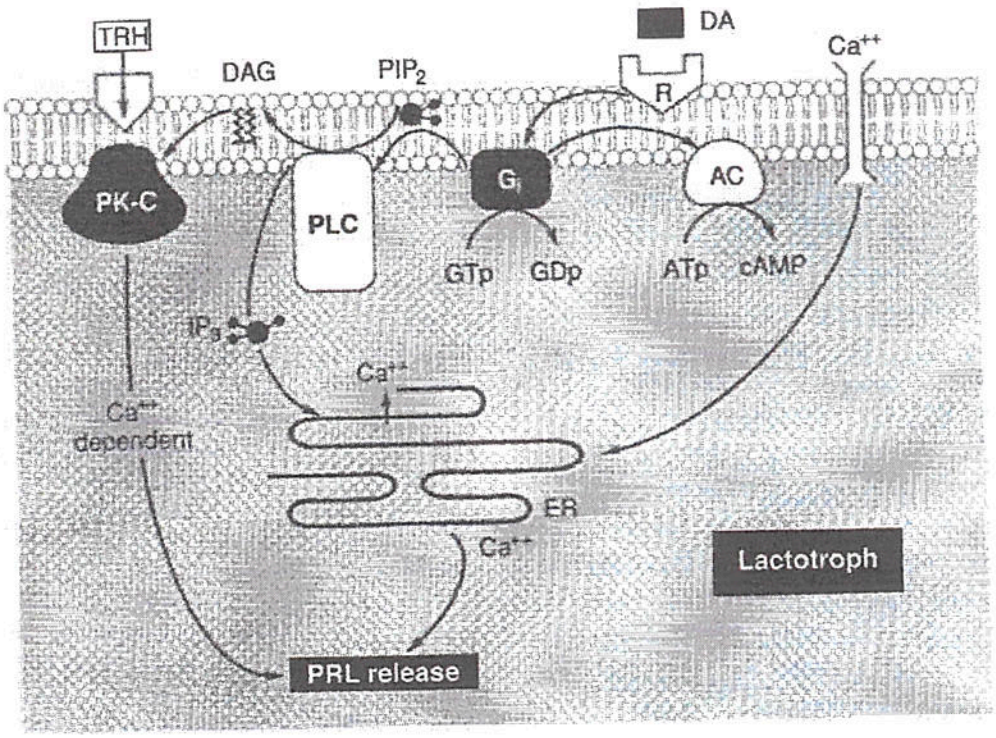

Figura 1: ilustração esquemática da liberação da prolactina.
Ação inibitória da dopamina (DA) pela atividade da proteína G_1 que inibe a adenilato ciclase (AC), AMPc e fosfolipase C (PLC).
A ação ativatória do *TRH* na liberação de prolactina ocorre com indução da proteína cinase C (PK-C) com liberação, cálcio (Ca^{++}) dependente, da prolactina pelo lactotrófo.
ER, retículo endoplasmático.
DAG, diacilglicerol.
PIP2, inositol difosfato.
PIP3, inositol trifosfato.
Adaptado de YEN, JAFFE, BARBIERI (1998).

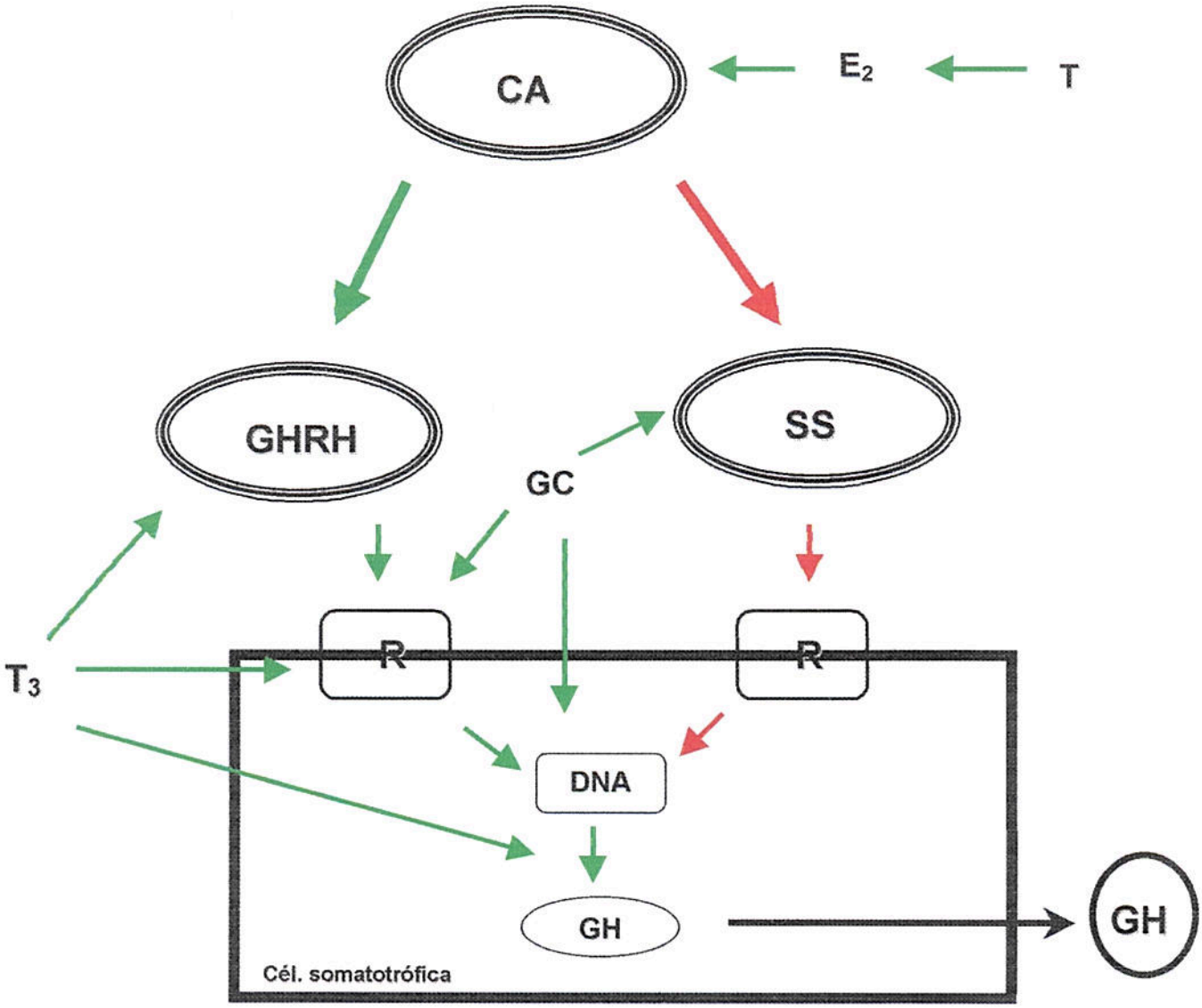

Figura 2: Controle da liberação do GH.
Setas em verde representam ação facilitatória, em vermelho inibitória.
CA, catecolaminas
GHRH, hormônio liberador do GH
SS, somatostatina
R, receptor de membrana
GC, glicocorticóides
E_2, estrógenos
T, testosterona
Adaptado de DEVESSA, LIMA E TRESGUERRES (1992).

OBJETIVOS

- AVALIAR A FASE LÚTEA, PELA BIÓPSIA ENDOMETRIAL E MEDIDA SÉRICA DE PROGESTERONA, EM MULHERES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA OU LEVE;

- AVALIAR A SECREÇÃO DE PRL APÓS TESTES DE ESTÍMULO COM TRH E METOCLOPRAMIDA EM MULHERES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA OU LEVE;

- AVALIAR A SECREÇÃO DO GH APÓS INFUSÃO DE TRH E METOCLOPRAMIDA EM MULHERES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA OU LEVE;

O corpo desta tese é composto por dois artigos originais, contendo os resultados e a discussão sobre o estudo da secreção da prolactina e hormônio do crescimento após testes de estímulo e a análise da fase lútea em mulheres inférteis com endometriose mínima e leve.

O primeiro artigo foi aceito para publicação na *Hormone Metabolic Research* em outubro deste ano e tem como objetivo a análise da função lútea neste grupo de mulheres.

O segundo manuscrito estudou as secreções do hormônio do crescimento e da prolactina após administração de TRH e metoclopramida, também contemplando pacientes com infertilidade e endometriose mínima e leve. Este último artigo foi enviado para publicação no *J Clin Endocrinol Metab* em novembro de 2000.

REFERÊNCIAS BIBLIOGRÁFICAS

ACIÈN, P.; LLORET, M.; GRAELLS, M. - Prolactin and its response to the luteinizing hormone-releasing hormone thyrotropin-releasing hormone test in patients with endometriosis before, during and after treatment with danazol. **Fertil. Steril., 51**: 774-80, 1989.

AISAKA, K.; ANDO, S.; KOKUHO, K.; KANEDA, S.; TAWADA, T.; YOSHIDA, K.; MORI, H. – Role of TRH or metoclopramide loading test for the diagnosis of so-called occulted of latent hyperprolactinemia. **Folia Endocrinolol., 63**:853-61, 1987.

ALBIBI, R.; MCCALLUM, W. – Metoclopramide: pharmacology and clinical application. **Annals of Intern. Med., 98**: 86-95, 1983.

ANAPLIOTOU, M.L.G.; SYRIGOS, K.; PANAYIOTAKAPOULOS, G.; KALOS, A.; BATRINOS, M.L. Paradoxical growth hormone response following thyroid-stimulating hormone administration in the polycystic ovary syndrome. **Horm. Res., 31**: 119-24, 1989.

ANDERSEN, A.N.; TABOR, A. – Prl, TSH, GH and LH responses to metoclopramide and breast-feeding in normal and hyperprolactinemic women. **Acta Endocrinologica, 100**: 177-83, 1982.

ARCE, V.; LIMA, L.; LOIS, N.; RODRIGUEZ, A.; DIAZ, M.J.; TRESGUERRESJ.A.F.; DEVESA, J. – Role of central dopaminergic pathways in the neural control of growth hormone secretion in normal men: studies with metoclopramide. **Neuroendocrinology, 53**: 143-9, 1991.

ASUKAI, K.; UEMURA, T.; MINAGUCHI, H. – Occult hyperprolactinemia in infertile women. **Fertil. Steril., 60**: 423-7, 1993.

AYERS, J.W.T.; BIRENBAUM, D.L.; MENON, K.M.J. – Luteal phase dysfunction in endometriosis: elevated progesterone levels in peripheral and ovarian veins during the follicular phase. **Fertil. Steril., 47** (6): 925-929, 1987.

BALLACH, J.; VANRELL, J.A. – Mild Endometriosis and Luteal Function. **Int. J. Fert., 30** (3): 4-6, 1985.

BANCROFT, K.; WILLIAMS, C.A.V.; ELSTEIN, M. - Minimal/mild endometriosis and infertility. A review. **Brit. J. Obstet. Gynaecol., 96**:454-60, 1989.

BANCROFT, K.; WILLIAMS, C.A. V.; ELSTEIN, M. – Pituitary-ovarian functionin women with minimal or mild endometriosis and otherwise unexplained infertility. **Clin. Endocrinol., 36**: 177-181, 1992.

BARRECA, A.; ARTINI, P.G.; DEL MONTE, P.; PONZANI, P.; PASQUINI, P.; CARIOLA, G. – In vivo and in vitro effect of growth hormone on estradiol secretion by human granulosa cells. **J. Clin. Endocrinol. Metab., 77**: 61-7, 1993.

BATISTA, M.; CARTLEDGE, T.P.; MERINO, M.J.; AXIOTIS, C.; PLATIA, M.P.; MERRIAM, G.R.; LORIAUX, D.L.; NIEMAN, L.K. - Midluteal phase endometrial biopsy does not accurately predict luteal function. **Fertil Steril., 59**:294-300, 1993.

CAHILL, D.J.; WARDLE, P.G.; MAILE, L.A.; HARLOW, C.R.; HULL, M.G.R.- Pituitary-ovarian dysfunction as a cause for endometriosis-associated and unexplained infertility. **Hum. Reprod., 10**: 3142-3146, 1995.

CAHILL, D.J.; HULL, M.G.R. Pituitary-ovarian dysfunction and endometriosis. **Hum. Reproduc. Update., 6**: 56-66, 2000.

CHEESMAN, K.L.; BEM-NUN, I.; CHATTERTON, R.T,; COHEN, M.R. – Relationship of luteinizing hormone, pregnanediol-3-glucuronide, and estriol-16- glucuronidein urine of infertile women with endometriosis. **Fertil. Steril., 38** (5): 542-548, 1982.

CHEESMAN, K.L.; CHEESMAN, S.D.; CHATTERTON, R.T.; COHEN, M.R. – Alterations in progesteron metabolism and luteal function in infertile women with endometriosis. **Fertil. Steril., 40** (5): 590-595, 1983.

CHIODERA, P.; COIRO, V.; ZANARDI, G.; VOLPI, R.; VALENTI, G.; BUTTURINI, U. – Effect of metoclopramide on serum GH levels in normal women. **Horm. Metab. Res., 14**: 103-4, 1982.

COHEN, H.N.; HAY, I.D.; THONSON, J.A.; LOGUE, F.; RATCLIFFE, W.A.; BEASTALL, G.H. – Metoclopramide stimulation: a test of growth hormone reserve in adolescents males. **Clinical Endocrinology, 11**: 89-93, 1979.

CORENBLUM, B.; TAYLOR, P.J. - A rationable for the use of bromocriptine in patients with amenorrhea and normoprolactinenia. **Fertil. Steril., 34**:239-43, 1980.

CUNHA-FILHO, J.S.; GROSS, J.L.; CASTILHO, M.; BRANDELLI, A.; LEMOS, N.A.; PASSOS, E.P. – Hyperprolactinemia and luteal insufficiency in infertile patients with mild/minimal endometriosis. Horm. Metab. Res.; In press., 2000.

DELPOZO, E.; WYSS, H.; TOLIS, G.; ALCANIZ, J.; CAMPANA, A .; NAFTOLIN, F. - Prolactin and deficient luteal function. **Obstet. Gynecol., 53**:282-7, 1979.

DEVESA, J.; LIMA, L.; TRESGUERRES, A.F. – Neuroendocrine control of growth hormone secretion in humans. **Trends Endocrinol. Metab., 3**: 175-83, 1992.

FILICORI, M.; BUTLER, J.P.; CROWLEY, W.F. - Neuroendocrine regulation of corpus luteum in the human: evidence of pulsatile progesterone secretion. **J. Clin. Invest., 73**:1638-43, 1984.

GARCIA-VELASCO, J.A.; ARICI, A. – Is the endometrium or oocyte/embryo affected in endometriosis? **Hum. Reprod., 14**: 77-89, 1999.

GARRIDO, N.; NAVARRO, J.; REMOHÍ, J.; SIMÓN, C.; PELLICER, A. Follicular hormonal environment and embryo quality in women with endometriosis. **Hum. Reprod. Update., 6**: 67-74, 2000.

GREGORIOU, G.; BAKAS, P.; VITORATOS, N.; PAPADIAS, K.; GOUMAS, K.; CHRYSSICOPOULOS, A.; CREATSAS, G. – Evaluation of serum prolactin levels in patients with endometriosis and infertility. **Gynaecol. Obstet. Invest., 48**: 48-51, 1999.

GINSBURG, K.A. - Luteal phase defect: Etiology, Diagnosis and Treatment. **Endocrinology and Metabolism Clinics of North America, 21** (1):85-104, 1992.

HARGROVE, J.T.; ABRAHAM, G.K. - Abnormal luteal function in endometriosis (Abstract). **Fertil. Steril., 34**:302, 1980.

HARLOW, C.R.; CAHILL, D.J.; MAILE, L.A.; TALBOT, W.M.; MEARS, J.; WARDLE, P.G.; HULL, M.G.R. - Reduced preovulatory granulosa cell steroidogenesis in women with endometriosis. **J. Clin. Endocrinol. Metab., 81**:426-9, 1996.

HAYASHI, N.; TEKETANI, Y.; MIZUNO, M. – Relationship between luteal function and prolactin in infertile women with endometriosis. **Acta Obstet. Gynaec. Jpn. 41**: 1720-24, 1989.

HE, YE. - Prolactin secretion in patients with endometriosis and its relationship to luteal phase defect and infertility. **Chung Hua Fu Chan Ko Tsa Chih 28**: 14-7, 1993.

HIRSCHOWITZ, J.S.; SOLER, N.G.; WORTSMAN, J. - The galactorrea endometriosis syndrome. **Lancet, 1**: 896-898, 1978.

ISHIBASHI, M.; YAMAJI, T. Effect of Thyrotropin-releasing hormone and bromoergocriptine on growth hormone and prolactin secretion in perfused pituitary adenoma tissues of acromegaly. **J. Clin. Endocrinol. Metab., 47**: 1251-6, 1978.

JONES, G.E.S. - Some newer aspects of the management of infertility. **JAMA 141**:1123, 1949.

JORDAN, J.; CRAIG, K.; CLIFTON, D.K.; SOULES, M.R. - Luteal phase defect: the sensitivity and specificity of diagnostic methods in common clinical use. **Fertil. Steril., 62**:54-62, 1994.

JORDAN, V.; DIEGUEZ, C.; LAFAFFIAN, I.; RODRIGUEZ-ARNAO, M.D.; GOMEZ-PAN, A.; HALL, R.; SCANLON, M.F. Influence of dopaminergic,

adrenergic and cholinergic blockade and TRH administration on GH responses to CRF 1-29. **Clin. Endocrinol., 29**: 291-8. 1986.

KALTSAS, T.; PONTIKIDES, N.; KRASSAS, G.E.; SEFERIADIS, K.; LOLIS, D.; MESSINIS, I.E. – Growth hormone response to thyrotrophin releasing hormone in women with polycystic ovarian syndrome. **Human. Reprod., 14**: 2704-8, 1999.

KATZ, E.; RICCIARELLI, E.; ADASHI, E. Y. – The potential relevance of growth hormone to female reproductive physiology and pathophysiology. **Fertil. Steril., 59**: 8-34, 1993.

KAUPPILA, A.; LEINONEN, P.; VIHKO, R.; YLOSTALO, P. – Metoclopramide induced hyperprolactinemia impairs ovarian follicle maturation and corpus luteum function in women. **J. Clin. Endocrinol. Metab., 54**: 955-60, 1982.

KONINCKX, P.R. - Is mild endometriosis a disease? **Hum. Reprod., 9**:2202-11, 1994.

KOSTAL, M.; TOSNER, J. – The influence of latent hyperprolactinemia on the levels of LH, FSH, E2 and T in the midfollicular phase of the cycle. **Arch. Gynecol. Obstet., 259**: 65-8, 1997.

KUSUHARA, K. - Luteal function in infertile patients with endometriosis. **Am. J. Obstet. Gynecol., 167**:274-7, 1992.

MACHIDA, T.; TAGA, M.; MINAGUCHI, H. – Prolactin secretion in endometriotic patients. **Eur. J. Obst. Gynecol. Reprod. Biol., 72**: 89-92, 1997.

MASALA, A.; DELITALA, G.; ALAGNA, S.; DEVILLA, L.; ROVASIO, P.; LOTTI, G. – Effect of Dopaminergis blockade on secretion of growth hormone and prolactin in man. **Metabolism, 27**(8): 921-6, 1977.

MASON, H.D.; MARTIKAINEN, H.; BEARD, R.W.; ANYAOKU, V.; FRANKS, S. – Direct gonadotrophic effect of growth hormone on oestradiol by human granulosa cells in vitro. **Journal of Endocrinology, 126**: R1-R4, 1990.

MASSARA, F.; TANGOLO, D.; GODANO, A.; GOFFI, S.; BERTAGNA, A.; MOLINATTI, G.M. – Effect of metoclopramide, domperidone and apomorphine on GH secretion in children and adolescents. **Acta Endocrinologica, 108**: 451-5, 1985.

MATALLIOTAKIS, I.; PANIDIS, D.; VLASSIS, G.; VAVILIS, D.; NEONAKI, M.; KOUMANTAKIS, E. - PRL, TSH and their response to the TRH test in patients with endometriosis before, during, and after treatment with danazol. **Gynecol. Obstet. Invest., 42**:183-6, 1996.

MATORRAS, R.; RODRÍGUEZ, F.; PÉREZ, C.; PIJOAN, J.L.; NEYRO, J.L.; RODRÍGUEZ-ESCUDERO, F.J. – Infertile woman with and without endometriosis: a case control study of luteal phase and other infertility conditions. **Acta Obst. Gynec. Scand., 75**: 826-831, 1996.

MATORRAS, R.; RODÍQUEZ, F.; PIJOAN, J.I.; RAMÓN, O.; TÉRAN, G.G.; RODRÍGUEZ-ESCUDERO, F. – Epidemilogy of endometriosis in infertile women. **Fertil. Steril., 63** (10): 34-38, 1995.

MENASHE, Y.; LUNEFELD, B.; PARIENTE, C.; FRENKEL, Y.; MASHIACH, S. – Can growth hormone increase, after clonidine administration, predict the

dose of human menopausal hormone needed for induction of ovulation? **Fertil. Steril., 53**: 432-4, 1990.

MOELOECK, F.A ; MOEGNY, E. - Endometriosis and luteal phase defect. **Asia-Oc.-J.-Obstet.-Gynaecol., 19**(2):171-6, 1993.

MUSE, K.N.; WILSON, E.A. - How does mild endometriosis cause infertility?. **Fertil. Steril., 38**: 145-152, 1982.

MUSE, K.N.; WILSON, E.A. ; JAWAD, M.J. - Prolactin hyperstimulation in response to thyrotropin-releasing hormone in patients with endometriosis. **Fertil. Steril., 38**: 419-422, 1982.

NISOLLE, M. & DONNEZ, J.- Peritoneal endometriosis, ovariam endometriosis, and adenomyotic nodules of the rectovaginal septum are three different entities. **Fertil. Steril., 68**:585-596, 1997.

NOYES, R.W.; HERTIG, A . T.; ROCK, J. - Dating the endometrial biopsy. **Fertil. Steril.,1**:1-25, 1950.

OLIVE, D.L.; SCHWARTZ, L.B. - Endometriosis. **N. Engl. J. Med. 328:**1759-1767, 1993.

OLIVE, D.L.; HANEY, A .F. - Endometriosis- Associated infertility: A critical review of therapeutic approaches. **Obstet. Gynecol. Surv., 41**:538-55, 1986.

OVESEN, P.; INGERSLEV, H.J.; ORSKOV, H.; LADET, T. – Effect of growth hormone on steroidogenesis, insulin-like growth factor-I (IGF-I) and IGF binding protein-1 production and DNA syntesis in cultured human luteinized granulosa cells. **Journal of Endocrinology, 140**: 313-9, 1994.

PALMA DIAS, R.; BRUGNARA, L.; BRUGNARA, L.; CAVAGNOLI, G.; CÉSAR, C.P.; FRONZA, R.; GUGEL, F.; FREITAS, F.; RAMOS, J.G.; PASSOS, E.P.- Indicações e achados diagnósticos em laparoscopias ginecológicas no Hospital de Clínicas de Porto Alegre. **Rev. ATM 95/1, 1**:11-16, 1995.

PANERAI, A. E.; SALERMO, F.; MENNESCHI, M.; COCCHI, D.; MÜLLER, E.E. Growth hormone and prolactin responses to thyrotropin-releasing hormone in patients with severe liver disease. **J. Clin. Endocrinil. Metab., 45**: 134-40, 1977.

PASSOS, E.P.; FREITAS, F.; FACIN, A.C.; CUNHA, J.S.L. - Infertilidade e técnicas de reprodução assistida. In: FREITAS, F.; MENKE, C.H.; RIVOIRE, W.A.; PASSOS, E.P. - **Rotinas em ginecologia.** Artes Médicas. Porto Alegre. 1997. Cap. 38.

PIADITIS, G.P.; KOUNADI, T.G.; RANGOU, D.B.; TROVAS, G.P.; KAKLAS, N.A.; TZONOU, A.J.; CHLOUVERAKIS. – Dysfunstion of the growth hormone/insulin like growth factor-I axis in women with polycystic ovarian syndrome. **Clinical Endocrinology, 42**: 635-40, 1995.

PITTAWAY, D.E.; MAXSON, W.; DANIELL, J.; HERBERT, C.; WENTZ, A.C. – Luteal phase defects in infertility patients with endometriosis. **Fertil. Steril., 39**: 712-713, 1983.

RADWANSKA, E.; HENIG, I.; DMOWSKI, W.P. – Nocturnal prolactin levels in infertile women with endometriosis. **J. Reprod. Med., 32**: 605-8, 1987.

RODRIGUES-ESCUDERO, F.J.; NEYRO, J.L.; CORCOSTEGUI, B.; BENITO, J.A. – Does minimal endometriosis reduce fecundity?. **Fertil. Steril., 50** (3): 522-524, 1988.

ROSSATO, P.; MINUTO, F.; GARRONE, S.; RAGNI, N. – Growth hormone response to clonidine in anovulatory infertile women resistant to clomiphene citrate stimulation. **Fertil. Steril., 73**:78-84, 2000.

SAMPSON, J.A. - Peritoneal endometriosis due to menstrual dissemination of endometrial tissue into peritoneal cavity. **Am. J. Obstet. Gynecol. 14**:422-35, 1927.

SEPPALA, M.; HIRVONEN, E.; RANTA, T. - Hyperprolactinemia and luteal insufficiency. **Lancet, 1**:229-34, 1976.

SPEROFF, L.; GLASS, R.H.; KASE, N.G. – Clinical Gynecologic Endocrinology and Infertility. Lippincott Williams & Wilkins Publishers, 6th Edition, 1999.

SERRI, A.; BEAUREGARD, H.; SOMMA, M.; COMTOIS, R.; JILWAN, N.; HARDY, J. – Defective dopaminergic regulation of prolactin in patients with hyperprolactinemia**. Clin. Invest. Med., 11**: 387-91, 1988.

STEINBERGER, E.; NADER, S.; RODRIGUEZ-RIGAU, L.; AYALA, C.; SMITH, K. – Prolactin response to thyrotropin-releasing hormone in normoprolactinemic patients with ovulatory dysfunction and its use for selection of canditates for bromocriptine therapy. **J. Endocrinol. Invest. 13**: 637-42, 1990.

STRAUSS III, J.F. & COUTIFARIS C. - The endometrium and myometrium: regulation and dysfunction. In: YEN, S.S.C. & JAFFE, R.B.- **Reproductive endocrinology.** Philadelphia, W.B. Saunders Company, 1998. p.218-256.

THE AMERICAN FERTILITY SOCIETY. - Revised American Fertility Society classification of endometriosis. **Fertil. Steril., 43**:351-2, 1985.

TOYA M, SAITO H, OHTA N, SAITO T, KANEKO T, HIROI M. - Moderate and severe endometriosis is associated with alterations in the cell cycle of granulosa cells in patients undergoing in vitro fertilization and embryo transfer. Fertil. Steril., 73(2): 344-50, 2000.

TUMMON, I.S.; MACLIN, V.M.; RADWANSKA, E.; BINOR,Z.; DMOWSKI, W.P. – Occult ovulatory dysfunction in women with minimal endometriosis or unexplained infertility. **Fertil. Steril., 50** (5): 716-720, 1988.

WILLIAMS, C. A. V.; OAK, M.K.; ELSTEIN, M. – Cyclical gonadotrophin and progesterone secretion in women with minimal endometriosis. **Clin. Reprod. Fertil., 4**: 259-268, 1986.

VANCE, M.L.; KAISER, D.L.; FROHMAN, L.A.; RIVIER, J.; VALE, W.W.; THORNER, M.O. – Role of dopamine in the regulation of Growth Hormone secretion: dopamine and bromocriptine augment growth hormone-releasing hormone stimulated GH secretion in normal man. **J. Clin. Endocrinol. Metab., 64**: 1136-41, 1987.

VÁZQUEZ-MATUTE, L.; CANALE, E.S.; ALGER, M.; ZÁRATE, A. Clinical use of metoclopramide test in the diagnosis of women with hyperprolactinemia. **Horm. Res., 10**: 207-12, 1979.

YEN S.S.C.- Prolactin in human reproduction. In: YEN, S.S.C. & JAFFE, R.B.- **Reproductive endocrinology.** Philadelphia, W.B. Saunders Company, 1998. p.257-283.

HYPERPROLACTINEMIA AND LUTEAL INSUFFICIENCY IN INFERTILE PATIENTS WITH MILD AND MINIMAL ENDOMETRIOSIS*

Short title: Infertility and endometriosis

João Sabino L. Cunha-Filho, MD†, Jorge L. Gross, MD‡, Nadiane A. Lemos, MD†, Adriano Brandelli, PhD†, Melissa Castillos, MD†, Eduardo P. Passos, MD, PhD†.

*Supported by Fundo de Incentivo à Pesquisa, Hospital de Clínicas de Porto Alegre, Brazil.

†Obstetrics and Gynecology Department, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil.

‡Endocrinology Division, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil.

Correspondence:

Dr. Eduardo P. Passos

Departamento de Ginecologia e Obstetrícia - Hospital de Clínicas de Porto Alegre

Rua Ramiro Barcelos, 2350 – 11º andar

90035-003 Porto Alegre, RS – Brazil

Phone/Fax: +55-51-346-7155

e-mail: epp@via-rs.net

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SUMMARY

The objective of the present paper was to assess the presence of hormonal alterations in infertile women with stage I or II endometriosis (Group III, n =20) comparing with fertile women without endometriosis (Group I, n=14) and fertile women with endometriosis (Group II, n=7). Serum levels of FSH, LH, estradiol, TSH, and PRL were measured between days 1 and 5 of the early follicular phase; in the luteal phase, three serum samples were collected for progesterone measurement, and endometrial biopsies were performed. Serum estradiol levels were lower (P=0.035) in infertile patients with endometriosis than in fertile patients without endometriosis. Six infertile patients with endometriosis presented prolactin levels above 20ng/ml. This was not observed in the other groups. Luteal insufficiency was more frequent in infertile patients with endometriosis (78.9%) than in fertile patients with (42.9%) or without endometriosis (0%). In a multiple logistic regression analysis, only the presence of endometriosis and infertility was significantly associated with luteal insufficiency. The serum levels of LH, FSH, and TSH were not significantly different among the groups. Luteal insufficiency and altered prolactin secretion were associated with endometriosis, and could be important mechanisms causing infertility in this group of patients.

Keywords: Hormonal alterations; progesterone; endometrial biopsy; prolactin.

INTRODUCTION

Endometriosis is estimated to affect between 10 and 15% of the female population (1) and is frequently (30-60%) associated with infertility (2). Several mechanisms have been suggested as underlying the etiology of infertility in patients with endometriosis. There is an anatomical basis for infertility in moderate or severe endometriosis patients; however, in patients with minimal or mild endometriosis, there is no clear cause of infertility (1,3-6).

An association between endometriosis and anovulation has also been suggested (3), as well as between abnormal folliculogenesis and alterations in the follicular and luteal phases. In addition, hormonal abnormalities have been observed in infertile patients with endometriosis, such as midluteal erratic progesterone secretion, altered luteinizing hormone peak, and PRL secretion (4,5,7-9).

The present study assessed the possible hormonal alterations associated with minimal and mild endometriosis and infertility by evaluating the luteal phase and the early follicular phase in infertile women with endometriosis compared with fertile women with and without endometriosis.

PATIENTS AND METHODS

Design

This study followed a case-control design. Patients with mild and minimal endometriosis and infertility (Group III) were compared with two control groups: fertile patients without endometriosis (Group I) and fertile patients with minimal or mild endometriosis (Group II). The patients were compared in terms of the serum levels of FSH, LH, estradiol, TSH, and PRL in the early follicular phase, and in terms of serial progesterone measurements and endometrial biopsy in the luteal phase.

Patients

Patients were selected among the women seen at the Gynecological Clinic at Hospital de Clínicas de Porto Alegre. We excluded patients with previous endocrine disorders, patients who were using drugs that could affect the parameters of the tests employed, and patients with contraindications for endometrial biopsy (current pelvic infection, IUD users, suspicion of pregnancy, unclear histologic or cytological alteration of the cervix uteri or endometrium). Patients whose infertility was not caused by endometriosis were also excluded.
A total of 41 patients were analyzed between March, 1997, and January, 1998. They were divided into three groups: Group I included 14 patients with proven fertility in the year prior to the study (not using any hormonal form of

contraception), who were submitted to laparoscopy for tubal ligation (control group); Group II included seven patients, all of them fertile and with stage I and II endometriosis (endometriosis control group) who underwent laparoscopy for investigation of pelvic pain or for tubal ligation; and Group III was formed by 20 infertile patients with a diagnosis of stage I and II endometriosis with normal results in other infertility tests (infertility and endometriosis group), in whom laparoscopy was performed for infertility investigation. Endometriosis was graded according to the classification of the American Society for Reproductive Medicine (10). All patients were informed of the procedures and spontaneously signed an informed consent form. The research project was approved by the Ethics Committee of the Hospital (IRB-equivalent).

The endoscopic procedure was carried out in all cases by the same investigator (JSLCF).

Measurements

Endometrial biopsy was performed between the 23rd and 26th days of the menstrual cycle. The first day of bleeding was considered as the first day of the cycle. Uterine fundus endometrium material was collected and placed in vials with formol for later anatomopathologic analysis. The samples were dated by the pathologist following the criteria of Noyes et al. (11). Luteal insufficiency was defined as a lag of more than 2 days between the histologic dating and the first day of the next menstrual period (following the endometrial biopsy).

Hormonal assessment was carried out by measuring serum levels of FSH, LH, estradiol, TSH and PRL during the first 5 days of the menstrual cycle. The prolactin measurements were done in a pool of three samples of equal volume. Progesterone was measured three times, at 3-day intervals, during the luteal phase. Luteal insufficiency was diagnosed when the sum of the three measurements was less than 30 ng/ml (12). All samples were centrifuged at 250 rpm for the separation of plasma, which was frozen at –20 °C for later analysis. The hormones were analyzed using chemiluminescence kits (Immulite Ltd., U.S.A.). The largest inter and intra-kit variation was 13% for progesterone; 15 and 16% for estradiol; 8.1 and 7.7% for FSH; 10.6 and 6.5% for LH; 17.5 and 13.8% for TSH; and 5.45 and 13.3% for PRL.

Statistical Analysis

Categorical data were analyzed with Fisher's exact test with Bonferroni correction due to the asymmetrical distribution observed. Continuos variables were compared among groups by means of the Mann-Whitney's U test (MWU), and Kruskal-Wallis' test (for variance analysis) non-parametric tests, followed by post hoc Dunn procedure for multiple comparisons.

The results were expressed as medians and 95% confidence intervals (CI) or range. P values < 0.05 (two-tailed) were considered to be statistically significant, with a power calculation of 80% for this sample size. The relationship of relevant variables with luteal insufficiency (dependent variable) was assessed by multiple logistic regression.

RESULTS

The median age for group I patients was 33 years (26-41 years). Group II patients were older (38 years; 33-40) than patients in group III (31 years; 26-39) (P=0.006/Kruskal-Wallis). The body mass index (BMI) was similar for all groups (P=0.25/Kruskal-Wallis).

The medians for serum levels of FSH, LH, TSH, PRL, and estradiol are shown in Table 1 with the respective 95% CI. Estradiol levels were different among the groups (P = 0.035/Kruskal Wallis). According to the post hoc Dunn test, we observed that this difference is significant only between the groups I and III (P<0.05).

Six patients in group III (30%) presented hyperprolactinemia, that is, PRL levels higher than 20 ng/ml. No such finding was observed for patients in groups I and II (P = 0.025/Fisher's exact test). When all the patients with endometriosis (groups II and III) were grouped, the PRL levels (11.80 ng/ml; 11.20-19.38) were higher than the PRL levels of fertile women without endometriosis (7.05 ng/ml; 5.60-11.35; P = 0.037/MWU), as is shown in Figure 1.

Luteal function was analyzed by the sum of progesterone measurements and endometrial biopsy. Regarding progesterone measurements, only samples collected at 3-day intervals in the luteal phase were considered for analysis of progesterone levels. We observed that out of 20 patients with infertility and endometriosis, 13 (65%) had the sum of progesterone levels lower than 30 ng/ml in three plasma samples. Otherwise, this finding was observed in only one fertile

patient with endometriosis and in none of fertile patients without endometriosis (figure 2). Post-hoc analysis disclosed that the proportion of luteal insufficiency was higher in infertile patients with endometriosis as compared to fertile patients without endometriosis (P=0.0015/Bonferroni correction).

A significant difference among groups was also observed in terms of endometrial biopsy results (P = 0.001/Fisher's exact test). None of the 14 cases studied in group I showed luteal insufficiency upon endometrial biopsy. In group II, 3 out of 7 patients (42.9%) presented an out of phase endometrium, and in group III, luteal insufficiency was diagnosed in 15 out of 19 patients (78.9%) (Figure 3). This difference is statistical significant between the groups I and III (P=0.00009/ Bonferroni correction).

The possible factors associated with luteal insufficiency (dependent variables) were analyzed by multiple logistic regression analysis. Two models were characterized: in one model the presence of luteal insufficiency was established following endometrial biopsy criteria; in the second model, diagnosis of luteal insufficiency was based on progesterone levels. Only the presence of endometriosis and infertility was significantly associated with luteal insufficiency as diagnosed by endometrial biopsy (OR:29.13, 95% CI:2.60- 326.42, P = 0.0062) or by progesterone assays (OR:12.31, 95% CI:1.06-143.65, P = 0.045) (table 2).

DISCUSSION

The present study was unique to include a group of fertile patients with endometriosis. Most other studies compare patients with endometriosis with patients with unexplained infertility; however, this last group could be a very heterogeneous group, encompassing patients with infertility resulting from various causes. This study was also unique in controlling estradiol, prolactin, and endometriosis as independent variables, and luteal phase as the dependent variable, using multiple logistic regression analysis. We observed that infertile patients with minimal or mild endometriosis had a higher prevalence of hyperprolactinemia as well as of luteal insufficiency. Furthermore, these patients presented alterations in the early follicular phase, characterized by a decrease in estradiol levels as well as by a higher prevalence of hyperprolactinemia and luteal insufficiency. This could be related with a disturbance of the pituitary-ovarian axis, as described by Cahil et al. (13).

The patients of the groups differed regarding the age. Probably this difference did not affect the results because the patients had similar levels of FSH, and had regular menstrual cycles.

Some authors have observed altered estrogen concentration in patients with infertility and endometriosis (4,5,13,14), while others did not observe a significant difference in estradiol secretion during the menstrual cycle in these patients (15-17). In our study, the low estrogen levels observed in infertile patients with endometriosis could be a result of increased PRL secretion.

There is no consensus regarding the association between PRL levels and endometriosis. Some authors observed that infertile women with endometriosis had a basal PRL level twice as large as that of the control group ($P > 0.05$). This difference became significant after the stimulus test with TRH (6,18). Others (8,9) found significantly higher basal PRL levels in patients with infertility and endometriosis. Brosens et al. (4), Matalliotakis et al. (19) and Matorras et al. (20), however, did not find a significant difference between the groups in terms of basal PRL levels.

The phenomenon of normal basal serum PRL levels associated with altered prolactin secretion after the TRH stimulation test has been called occult or masked hyperprolactinemia (21), and could be the cause of luteal phase defects in patients with normal PRL values. Moreover, luteal insufficiency is a multifactorial disease (22), involving not only prolactin secretion, but also LH secretion and ovarian dysfunction.

According to our data, only patients with infertility and endometriosis presented PRL levels higher than 20 ng/ml in the early follicular phase. This suggests a difference in prolactin secretion between fertile patients and infertile patients with endometriosis.

Several authors have found lower progesterone levels in women with infertility and endometriosis (14-16,23,24). Other authors (20,25-28) did not associate an increase in the prevalence of luteal phase defects with infertility and endometriosis. Moreover, the progesterone cut-off point used to define luteal insufficiency and the characterization of the control group varied widely in these last

studies. Our findings agree with those of Williams et al. (15) in that there is an association between endometriosis and luteal insufficiency, as detected by the progesterone assay. Recent studies (29) have stressed the importance of the luteal phase in patients with endometriosis. Indeed, this is a controversial issue, especially in patients at mild/minimal stages of endometriosis, without tubal occlusion.

In conclusion, patients with stage I and II endometriosis (as defined by the American Society for Reproductive Medicine) present an increase in the prevalence of luteal insufficiency, probably related to increased PRL levels. These mechanisms interact and could be etiologically relevant for the infertility in women with minimal or mild endometriosis. Endometriosis should be viewed as a syndrome with peritoneal, immunologic, and also hormonal abnormalities causing infertility.

REFERENCES

1 Olive DL, Schwartz, LB: Endometriosis. N Engl J Med 328:1759-1767 (1993).

2 Koninckx PR: Is mild endometriosis a disease? Hum Reprod 9:2202-2211 (1994).

3 Soules MR, Malinak LR, Bury R, Poindexter A: Endometriosis and anovulation: A coexisting problem in the infertile female. Am J Obstet Gynecol 125: 412-417 (1976).

4 Brosens IA, Koninckx PR, Corveleyn PA: A study of plasma progesterone, oestradiol-17B, PRL and LH levels, and of the luteal phase appearance of the ovaries in patients with endometriosis and infertility. Br J Obstet Gynaecol 85:246-250 (1978).

5 Cheesman KL, Bem-Nun I, Chatterton RT, Cohen MR: Relationship of luteinizing hormone, pregnanediol-3-glucuronide, and estriol-16- glucuronidein urine of infertile women with endometriosis. Fertil Steril 38:542-548 (1982).

6 Muse KN, Wilson EA: How does mild endometriosis cause infertility? Fertil Steril 38:145-152 (1982).

7 Muse KN, Wilson EA, Jawad MJ: Prolactin hyperstimulation in response to thyrotropin-releasing hormone in patients with endometriosis. Fertil Steril 38:419-422 (1982).

8 Acién P, Lloret M, Graells M: Prolactin and its response to the luteinizing hormone-releasing hormone thyrotropin-releasing hormone test in patients

with endometriosis before, during and after treatment with danazol. Fertil Steril 51:774-780 (1989).

9 He YE: Prolactin secretion in patients with endometriosis and its relationship to luteal phase defect and infertility. Chung Hua Fu Chan Ko Tsa Chih 28:14-17 (1993).

10 The American Fertility Society: Revised American Fertility Society classification of endometriosis. Fertil Steril 43:351-352 (1985).

11 Noyes RW, Hertig AT, Rock J: Dating the endometrial biopsy. Fertil Steril 1:1-25 (1950).

12 Jordan J, Craig K, Clifton DK, Soules MR: Luteal phase defect: the sensitivity and specificity of diagnostic methods in common clinical use. Fertil Steril 62:54-62 (1994).

13 Cahill DJ, Wardle PG, Maile LA , Harlow CR, Hull MG: Pituitary-ovarian dysfunction as a cause for endometriosis-associated and unexplained infertility. Hum Reprod 10:3142-3146 (1995).

14 Tummon IS, Maclin VM, Radwanska E, Binor Z, Dmowski WP: Occult ovulatory dysfunction in women with minimal endometriosis or unexplained infertility. Fertil Steril 50:716-720 (1988).

15 Williams CAV, Oak MK, Elstein M: Cyclical gonadotrophin and progesterone secretion in women with minimal endometriosis. Clin Reprod Fertil 4:259-268 (1986).

16 Bancroft K, Williams CAV, Elstein M: Pituitary-ovarian functioning women with minimal or mild endometriosis and otherwise unexplained infertility. Clin Endocrinol 36:177-81 (1992).

17 Barry-Kinsella C, Sharma SC, Cottell E, Harrison RF: Mid to late luteal phase steroids in minimal stage endometriosis and unexplained infertility. Eur J Obstet Gynecol Reprod Biol 54:113-118 (1994).

18 Machida T, Taga M, Minaguchi H: Prolactin secretion in endometriotic patients. Eur J Obstet Gynecol Reprod Biol 72:89-92 (1997).

19 Matalliotakis I, Panidis D, Vlassis G, Vavilis D, Neonaki M, Koumantakis E: PRL, TSH and their response to the TRH test in patients with endometriosis before, during, and after treatment with danazol. Gynecol Obstet Invest 42:183-186 (1996).

20 Matorras R, Rodríguez F, Pérez C, Pijoan JI, Neyro JL, Rodríguez Escudero FJ: Infertile woman with and without endometriosis: a case control study of luteal phase and other infertility conditions. Acta Obstet Gynecol Scand 75:826-831 (1996).

21 Asukai K, Uemura T, Minaguchi H: Occult hyperprolactinemia in infertile women. Fertil Steril 60:423-427 (1993).

22 Hinney B, Henze C, Kuhn W, Wuttke W: The corpus luteum insufficiency: a multifactorial disease. J Clin Endocrinol Metab 81:565-570 (1996).

23 Hargrove JT, Abraham GK: Abnormal luteal function in endometriosis. Fertil Steril 34:302 (1980).

24 Cheesman KL, Cheesman SD, Chatterton RT, Cohen MR: Alterations in progesterone metabolism and luteal function in infertile women with endometriosis. Fertil Steril 40:590-595 (1983).

25 Pittaway DE, Maxson W, Daniell J, Herbert C, Wentz AC: Luteal phase defects in infertility patients with endometriosis. Fertil Steril 39:712-713 (1983).

26 Ballach J, Vanrell JÁ: Mild endometriosis and luteal function. Int J Fertil 30:4-6 (1985).

27 Kusuhara K: Luteal function in infertile patients with endometriosis. Am J Obstet Gynecol 167:274-277 (1992).

28 Moeloeck FA, Moegny E: Endometriosis and luteal phase defect. Asia Oceania J Obstet Gynaecol 19:171-176 (1993).

29 Garcia-Velasco JA, Arici A: Is the endometrium or oocyte/embryo affected in endometriosis? Hum Reprod 14 (2):77-89 (1999).

Requests for reprints should be addressed to:

Dr. Eduardo P. Passos

Departamento de Ginecologia e Obstetrícia - Hospital de Clínicas de Porto Alegre

Rua Ramiro Barcelos, 2350 – 11º andar - 90035-003 Porto Alegre, RS – Brazil

Table 1. Median and 95%CI for serum levels of FSH, LH, estradiol, PRL, and TSH in the early follicular phase:

	Group I Fertile without endometriosis	Group II Fertile with endometriosis	Group III Infertile with endometriosis	Kruskal-Wallis Statistics (P)
LH	2.65	3.30	3.20	0.484
mIU/ml	(1.94-4.03)	(0.89-5.74)	(2.86-5.65)	
FSH	3.60	4.00	4.40	0.242
mIU/ml	(2.93-4.38)	(1.18-7.18)	(3.36-5.18)	
Estradiol	94.00	108.00	43.50	0.035
pg/ml	(23.83-299.74)	(41.81-207.62)	(34.70-110.41)	
Prolactin	7.05	11.50	14.00	0.112
ng/ml	(5.60-11.35)	(7.85-16.50)	(1.89-23.40)	
TSH	1.49	2.20	1.25	0.103
μIU/ml	(1.12-2.13)	(1.40-3.61)	(1.00-2.30)	

Table 2. Multiple logistic regression analysis of factors associated with luteal phase insufficiency

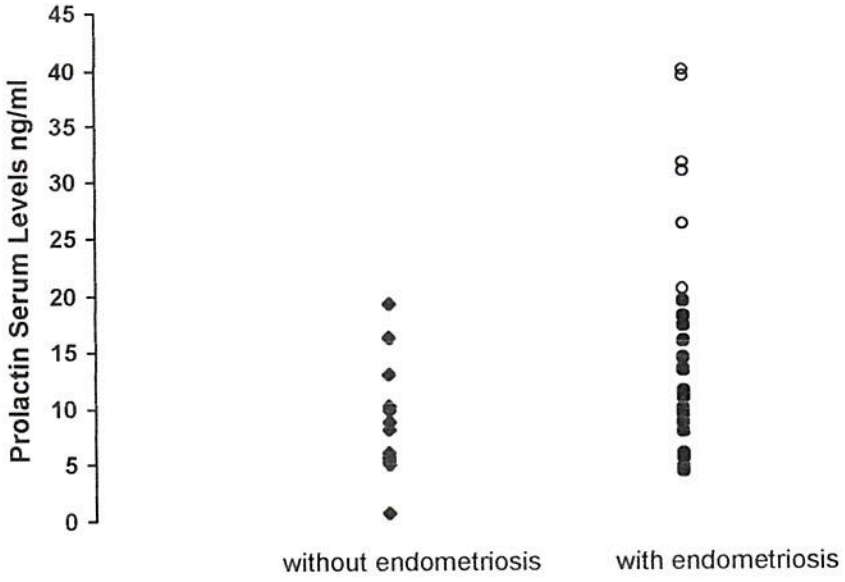
Model 1: dependent variable: endometrial biopsy criteria					
	B	P	R	OR	CI 95%
Estradiol	-0.0044	0.40	0.0000	0.10	0.99-1.01
Prolactin	0.0503	0.39	0.0000	1.05	0.94-1.18
Group II	2.1029	0.11	0.0938	8.19	0.6-111.78
Group III	3.3717	0.0062	0.3146	29.13	2.60-326.42
Model 2: dependent variable: serum progesterone measurements criteria					
	B	P	R	OR	CI 95%
Estradiol	-0.0012	0.72	0.0000	1.00	0.99-1.01
Prolactin	0.1560	0.05	0.1799	1.17	1.00-1.37
Group II	0.1331	0.93	0.0000	1.14	0.05-26.72
Group III	2.5105	0.0452	0.1933	12.31	1.06-143.65

Figure Legends

Figure 1: Serum prolactin levels in women with and without endometriosis. Fertile patients without endometriosis (♦, n=14); fertile and infertile patients with endometriosis (•, n=27); only infertile patients (○) showed serum prolactin levels above 20 ng/ml (hyperprolactinemia), P = 0.037 (Mann-Whitney-U test).

Figure 2: Total progesterone serum levels, sum of three samples, for fertile women without endometriosis (♦, n=14), fertile women with endometriosis (▲, n=7) and infertile women with stage I or II endometriosis (■, n=20).
The open triangle and squares represent patients with a sum of three progesterone measurements lower than 30 ng/ml (luteal phase insufficiency).
P = 0.0015/Fisher's exact test with Bonferroni correction.

Figure 3: Luteal insufficiency diagnosed by endometrial biopsy in Group I (fertile women without endometriosis, n=14), Group II (fertile women with endometriosis, n=7), and Group III (infertile women with stage I or II endometriosis, n=20).
P=0.00009 (Fisher's exact test with Bonferroni correction).



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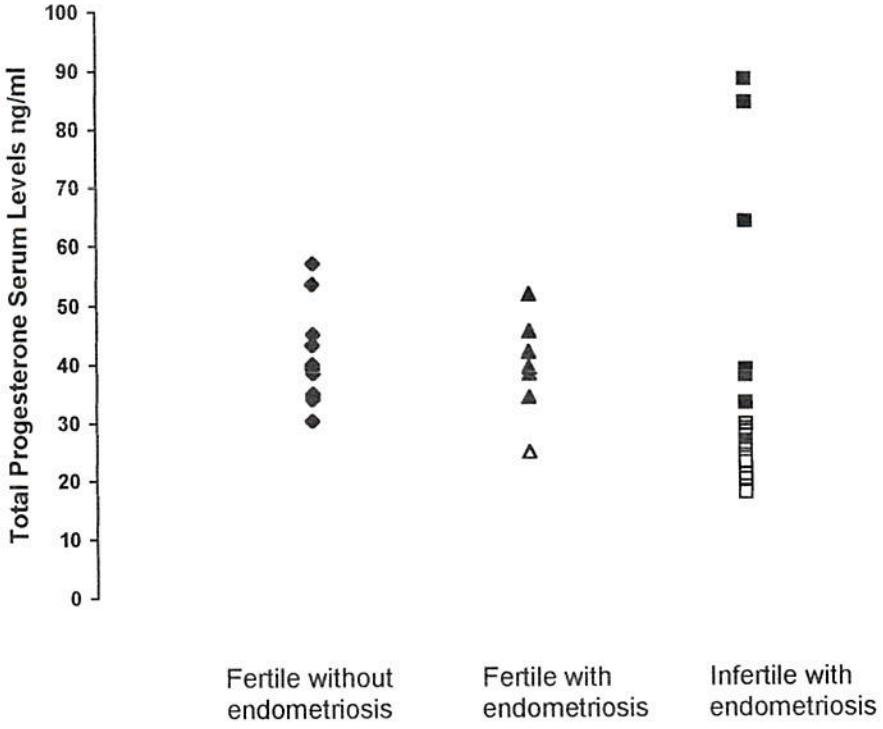

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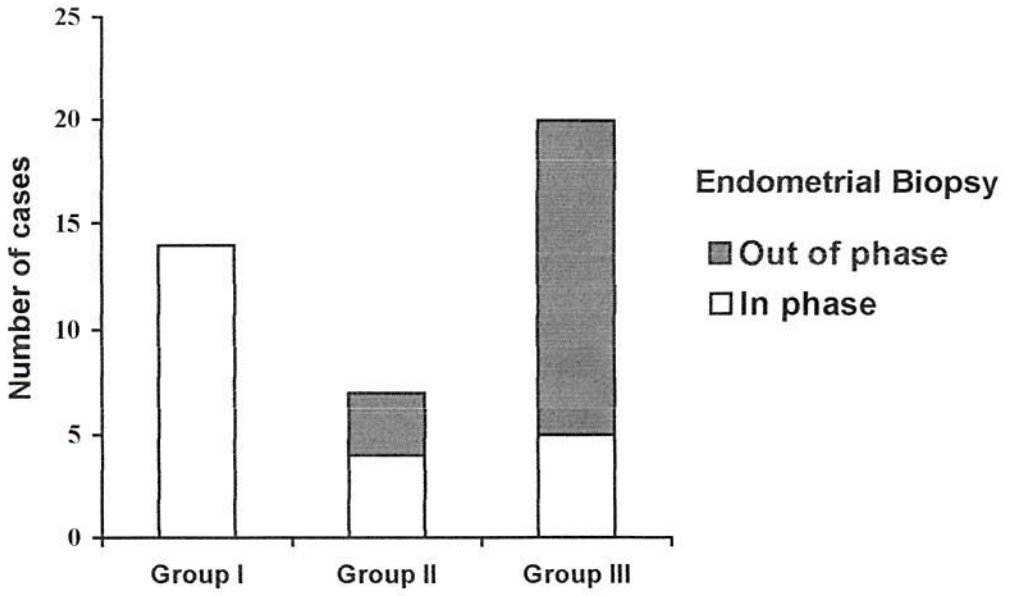

P=0.00009, Fisher's exact test with Bonferroni correction.

Prolactin and growth hormone secretion after TRH infusion and dopaminergic blockade in infertile patients with mild/minimal endometriosis*

João Sabino L. Cunha-Filho, MD†

Jorge L. Gross, MD‡

Melissa Castilhos, MD†

Nadiane Albuquerque Lemos, MD†

Eduardo Costa Dias, MD†

Daniela Vettori, MD†

Eduardo P. Passos, MD, PhD†

Hospital de Clínicas de Porto Alegre and Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

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†Obstetrics and Gynecology Department, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul.

‡Endocrinology Division, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul.

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Abstract

Objective: To investigate the secretion of prolactin and growth hormone after metoclopramide and TRH infusion in infertile patients with mild and minimal endometriosis.

Design: Case-control study.

Setting: Tertiary care center clinic.

Patients: sixty-four women participated in the study: 33 fertile patients without endometriosis; ten fertile endometriotic patients (mild/minimal) who underwent tubal ligation; and 21 patients with minimal or mild endometriosis undergoing infertility investigation.

Intervention: Patients underwent to laparoscopy for tubal ligation or infertility investigation. Blood samples were collected at the beginning of the follicular phase of the menstrual cycle for hormonal analysis. The dopaminergic blockade was achieved with 10 mg of intravenous metoclopramide, and, in the next cycle, 200 μg of TRH were administered. Samples were collected at 15 minute intervals (-15, 0, 15, 30, 45, 60 minutes). The secretion pattern of serum prolactin and growth hormone before and after dopaminergic blockade and TRH were compared.

Results: Basal PRL serum levels were higher in patients with endometriosis (P=0.006), and these patients presented higher prolactin levels than patients without endometriosis 15 and 30 minutes after TRH stimulation (P=0.0001 and 0.036). The serum prolactin levels after dopaminergic blockade did not differ between the groups. Moreover, infertile women with endometriosis showed lower estradiol serum levels (P=0.021) than fertile patients with endometriosis. The analysis of GH showed that fertile patients with endometriosis had higher serum GH levels 30 minutes after TRH administration.

Conclusions: The TRH stimulation test revealed an occult hyperprolactinemia in women with mild/minimal endometriosis, and only those with infertility demonstrated lower serum estradiol levels which may contribute to infertility. Otherwise, the dopaminergic pathway is not associated with abnormal

prolactin secretion in endometriotic patients. Our results suggest a relation between an occult hyperprolactinemia, altered estradiol secretion and infertility, probably due to a follicular and oocyte dysfunction, in patients with mild/minimal endometriosis.

INTRODUCTION

Endometriosis, a disease which has been associated with infertility (Koninckx, 1994), is estimated to affect between 10 and 15% of the female population (Olive and Schwartz, 1993). A number of mechanisms have been suggested as underlying the etiology of infertility in patients with endometriosis. Patients with moderate or severe endometriosis have an anatomical basis for their infertility; until this moment, however, the origin of infertility in patients with minimal or mild endometriosis has not been clearly defined (Olive and Schwartz, 1993; Muse and Wilson, 1993; Olive and Haney, 1986; Candiani et al., 1991; Inoue et al., 1992).

Serum prolactin and growth hormone (GH) levels and secretion are closely related to reproductive status. Prolactin has a pulsatile and circadian cycle, and the main regulatory mechanism for prolactin secretion is the inhibitory action of dopamine (Fossati et al., 1989; Kauppila et al., 1986; Fujimoto et al., 1990; Inaudi et al., 1992; Veldhuis et al., 1989; Frohman, 1995; Schlaff, 1986; Barbarino et al., 1985; Katz et al., 1993; Piaditis et al., 1995). The pattern of prolactin secretion can be studied by inhibiting dopaminergic mechanisms, or by stimulating the release of prolactin by means of a thyrotropin-releasing hormone (TRH) stimulation test (Fossati et al., 1989; Kauppila et al., 1986; Frohman, 1995; Schlaff, 1986; Barbarino et al., 1985).

A hormonal alteration of prolactin secretion could be at the origin of infertility in patients with endometriosis (Acièn et al., 1989; Muse et al., 1982; Machida et al.,

1997; Radwanska et al., 1987; He, 1993), affecting their oocyte maturation and folliculogenesis (Cahill, 2000). Although some authors demonstrated that their infertile endometriotic patients had normal basal serum prolactin levels, these levels were altered following a TRH stimulation test, suggesting an association between this altered secretion pattern and these patient's infertility (Muse et al., 1982; Gregoriou et al., 1999). Still, this association is disputed by some authors (Matalliotakis et al., 1996; Panisdis et al., 1992).

Patients who present luteal phase defects (Archer, 1987) or infertility (Asukai et al., 1993), but have normal serum levels of PRL, have shown an exaggerated response to the TRH stimulation test or to metoclopramide administration (dopaminergic blockade). In such patients, this abnormal PRL secretion reveals an otherwise occult state of hyperprolactinemia, which could be the cause of infertility (Asukai et al., 1993; Aisaka et al., 1987; Steinberger et al., 1990; Kostal et al., 1997; Hayashi et al., 1989).

The regulation of GH secretion is very controversial; it is modulated by various mechanisms and neuro-substances (Devesa et al., 1991). In some conditions, such as acromegaly, polycystic ovarian syndrome (PCOS), anorexia nervosa, liver disease, and depression, GH secretion is impaired after TRH stimulation (Kaltsas et al., 1999; Ishibashi, Yamaji, 1978; Anapliotou et al., 1989; Panerai et al., 1976; Irie et al., 1972; Takahashi et al., 1975).

There is no consensus regarding GH secretion after metoclopramide infusion (dopaminergic blockade), and different investigators have reached

different results (Masala et al., 1978; Cohen et al., 1979; Chiodera et al., 1982; Jordam et al., 1985; Vance et al., 1987; Arce et al., 1991).

Anovulation and infertility could be related with the altered secretion of growth hormone (GH), as proposed by Ovesen et al. (1994). GH may act as a co-gonadotropin, and the literature supports the notion that the somatotrophic axis is associated with the reproductive process and with gonadal function (Mason et al., 1990; Katz et al., 1993; Rossato et al., 1999).

Therefore, the aim of the present study was to investigate a subtle alteration in prolactin and GH levels after dopaminergic blockade, and TRH infusion in infertile patients with mild and minimal endometriosis, and to relate this physiological event to the presence of infertility.

MATERIALS AND METHODS

Design

A case control study was designed to analyze the relationship between infertility in endometriosis and an altered prolactin and GH secretion pattern. A total of 64 patients were studied between March, 1997, and June, 2000. They were selected among patients seen at the Gynecological Clinic at *Hospital de Clínicas de Porto Alegre* (HCPA) and were divided into three groups, according to the presence of infertility and/or endometriosis.

Patients

Group I (control group) consisted of 33 patients with proven fertility and no endometriosis underwent to laparoscopy for tubal ligation. Group II consisted of 10 fertile patients with endometriosis, and group III was formed by 21 patients with infertility and mild/minimal endometriosis, according to the classification proposed by the American Society for Reproductive Medicine (25).

Patients with previous endocrine disorders were excluded from the study, along with patients who were using drugs that could affect the parameters of the tests performed. Also excluded were patients with a history of allergic reaction to metoclopramide. Finally, patients whose infertility was not caused by endometriosis were also excluded from the study.

All participants were informed of the procedures involved in the study and spontaneously signed an informed consent form. The research project was

approved by the Ethics Committee of the Hospital, and followed the guidelines set by the Helsinky Declaration regarding human experimentation.

All endoscopic procedures were carried out by the same investigator (JSLCF).

Measurements

Hormonal assessment for analysis of PRL and GH secretion were done in the early follicular phase. Prior to dopaminergic blockade with metoclopramide or TRH infusion, patients stayed at rest and were submitted to an 8-hour fast. An IV catheter was placed in the antecubital vein 30 minutes before sample collection. After that, 10 mg of metoclopramide or 200 μg of TRH were administered intravenously. Collections were made at 15 minute intervals, for a total of 6 collections, at –15, 0, 15, 30, 45 and 60 minutes. TRH or metoclopramide were performed at random in two sequential menstrual cycles. The –15 and 0 samples were used to measure estradiol, glucose, insulin, IGF-1, TSH and basal serum PRL and GH levels.

All samples were centrifuged at 250 rpm for separation of plasma, which was frozen at –20°C for later analysis. Hormones were analyzed using chemiluminescence kits (Immulite Ltd., USA), IGF-1 was analyzed with IRMA kit from Nichols Institute Diagnostics USA and glucose with the Glico-DH method (Merck Mega, Germany). The largest inter and intra-kit variation was 5.45 and 13.3%, respectively, for prolactin; 15 and 16% for estradiol; 17.5 and 13.8% for TSH; 6.1 and 6.5% for GH; 7.6 and 4.3% for insulin, and 15.8 and 4.6% for IGF-1.

The kits did not show a significant cross-reactivity between the hormones measured.

Statistical Analysis

PRL and GH secretory patterns were compared in the same group by using a model for repeated measures (Friedman test). The hormonal analysis among the groups was performed by Kruskal-Wallis' test (KW), and the Dunn post hoc test was done to evaluate the significant differences between the groups.

The significance level was 0.05 (two-tailed).

RESULTS

The relevant clinical and hormonal characterization of the three groups are showed in table 1. Fertile patients with endometriosis are older than patients of the other two groups, infertile patients with endometriosis had lower estradiol serum levels than fertile patients with endometriosis. Fertile and infertile patients with endometriosis had higher baseline serum prolactin levels than fertile patients without endometriosis. Body mass index (BMI), IGF-1, glucose, insulin and TSH serum levels were similar for the three groups (table 1).

The comparison of prolactin serum levels after TRH infusion was significantly different for all groups. Fertile patients with endometriosis (median: 76.65 ng/ml; 95%IC: 26.60-142.70) and infertile women with endometriosis (median: 72.30 ng/ml; 95%IC: 41.91-493.35) presented higher levels of prolactin 15 minutes after TRH administration (P=0.0001/KW) than fertile patients without endometriosis (median: 38.50 ng/ml; 95%IC: 13.54-120.50). This finding was confirmed after 30 minutes of TRH infusion: group I (median: 30.10 ng/ml; 95%CI: 14.71-119.00), group II (median: 66.25 ng/ml; 95%CI: 23.50-99.20) and group III (median: 52.70 ng/ml; 95%CI: 12.81-478.50) (P=0.036/KW) (figure 1).

The analysis of prolactin secretion after dopaminergic blockade showed no difference between the three groups. PRL secretion had a maximum response after 30 minutes, with a median and 95%CI of 199.00 ng/ml (75.84-436.40) for Group I, 233.75 ng/ml (83.70-289.25) for Group II and 218.50 ng/ml (18.20-476.40) for Group III (P= 0.110/KW) (figure 2).

The secretion of GH after dopaminergic blockade was not different between the three groups (P>0.05/KW) (table 2). Otherwise, fertile patients with endometriosis showed higher GH serum levels after TRH administration on 15 (P=0.018/KW) and 30 minutes (P=0.026/KW) than fertile patients without endometriosis (table 3). However, after Dunn test, these differences are significantly only on 30 minutes (P<0.05).

Serum GH levels were not different after TRH administration between fertile and infertile patients with endometriosis.

DISCUSSION

In the present study, we observed an higher PRL serum levels in patients with endometriosis at baseline, and after 15 and 30 minutes of TRH administration. Also, infertile patients with endometriosis had lower estradiol serum levels, and fertile women with endometriosis showed an altered GH secretion after TRH administration. Moreover, the dopaminergic blockade did not show an abnormal PRL or GH secretion. These results suggest the existence of a central prolactin secretion dysfunction in patients with endometriosis, not involving the dopaminergic system. As far as we know, no study has previously analyzed the dopaminergic and TRH pathways in this particular group of infertile women, and also in studying fertile women with endometriosis.

Hirschowitz et al. (1978) were the first to describe a likely association between endometriosis and galactorrhea. Other authors observed that infertile women with endometriosis had a basal PRL level twice as high as that of the control group, a difference which was not statistically significant ($P > 0.05$). However, this difference became significant after the stimulus test with TRH, indicating the existence of a direct relation between endometriosis stage and the levels of PRL (Acièn et al., 1989; Muse et al., 1982). Many authors have studied PRL secretion after TRH stimulation tests; however, only one group of authors (Wallace et al., 1984) studied PRL secretion in relation to the dopaminergic pathway. This authors detected an increase in prolactin secretion after metoclopramide infusion in patients with endometriosis, and observed also that it is

possible to control the over-secretion of prolactin in endometriotic patients by lowering the estrogenic stimulus. Still, such a conclusion is questioned in other studies (Matalliotakis et al., 1996; Panidis et al., 1992), in which at baseline and after TRH stimulation, serum PRL levels of infertile patients with endometriosis were not statistically higher.

Recently (Gregoriou et al., 1999), a study with patients in all stages of endometriosis presenting infertility demonstrated that an occult hyperprolactinemia may be the main cause of infertility. Others (Asukai et al., 1993; Aisaka et al., 1987; Steinberger et al., 1990; Kostal et al., 1997; Hayashi et al., 1989) have also demonstrated that metoclopramide or TRH can be used for the diagnosis of occult hyperprolactinemia, a condition associated with infertility, probably resulting from an ovulatory dysfunction.

We demonstrated that PRL secretion is abnormal, in patients with endometriosis, only after direct protein kinase C stimulation (TRH pathway), however the dopaminergic blockade did not demonstrated any dysfunction on PRL secretion in those patients. Most likely, the mechanism involved in the abnormal PRL secretion does not include an inhibitory dopaminergic effect.

Patients with latent hyperprolactinemia have lower serum levels of LH and estradiol in the midfollicular phase (Kostal et al., 1997). We can speculate that this finding, higher PRL levels after TRH, could be associated with the follicular dysfunction and ovulatory abnormalities in infertile patients with endometriosis. In fact, an altered steroidogenesis with decreased steroids release was already described (Harlow et al., 1996), and we observed a lower serum estradiol levels in

infertile patients with endometriosis, probably as a consequence of higher PRL secretion. Others also demonstrated the impairment in oocyte quality and follicular development in infertile patients with endometriosis, these findings may be associated with an abnormal ovulatory mechanism (Garrido et al., 2000; Cahill and Hull, 2000).

We have previously described a higher prevalence of hyperprolactinemia, luteal insufficiency, and lower estradiol secretion in infertile patients with endometriosis (Cunha-Filho, et al., 2000). We also agree with Asukai and colleagues (1993) in the sense that we believe that such alteration (occult hyperprolactinemia) affects follicular development and may promote a luteal phase abnormality, possibly contributing to infertility in patients with endometriosis.

GH is associated with the reproductive function and prognosis of infertile patients, mainly due to its co-gonadotrophic modulation. Also, GH stimulates estradiol secretion; human granulosa cells have GH receptors, and an association between infertility and impaired GH secretion has been demonstrated (Mason et al., 1990; Ovesen et al., 1994).

GH secretion after metoclopramide infusion could be stimulated by an inhibitory effect on SS secretion and on the adrenergic tonus (Arce et al., 1991; Cohen et al., 1978; Vance et al., 1987). However, this conclusion is disputed by others that did not find a difference in terms of GH secretion after dopaminergic blockade (Jordam et al., 1986; Masala et al., 1978). This disagreement could be explained by differences in the metoclopramide infusion protocols and in the control groups.

Our results showed that metoclopramide administration did not alter GH secretion in either fertile or infertile patients, probably because GH secretion is modulated by different pathways, and various neurohormones and transmitters are involved. The central role of dopamine in GH secretion depends mainly on its effect on adrenergic transmission to SS neurons, so that it is possible to conclude that dopamine is a modulator, and that it does not have a major role in GH neuroregulation (Devessa et al., 1991).

The exact mechanism causing abnormal GH secretion after TRH is unknown. In some disorders (acromegaly, PCOS, anorexia nervosa, liver disease and depression), GH secretion after TRH administration is altered. Some investigators believe that TRH could stimulate pituitary cells in a similar way as protein kinase C stimulates PRL secretion, or by acting on the hypothalamus (Kaltsas et al., 1999; Ishibashi, Yamaji, 1978; Anapliotou et al., 1989; Panerai et al., 1976; Irie et al., 1972; Takahashi et al., 1975).

Indeed, we may speculate that the enhanced GH secretion in fertile patients with endometriosis can be related with an higher estradiol levels, since estradiol influence GH secretion.

In conclusion, patients with endometriosis have a dysfunction in PRL secretion after TRH infusion (protein kinase C stimulation), however the central dopaminergic system is not affected. Moreover, infertile patients with endometriosis had an altered serum estradiol levels, probably due to a higher PRL secretion. Such alteration in PRL secretion and decreased estradiol, which functions as a control mechanism for reproductive status, is strongly related to a

dysfunction of the hypothalamic-hypophyseal-ovarian axis, and could be the cause of infertility in women with endometriosis without tubal occlusion.

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REFERENCES

1. ACIÈN P, LLORET M, GRAELLS M. Prolactin and its response to the luteinizing hormone-releasing hormone thyrotropin-releasing hormone test in patients with endometriosis before, during and after treatment with danazol. **Fertil Steril** 1989;51:774-80.
2. AISAKA, K.; ANDO, S.; KOKUHO, K.; KANEDA, S.; TAWADA, T.; YOSHIDA, K.; MORI, H. – Role of TRH or metoclopramide loading test for the diagnosis of so-called occulted of latent hyperprolactinemia. **Folia Endocrinolol., 63**:853-61, 1987.
3. ANAPLIOTOU, M.L.G.; SYRIGOS, K.; PANAYIOTAKAPOULOS, G.; KALOS, A.; BATRINOS, M.L. Paradoxical growth hormone response following thyroid-stimulating hormone administration in the polycystic ovary syndrome. **Horm. Res., 31**: 119-24, 1989.
4. ARCE, V.; LIMA, L.; LOIS, N.; RODRIGUEZ, A.; DIAZ, M.J.; TRESGUERRESJ.A.F.; DEVESA, J. – Role of central dopaminergic pathways in the neural control of growth hormone secretion in normal men: studies with metoclopramide. **Neuroendocrinology, 53**: 143-9, 1991.
5. ARCHER DF. Prolactin response to thyrotropin-releasing hormone in women with infertility and/or randomly elevated serum prolactin levels. **Fertil Steril** 1987;47:559-64.
6. ASUKAI K, UEMURA T, MINAGUCHI H. Occult hyperprolactinemia in infertile women. **Fertil Steril** 1993;60:423-7.

7. BARBARINO A, MARINIS LD, MANCINI A, MENINI E, D'AMICO C, PASSERI M, ET AL. Prolactin dynamics in patients with non-secreting tumours of the hypothalamic-pituitary region. **Acta Endocrinol** 1985;110:10-6.
8. CANDIANI GB, VERCELLINI P, FEDELE L. Mild endometriosis and infertility: A critical review of epidemiologic data, diagnostic pitfalls, and classification limits. **Obstet Gynecol Surv** 1991;46:374-82.
9. CAHILL, D.J.; HULL, M.G.R. Pituitary-ovarian dysfunction and endometriosis. **Hum. Reproduc. Update., 6**: 56-66, 2000.
10. CHIODERA, P.; COIRO, V.; ZANARDI, G.; VOLPI, R.; VALENTI, G.; BUTTURINI, U. – Effect of metoclopramide on serum GH levels in normal women. **Horm. Metab. Res., 14**: 103-4, 1982.
11. COHEN, H.N.; HAY, I.D.; THONSON, J.A.; LOGUE, F.; RATCLIFFE, W.A.; BEASTALL, G.H. – Metoclopramide stimulation: a test of growth hormone reserve in adolescents males. **Clinical Endocrinology, 11**: 89-93, 1979.
12. CUNHA-FILHO, J.S.; GROSS, J.L.; CASTILHO, M.; BRANDELLI, A.; LEMOS, N.A.; PASSOS, E.P. – Hyperprolactinemia and luteal insufficiency in infertile patients with mild/minimal endometriosis. **Horm. Metab. Res. In press**, 2000.
13. DEVESA, J.; LIMA, L.; TRESGUERRES, A.F. – Neuroendocrine control of growth hormone secretion in humans. **Trends Endocrinol. Metab., 3**: 175-83, 1992.
14. FOSSATI P, DEWAILLY Y, COMBES M-E, RACADOT A, LEMAIRE C, MÉRGNARGUES I. Classification des hyperprolactinémies en pathologie.

Intérêt du test séquentiel TRH-Metoclopramide. **Franç Endocrinol Clin** 1989;30:331-7.

15. FROHMAN LA. Disease of anterior pituitary. In: FELIG P, BAXTER JD, FROHMAN LA. **Endocrinology and metabolism**. New York: McGraw Hill Inc., 1995:289-383.
16. FUJIMOTO VY, CLIFTON DK, COHEN NL, SOULES MR. Variability of serum prolactin and progesterone levels in normal women: the relevance of single hormone measurements in the clinical setting. **Obstet Gynecol** 1990;76:71-8.
17. GARRIDO, N.; NAVARRO, J.; REMOHÍ, J.; SIMÓN, C.; PELLICER, A. Follicular hormonal environment and embryo quality in women with endometriosis. **Hum. Reprod. Update., 6**: 67-74, 2000.
18. GREGORIOU, G.; BAKAS, P.; VITORATOS, N.; PAPADIAS, K.; GOUMAS, K.; CHRYSSICOPOULOS, A.; CREATSAS, G. – Evaluation of serum prolactin levels in patients with endometriosis and infertility. **Gynaecol. Obstet. Invest., 48**: 48-51, 1999.
19. HAYASHI, N.; TEKETANI, Y.; MIZUNO, M. – Relationship between luteal function and prolactin in infertile women with endometriosis. **Acta Obstet. Gynaec. Jpn. 41**: 1720-24, 1989.
20. HARLOW, C.R.; CAHILL, D.J.; MAILE, L.A.; TALBOT, W.M.; MEARS, J.; WARDLE, P.G.; HULL, M.G.R. - Reduced preovulatory granulosa cell steroidogenesis in women with endometriosis. **J. Clin. Endocrinol. Metab., 81**:426-9, 1996.

21. HE YE. Prolactin secretion in patients with endometriosis and its relationship to luteal phase defect and infertility. **Chung Hua Fu Chan Ko Tsa Chih** 1993;28:14-7.

22. HIRSCHOWITZ JS, SOLER NG, WORTSMAN J. The galactorrhea endometriosis syndrome. **Lancet** 1978;1:896-8.

23. INAUDI P, REYMOND MJ, REY F, GENAZZANI AD, BÉRAUD, TL. Pulsatile secretion of gonadotropins and prolactin during the follicular and luteal phases of the menstrual cycle: Analysis of instantaneous secretion rate and secretory concomitance. **Fertil Steril** 1992;58:51-9.

24. INOUE M, YOSHIMUNE K, HONDA I. The impact of endometriosis on the reproductive outcome of infertile patients. **Am J Obstet Gynecol** 1992;167:278-82.

25. IRIE, M.; TSUSHIMA, T. – Increase of serum growth hormone concentration following thyrotrophin releasing injection in patients with acromegaly or gigantism. **J. Clin. Endocrinol. Metab., 47**:1251-6, 1972.

26. ISHIBASHI, M.; YAMAJI, T. Effect of Thyrotropin-releasing hormone and bromoergocriptine on growth hormone and prolactin secretion in perfused pituitary adenoma tissues of acromegaly. **J. Clin. Endocrinol. Metab., 47**: 1251-6, 1978.

27. JORDAN, V.; DIEGUEZ, C.; LAFAFFIAN, I.; RODRIGUEZ-ARNAO, M.D.; GOMEZ-PAN, A.; HALL, R.; SCANLON, M.F. Influence of dopaminergic, adrenergic and cholinergic blockade and TRH administration on GH responses to CRF 1-29. **Clin. Endocrinol., 29**: 291-8. 1986.

28. KAUPPILA A, HEIKKINEN J, VIINIKKA L. Dynamic evaluation of prolactin secretion by successive TRH and metoclopramide stimulations. **Acta Endocrinol** 1986;111:10-6.

29. KALTSAS, T.; PONTIKIDES, N.; KRASSAS, G.E.; SEFERIADIS, K.; LOLIS, D.; MESSINIS, I.E. – Growth hormone response to thyrotrophin releasing hormone in women with polycystic ovarian syndrome. **Human. Reprod., 14**: 2704-8, 1999.

30. KATZ, E.; RICCIARELLI, E.; ADASHI, E. Y. – The potential relevance of growth hormone to female reproductive physiology and pathophysiology. **Fertil. Steril., 59**: 8-34, 1993.

31. KONINCKX PR. Is mild endometriosis a disease? **Hum Reprod** 1994;9:2202-11.

32. KOSTAL, M.; TOSNER, J. – The influence of latent hyperprolactinemia on the levels of LH, FSH, E2 and T in the midfollicular phase of the cycle. **Arch. Gynecol. Obstet., 259**: 65-8, 1997.

33. MACHIDA T, TAGA M, MINAGUCHI H. Prolactin secretion in endometriotic patients. **Eur J Obst Gynecol Reprod Biol** 1997;72:89-92.

34. MASON, H.D.; MARTIKAINEN, H.; BEARD, R.W.; ANYAOKU, V.; FRANKS, S. – Direct gonadotrophic effect of growth hormone on oestradiol by human granulosa cells in vitro. **Journal of Endocrinology, 126**: R1-R4, 1990.

35. MASSARA, F.; DELITALA, G.; ALAGNA, S.; DEVILLA, L.; RAVASIA, P.P.; LOTTI, G. – Effect of dopaminergic blockade on the secretion of growth hormone and prolactin in man. **Metabolism, 27**: 921-6, 1978.

36. MATALLIOTAKIS I, PANIDIS D, VLASSIS G. PRL, TSH and their response to the TRH test in patients with endometriosis before, during, and after treatment with danazol. **Gynecol Obstet Invest** 1996;42:183-6.

37. MUSE KN, WILSON EA, JAWAD MJ. Prolactin hyperstimulation in response to thyrotropin-releasing hormone in patients with endometriosis. **Fertil Steril** 1982;38:419-22.

38. MUSE KN, WILSON EA. How does mild endometriosis cause infertility? **Fertil Steril** 1993;38:145-52.

39. OLIVE DL, SCHWARTZ LB. Endometriosis. **N Engl J Med** 1993;328:1759-67.

40. OLIVE DL, HANEY AF. Endometriosis-associated infertility: A critical review of therapeutic approaches. **Obstet Gynecol Surv** 1986;41:538-55.

41. OVESEN, P.; INGERSLEV, H.J.; ORSKOV, H.; LADET, T. – Effect of growth hormone on steroidogenesis, insulin-like growth factor-I (IGF-I) and IGF binding protein-1 production and DNA synthesis in cultured human luteinized granulosa cells. **Journal of Endocrinology, 140**: 313-9, 1994.

42. PANERAI, A. E.; SALERMO, F.; MENNESCHI, M.; COCCHI, D.; MÜLLER, E.E. Growth hormone and prolactin responses to thyrotropin-releasing hormone in patients with severe liver disease. **J. Clin. Endocrinol. Metab., 45**: 134-40, 1977.

43. PIADITIS, G.P.; KOUNADI, T.G.; RANGOU, D.B.; TROVAS, G.P.; KAKLAS, N.A.; TZONOU, A.J.; CHLOUVERAKIS. – Dysfunction of the growth hormone/insulin like growth factor-I axis in women with polycystic ovarian syndrome. **Clinical Endocrinology, 42**: 635-40, 1995.

44. PANIDIS D, VAVILIS D, ROUSSO D, PANIDOU E, KALOGEROPOULOS A. Provocative tests of prolactin before, during and after long-term danazol treatment in patients with endometriosis. **Gynecol Endocrinol** 1992;6:19-24.

45. RADWANSKA E, HENIG I, DMOWISKI WP. Nocturnal prolactin levels in infertile women with endometriosis. **J Reprod Med** 1987;32:605-8.

46. ROSSATO, P.; MINUTO, F.; GARRONE, S.; RAGNI, N. – Growth hormone response to clonidine in anovulatory infertile women resistant to clomiphene citrate stimulation. **Fertil. Steril., 73**:78-84, 2000.

47. SCHLAFF WD. Dynamic testing in reproductive endocrinology**. Fertil Steril** 1986;45:589-606.

48. STEINBERGER, E.; NADER, S.; RODRIGUEZ-RIGAU, L.; AYALA, C.; SMITH, K. – Prolactin response to thyrotropin-releasing hormone in normoprolactinemic patients with ovulatory dysfunction and its use for selection of canditates for bromocriptine therapy. **J. Endocrinol. Invest. 13**: 637-42, 1990.

49. TAKAHASHI, S.; KONDO, H.; YOSHIMURA, M. – Enhanced growth hormone response to TRH injection in bipolar depressed patients. **Folia Psychiatr. Neurol., 29**: 215-20, 1975.

50. THE AMERICAN FERTILITY SOCIETY. Revised American Fertility Society classification of endometriosis. **Fertil Steril** 1985;43:351-2.

51. WALLACE AM, LEES DAR, ROBERTS ADG, GRAY CE, MCLAREN EH, LOW RAL. Danazol and prolactin status in patients with endometriosis**. Acta Endocrinol** 1984;107:445-9.

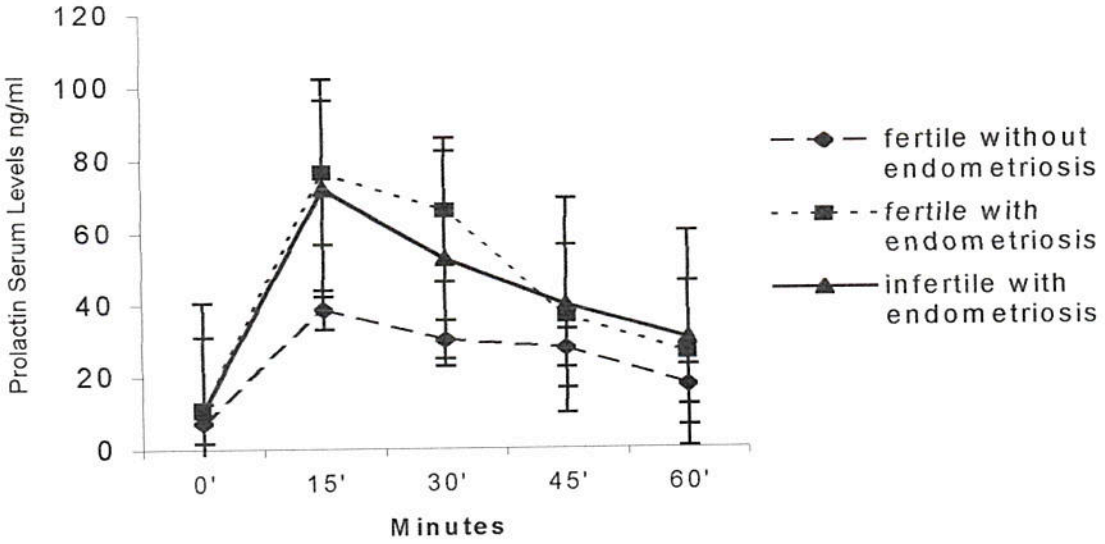
52. VANCE, M.L.; KAISER, D.L.; FROHMAN, L.A.; RIVIER, J.; VALE, W.W.; THORNER, M.O. – Role of dopamine in the regulation of Growth Hormone secretion: dopamine and bromocriptine augment growth hormone-releasing hormone stimulated GH secretion in normal man. **J. Clin. Endocrinol. Metab., 64**: 1136-41, 1987.

53. VELDHUIS JD, EVANS WS, STUMPF PG. Mechanisms that observe estradiol's induction of increased prolactin concentrations: Evidence of amplitude modulation of spontaneous prolactin secretory bursts**. Am J Obstet Gynecol** 1989;161:1149-58.

Table 1: Clinical characteristics and hormone serum levels

	Fertile without endometriosis n=33	Fertile with endometriosis n=10	Infertile with endometriosis n=21	Statistics Kruskal-Wallis P
Age	33	38	31	0.003
	(26-43)	(33-39)	(23-40)	
BMI	23.80	22.85	22.00	0.126
(kg/m^2)	(17.06-32.13)	(20.50-28.02)	(17.51-28.68)	
Estradiol	52.10	122.00	48.40	0.021
(pg/ml)	(22.17-218.90)	(40.00-269.20)	(23.67-157.80)	
TSH	1.47	1.68	1.50	0.427
(μIU/ml)	(0.45-5.66)	(0.26-5.64)	(0.53-3.47)	
PRL	7.20	10.96	10.55	0.006
(ng/ml)	(3.27-17.29)	(3.25-11.96)	(3.96-33.02)	
Glucose	85.00	89.50	77.00	0.100
(mg/dl)	(70.60-107.10)	(71.00-95.50)	(69.00-96.20)	
Insulin	9.60	7.05	6.00	0.130
(μIU/ml)	(2.00-19.50)	(4.00-16.35)	(2.10-18.02)	
IGF-1	306.73	272.92	285.38	0.858
(ng/ml)	(80.53-505.46)	(146.45-365.54)	(140.06-416.81)	

Values are expressed as medians and 95% confidence intervals (95%CI)



P=0.0001 and P=0.036 for a Kruskal-Wallis test, 15 and 30 minutes after TRH.

Figure 1: Prolactin serum levels after TRH administration, median ± SD.

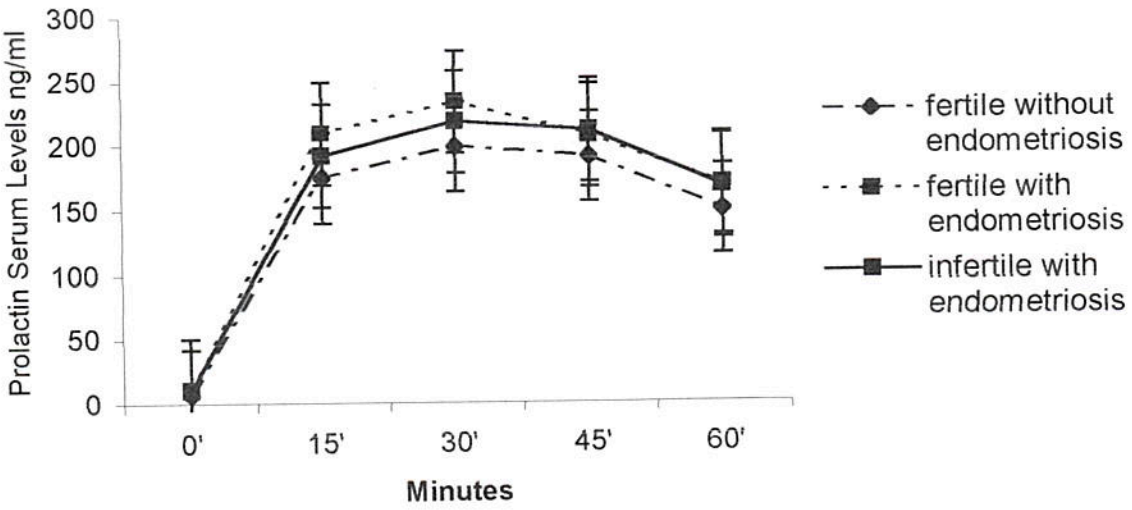


Figure 2: Prolactin serum levels after metoclopramide infusion, median ± SD.

P>0.05/KW test.

Table 2: Growth hormone serum levels (ng/ml) after metoclopramide administration, medians and 95% CI:

	Fertile without endometriosis	Fertile with endometriosis	Infertile with endometriosis	Statistics Kruskal-Wallis P
Basal	0.48	1.88	0.24	0.403
	(0.13-2.60)	(1.20-2.10)	(0.21-2.60)	
15 minutes	0.50	1.80	0.52	0.598
	(0.18-2.00)	(0.67-1.90)	(0.18-2.40)	
30 minutes	0.35	1.80	0.94	0.367
	(0.17-1.30)	(0.20-2.00)	(0.21-2.00)	
45 minutes	0.60	1.95	0.74	0.344
	(0.22-1.00)	(0.70-2.10)	(0.25-3.00)	
60 minutes	0.48	1.85	0.56	0.518
	(0.18-1.40)	(0.40-2.40)	(0.29-3.90)	

Table 3: Growth hormone serum levels (ng/ml) after TRH infusion, medians and 95% CI:

	Fertile without endometriosis	Fertile with endometriosis	Infertile with endometriosis	Statistics Kruskal-Wallis P
Basal	0.43	1.65	1.20	0.194
	(0.51-2.10)	(0.25-3.56)	(1.05-4.05)	
15 minutes	0.52*	2.15*	1.50	0.018
	(0.44-1.24)	(0.88-3.03)	(0.99-3.84)	
30 minutes	0.36**	1.70**	0.80	0.026
	(0.34-0.78)	(0.73-2.11)	(0.57-2.02)	
45 minutes	0.32	0.83	0.34	0.130
	(0.24-0.71)	(0.38-1.73)	(0.27-1.05)	
60 minutes	0.27	0.47	0.28	0.198
	(0.23-0.57)	(0.23-1.51)	(0.86-4.03)	

*P>0.05, Dunn procedure.
**P<0.05, Dunn procedure.

CONCLUSÕES

- PACIENTES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA E LEVE APRESENTAM MAIOR PREVALÊNCIA DE HIPERPROLACTINEMIA.

- MULHERES INFÉRTEIS COM ENDOMETRIOSE MÍNIMA E LEVE APRESENTAM MAIOR PREVALÊNCIA DE INSUFICIÊNCIA LÚTEA.

- PACIENTES COM ENDOMETRIOSE APRESENTAM NÍVEIS SÉRICOS BASAIS DE PROLACTINA SUPERIORES AOS DAS MULHERES FÉRTEIS SEM ENDOMETRIOSE.

- PACIENTES INFÉRTEIS COM ENDOMETRIOSE APRESENTAM NÍVEIS SÉRICOS DE ESTRADIOL INFERIORES.

- EXISTE UMA ALTERAÇÃO NA SECREÇÃO DE PROLACTINA APÓS INFUSÃO DE TRH EM MULHERES COM ENDOMETRIOSE.

- APÓS ADMINISTRAÇÃO DE METOCLOPRAMIDA (INIBIÇÃO DOPAMINÉRGICA) NÃO EXISTE UMA ALTERAÇÃO SIGNIFICATIVA NOS NÍVEIS SÉRICOS DE PROLACTINA EM PACIENTES FÉRTEIS E INFÉRTEIS COM ENDOMETRIOSE.

- A SECREÇÃO DO HORMÔNIO DO CRESCIMENTO FAZ-SE DE FORMA EXACERBADA, APÓS INFUSÃO DE TRH, EM MULHERES FÉRTEIS COM ENDOMETRIOSE, ESTA ALTERAÇÃO PODE ESTAR RELACIONADA A UM MAIOR ESTÍMULO ESTROGÊNICO.

- APÓS ADMINISTRAÇÃO DE METOCLOPRAMIDA, A SECREÇÃO DO HORMÔNIO DO CRESCIMENTO NÃO DIFERE ENTRE OS GRUPOS DE MULHERES FÉRTEIS COM E SEM ENDOMETRIOSE E INFÉRTEIS COM ENDOMETRIOSE.

- A SECREÇÃO ANÔMALA DE PROLACTINA COM PROVÁVEL REPERCUSSÃO NA SÍNTESE DE ESTERÓIDES, OBSERVADA PELA REDUÇÃO DOS NÍVEIS SÉRICOS DE ESTRADIOL EM MULHERES INFÉRTEIS COM ENDOMETRIOSE, E O AUMENTO NA PREVALÊNCIA DE INSUFICIÊNCIA LÚTEA RELACIONAM-SE COM A INFERTILIDADE, UMA VEZ QUE NÃO EXISTE DANO TUBÁRIO NESTE GRUPO DE PACIENTES.

CONSIDERAÇÕES FINAIS

O presente estudo evidenciou um aumento da prevalência de insuficiência lútea e hiperprolactinemia em mulheres inférteis com endometriose mínima ou leve. Este grupo de pacientes também apresentou alterações na secreção de prolactina após estímulo com TRH e baixos níveis séricos de estradiol se comparados aos outros grupos.

O objetivo desta pesquisa foi estudar mulheres com infertilidade e endometriose sem dano tubário para propor um mecanismo que possa explicar a infertilidade neste grupo de pacientes. Os achados encontrados relacionam-se e associam-se com a falha reprodutiva descrita neste grupo de mulheres.

Esta tese não tem como objetivo explorar a gênese fisiopatológica destas alterações, porém podemos propor que a endometriose deve ser investigada e tratada como uma síndrome com uma série de alterações imunológicas, peritoneais, uterinas e hormonais.

O tratamento deste grupo, mulheres inférteis com endometriose sem dano anatômico, é bastante controverso e com resultados discordantes. Atualmente, preconiza-se a cauterização dos focos como primeira opção, porém, mesmo após este procedimento, o prognóstico reprodutivo destas pacientes é baixo.

Este estudo abre uma série de questionamentos e especulações novas neste sentido. Poderíamos, por exemplo, diminuir a tonicidade da secreção da prolactina administrando uma droga agonista dopaminérgica antes ou durante a

indução da ovulação, tentando melhorar o meio-ambiente hormonal destas pacientes.

Esta tese faz parte de uma linha de pesquisa, estudo hormonal de pacientes inférteis com endometriose mínima e leve, e seus resultados apenas abrem uma série de especulações sobre a fisiopatogênese dos distúrbios aqui relatados, assim como a investigação de outras alterações hormonais relacionados com a endometriose: a análise do fluido folicular, o estudo da receptividade endometrial (calcitonina) e de outros marcadores de função lútea (ocitocina e prolactina endometriais) e a avaliação da secreção de *LH* após estímulo estrogênico que podem contribuir para uma melhor compreensão e, consequentemente, tratamento destas pacientes.