

## *Study of serum hormones and sialic acid levels of non-responsive infertile hyperprolactinemic women to medical therapy*

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### **Abstract :**

The study was performed on (56) infertile hyperprolactinemic women who fail in response to the medical therapy age (20-35) years, and (30) normoprolactinemic women were performed as control . The patients were divided between two kinds of treatments ,as follow =group 1 consisted of patients treated with Bromocriptine alone ,5 mg of drug was taken orally for 10 days , group 2 were of treated patients with 5 mg of Bromocriptine associated with 50mg of clomiphene, the drugs were taken orally for 5 days respectively .

Serum hormones and sialic acid levels were measured .The results indicated that ,PRL and SA levels were significantly higher (  $p < 0.05$  ) in both groups 1 and 2 compared to control , while there is no difference (  $p < 0.05$  ) in FSH and LH levels of both groups 1 and 2 compared to control, except LH levels was higher in group 2 versus group 1 and control .

In conclusion :

- 1- since SA is present just the structure of the two isoforms of prolactin glycosylated prolactin (G-PRL) and big big prolactin (bbPRL) , it may be used as indicator to defined the type of elevated PRL and then , monitoring appropriate therapy .
- 2- the inappropriate treatment for these patients may be the cause of failure response

### **Introduction**

It is well known that, the serum prolactin levels ranged from 20 to 25 ng/ml (1).The elevation of serum prolactin level above the normal range is called hyperprolactinemia (2)

Hyperprolactinemia is one of the major causative factors of unovulatory sub fertility (3).There is a relationship between increased prolactin and decreased gonadotrophin secretion, a specific role for prolactin itself in suppressing(4).The causes of hyperprolactinemia include : the growth of a prolactin-producing adenoma; other tumour of the pituitary region which block in the inhibitory influence of the hypothalamus; certain endocrine disease; polycystic ovary syndrome and certain drug(5).There are three treatment options to increase fertility in anovulatory hyperprolactinemic women: surgery,

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dopamine agonists and ovarian stimulation (6).Bromocriptine is by far the most widely used drug. A daily dosage restores normal serum prolactin concentration in about 85% of patients , leading to restoration ovarian function (7). Clomiphene is the most effective treatment for women with normogonadotrophic anovulation ,in order to induced ovarian stimulation (5).

The monomeric prolactin (mPRL) is twenty three kilo dalton (23KD) a protein hormone and secreted by aclass of cells known as mamotrophs in the pituitary gland (8).In humans PRL consist of 199 amino acid with 3 sulfide bridges (9,10). Studies in the last few years have shown that, prolactin exist in several molecular forms, some arising from posttransilation modifications and others from genetically determine factors (11). The multiple forms of prolactin varied in their physiochemical and biological characteristic. These forms were comprised of glycosylated human prolactin and macroprolactin (12,13).

The previous other works suggested that, a positive relationship between serum sialic acid (SA) and various pathological condition.Thus,several different mechanisms may lead to increased sialic acid concentration (14).

The objective of the present study was to investigat the cause of non response of the infertility hyperprolactinemic women to the medical therapy .

### **Subjects and methods**

The present study was undertaken on 56 infertile hyperprolactinemic women who attended into Kamal Al-Samaray hospital for infertility. The patients were below the age of 35years. All patients included in the study were non responders to both the two classes of medical therapy and non responder patients were suffering from infertility, amenorrhea and menstrual irregularities. The non responder patients were divided into 2 groups, In group 1: The patients (n=26) don't respond to at all to the treatment with, Bromocriptine (Parlodel by Sandoz switzezerland) and the treatment was administrated orally in dose 5mg/day for 10 days. Group 2 : The patients (n=30)don't respond at all to the same treatment for Bromocriptine , followed by 50 mg Clomiphene (Clommid by marion merrel Daw-Italy),The treatment was administrated orally for 5 days. The normoprolactinemic women were based as control group (n=30).

Venous blood sample (5ml) were drawn from control and patients at 2-4 days (follicle phase) of menstrual cycle. Each samples were centrifuged for serum separation. The serum samples divided into two parts one of them for hormonal test (PRL,FSH and LH) which performed by enzyme immunoassay method using commercial kit (biomerik),and the second Part was kept at 25c<sup>0</sup> until use for sialc acid measurements, which performed according to resorcinol method described by Svennerholm (15),statistical analysis of data and and level of significance were analyzed by using T-test (16).

### **Results**

Hyperprolactinemic patients who had taken either Bromocriptine alone (Group 1) or followed by additional treatment with clomiphene (Group2) in this

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study , showed a significant difference ( $p<0.05$ ) in the serum PRL and SA levels , compared to control group . (Mean  $\pm$  S.E) serum PRL and SA levels were higher in both group1 and group2 (PRLG1/37.51 $\pm$ 2.6 ng/ml ;PRL G2/31.51  $\pm$  0.8 ng/ml; SAG1 /74.7  $\pm$ 2.9  $\mu$ g/ml; SAG2/73.2 $\pm$ 2  $\mu$ g/ml ) than in control (PRLng/ml15.8 $\pm$  0.7 and SA/61 $\pm$ 1.8  $\mu$ g/ml) respectively . The result was obtained when compared the serum LH level of group1 with control (9 $\pm$ 1.8 m IU /MI VS . 4.3 $\pm$ 0.3m IU/MI) , while the difference between group2 and control in the serum levels of LH were not significant,There were no significant difference between all groups in serum FSH levels. Serum PRL and LH levels of group2 were significantly (31.5 $\pm$ 0.8 m IU/ML and 4.6 $\pm$ 0.4m IU/ML ) ( $P<0.05$ ) . While no significance difference in the serum FSH and SA levels in both group2 and group1 (Table 1).

Table-1: The serum hormones and sialic acids levels in normoprolactinemic women (control) and treated hyperprolactinemic patients (G1 and G2).

	No.	Mean $\pm$ S.E.			
		PRL Ng/ml	FSH mIU/ml	LH mIU/ml	SA $\mu$ g/ml
Standard values		1.3-25	3.2-12	1.5-8	46-71
Control group	30	* 15.8 $\pm$ 0.9	6.4 $\pm$ 0.4	* 4.3 $\pm$ 0.3	* 61 $\pm$ 1.8
Group1	26	*/ 37.5 $\pm$ 2.6	6.8 $\pm$ 0.8	*/ 9 $\pm$ 1.8	* 74.7 $\pm$ 2.9
Group2	30	*/ 31.5 $\pm$ 0.8	5.3 $\pm$ 0.2	 4.6 $\pm$ 0.4	* 73.2 $\pm$ 2

\* $P< 0.05$  significantly different from control group.

•  $P< 0.05$  significantly different from control group 1.

## Discussion

Although the previous other works had reported that ,Bromocriptine treatment in adequate doses, lowers the plasma PRL level and restores the menstruation and ovulation in 80% of patients ( 17 ,18) the result of the present study reported that ,markedly reduced responsiveness to treatment has obtained in treated patients with Bromocriptine alone or associated patients with Clomiphene, whereas the serum PRL levels were significantly higher in both treated patients (Group 1 and 2) than normoprolactinemic women (control).In this study, the treatment with Bromocriptine /Clomiphene did not have the beneficial effects on restoration the serum PRL levels in to physiological range, In spite of the role of Clomiphene as adjunct to reduce hyperprolactinemia during ovarian hyperstimulation.This result consistent with the previous

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research that had shown Clomiphene unable to reduce luteal phase hyperprolactinemia (19).

The significant rising in LH levels of G1 in the present study may be due to Clomiphene administration.

As well as ,our data indicated that the serum sialic acid levels were higher in both treated G1 and 2 than control. This finding is consist with the recent previous work that had shown the rising serum sialic acid in the untreated hyperprolactinemic women (20).

Sialic acid residues were present on multiple oligosaccharide of FSH molecule (21), and the two forms of Prolactin (glycosylated Prolactin (G-PRL)) and macroprolactin (big big PRL/bbPRL)carbohydrate content of G-PRL form was found to be consist with N-linked oligosacchrides of mannos-rich structure and complex units terminated in sialic acid (22) . macromolecular (bbPRL) form of Prolactin has been characterized as complex of PRL with Ig G anti-body (23). In addition to,previous reports demonstrated that,the high content of sialic acid is present in glycans structure of IgG (24)Therefore, in this study, the elevation of serum sialic acid level may be due to the presence one of two forms of G-PRL and bbPRL, but unlikely to be derived from FSH molecules ,Scince our data showed that ,the serum FSH Level in treated patients of both groups 1 and 2 are found to be similar to those reported in control but G2. In our study gather elevation in the serum PRL and SA levels confirm this explanation.

Concluded from our results that, sialic acid can be useful indicator for diagnosis the types of hyperprolactinemia and then ,monitoring appropriate therapy. The failure response may be due to the unappropriat treatment.

We recommend that , further studies must be performed to define the exact pathophysiological mechanisim of non responders in order to develop affective treatment.

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## دراسة مستوى الهرمونات و حمض السياليك في مصل النساء العقيمات المصابات بفرط البرولاكتين غير المستجيبات للعلاج الطبي

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### الخلاصة :

استخدمت في الدراسة 56 امرأة عقيمة مصابة بفرط البرولاكتين واللواتي أخفقن في الاستجابة للعلاج الطبي ،بعمر ( 20-35 ) سنة ، واستخدمت 30 امرأة سوية البرولاكتين ( كسيطرة ) .  
قسمت المريضات استنادا إلى نوع العلاج الذي كن قد تناولنه إلى مجموعتين كالآتي :  
\*المجموعة الأولى : نساء تتاولن (5) مللي غرام من دواء البروموكريبتين فمويا لمدة ( 10 ) أيام.  
\*المجموعة الثانية : نساء تتاولن ( 5) مللي غرام من البروموكريبتين بالارتباط مع (50) مللي غرام من دواء الكلوميدين فمويا لمدة (5) أيام.

تم قياس مستوى الهرمونات وحمض السياليك في مصل دم المريضات ، أشارت النتائج إلى حصول ارتفاع معنوي ( $P<0.05$ ) في مستوى آل PRL و آل SA في كلتا المجموعتين 1 و2 بالمقارنة مع السيطرة . بينما لم يسجل فرق معنوي ( $P<0.05$ ) في مستوى آل FSH و آل LH لكلتا المجموعتين 1 و2 بالمقارنة مع السيطرة باستثناء ارتفاع معنوي في مستوى آل LH في المجموعة 2مقابل المجموعتين أو السيطرة.

### الاستنتاج:

- 1- لكون حمض السياليك يدخل في تركيب البرولاكتين في نمط (bbPRL,GPRL) فقط فمن الممكن أن يستخدم كمؤشر لتحديد نمط البرولاكتين المرتفع ومن ثم ضبط العلاج المناسب .
- 2- أن العلاج غير الملائم لهؤلاء المريضات ربما هو سبب أخفاق الاستجابة .