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The Relationship of Circulating Prolactin Levels to The Size of Primary Tumor in Breast Cancer Patients

[Meme Kanserli Hastalarda Primer Tümör Boyutu ile Dolaşımdaki Prolaktin Düzeylerinin İlişkisi]

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ABSTRACT

Rationale: data about the role of prolactin (PRL) in breast cancer patients are controversial. This hormone might potentially play an important role in breast cancer initiation and development in rodents, as well as, at least partly, in humans. The aim of this study was to investigate the possible relationship between circulating levels of PRL and parameters of primary tumor in breast cancer patients.

The main experimental group consisted of 46 female patients with histologically confirmed diagnosis of breast cancer. There were two control groups: apparently clinically healthy women (40), and female patients with other types and locations of cancer (33). Baseline levels of PRL were determined both in the main and in control groups. Parameters of primary tumor (histological diagnosis, degree of differentiation, location, and size) have been determined. Results were processed by means of nonparametric tests.

The circulating levels of PRL before treatment were significantly higher in breast cancer patients in comparison to controls. The average size of the primary tumor in breast cancer patients with hyperprolactinemia before treatment was significantly higher ($U=125.5$, $p<0.01$) than in those with normoprolactinemia. The calculated correlation coefficient between PRL and the size of primary tumor in hyperprolactinemic breast cancer patients was statistically significant ($r=0.68$, $p<0.0001$). In normoprolactinemic, and in patients with other locations of cancer such a correlation did not exist.

Serum levels of PRL are, probably, directly dependable on the size of primary tumor in breast cancer patients, especially in those with hyperprolactinemia, but this is not differentiation dependent phenomenon.

Key Words: prolactin, breast cancer, primary tumor.

ÖZET

Meme kanserinde prolaktinin (PRL) rolü konusundaki veriler çelişkilidir. Bu hormon meme kanserinin başlaması ve gelişmesinde, insanda kısmen, kemirgen hayvanlarda ise önemli bir role sahip olabilmektedir.

Bu çalışmanın amacı PRL'nin dolaşımdaki seviyeleri ile meme kanseri hastalarının primer tümör parametreleri arasındaki olası ilişkiyi araştırmaktır. Deneysel grubu meme kanseri teşhisleri histolojik olarak onaylanmış 46 kadın hasta oluşturmaktadır. İki kontrol grubu ile çalışılmıştır: klinik olarak sağlıklı bulunan kadınlar (40) ve diğer kanser tiplerinden birine sahip kadın hastalar (33). Primer tümör parametreleri (histolojik tanı, farklılaşmanın derecesi, yerleşimi ve büyüklüğü) saptanmış ve sonuçlar nonparametrik testler kullanılarak değerlendirilmiştir.

Tedaviye başlanmadan önce ölçülen PRL'nin dolaşım düzeyleri, kontrol gruplarıyla karşılaştırıldığında meme kanserli hastalarda önemli derecede yüksek bulunmuştur. Tedavi öncesi hiperprolaktinemili meme kanseri hastalarının primer tümörlerinin ortalama boyutu ($U=125.5$, $p< 0.01$), normoprolaktinemiye sahip olanlardan önemli derecede daha büyüktür. Hiperprolaktinemili meme kanseri hastalarında primer tümör büyüklüğü ile PRL arasındaki hesaplanan korelasyon sabiti istatistiksel olarak önemlir ($r = 0.68$, $p< 0.0001$). Normoprolaktinemiye sahip olan meme kanserli hastalar ve diğer tümor tiplerine sahip olan hastalarda bu korelasyon tespit edilmemiştir.

Özellikle hiperprolaktinemili meme kanseri hastalarında primer tümör büyüklüğünün PRL'nin serum düzeylerine doğrudan bağlı olabildiği, ama bu durumun farklılaşmaya bağlı bir durum olmadığı söylenebilir.

Anahtar Kelimeler: prolaktin, meme kanseri, primer tümör

INTRODUCTION

Normal breast tissue cells are sensitive to a number of hormones among which estrogens and prolactin (PRL) have the most dramatical effect. Malignant breast tissue cells can behave in this respect similar to normal cells. This mostly depends on the degree of their differentiation and growth. More differentiated cells usually possess hormone receptors on their membranes very much alike normal cells, and according to that, can respond to normal hormonal stimulation. Less differentiated cells escape normal hormonal regulation from loss of hormone receptors.

Data about the function of PRL in mammary neoplasia are controversial. PRL was first recognised as a hormone that might significantly contribute to both the pathogenesis and progression of rodent mammary neoplasia (1). It is a major tumor mitogen in rodents, and excess PRL has been shown to induce breast cancer in mice (2, 3). The role of PRL in human breast cancer has been poorly understood until recently. The contribution of PRL to the pathogenesis and progression of human breast cancer at the cellular, transgenic, and epidemiological levels is increasingly appreciated. There is firm evidence documenting a direct stimulatory effect of PRL on mammary epithelial cells (4), and breast cancer cells (5, 6, 7) in vitro. Acting at the endocrine and autocrine/paracrine levels, PRL functions to stimulate the growth and motility of the human breast cancer cells (8). The majority of breast cancers express prolactin receptor (PRLR), meaning PRL was active in the tumors (8, 9, 10). The PRL/PRLR complex associates with, and activates several signaling networks that are shared with other members of the cytokine receptor superfamily (10). It was recently identified intranuclear function of PRL in the initiation of the human breast cancer (11, 12). There also are some epidemiological data suggest a relatively strong positive assotiation between circulating levels of PRL and breast cancer risk in women (13, 14).

With the goal of evaluation of clinical usefullness of PRL as a potential marker in breast cancer patients we examined correlations of its circulating levels with some parameters of primary tumor (size, and degree of differentiation).

PATIENTS AND METHODS

Patients

It was prospective randomised study with defined cryteria for patients inclusion.

The inclusion cryteria

A. Breast cancer patients:

- Non or occasional smokers,
- Non alcoholic drinkers,
- Occasional or no coffee drinkers,
- No other drug users,
- No other major illnesses,
- Histologically proven breast cancer,
- Currently under no treatment,
- Age interval 38-65 years,
- General clinical status – fair.

B. Other cancer patients:

Besides the major cancer illness, other cryteria were exactly the same as in main experimental group.

C. Healthy control group:

The inclusion cryteria for the healthy control group, besides the absence of malignant or any other major debilitating disease, were the same as in other two groups.

The main experimental group consisted of 46 female histologically confirmed breast cancer patients with an age interval 38-65 years. The circulating levels of PRL were measured in all of them before any therapy (baseline levels), and then at regular time intervals during the observation period. Parameters of primary tumor (histological diagnosis, degree of differentiation, location, and size) have been determined in all of them. This group was further divided into two subgroups according to their serum PRL before therapy: hyperprolactinemic ones (23 patients), with average circulating PRL of more than 520 mU/L; and normoprolactinemic (23 patients), having PRL between 52 and 520 mU/L.

There were two control groups designated as I, and II. Group I consisted of 40 clinically healthy women with an age interval 38-65 years. All of them had normal mammograms. The circulating levels of PRL were measured in all of them at least three times during the observation period.

Group II consisted of 33 female patients having cancer of different histological origin and location with an age interval 38-65 years. Majority of them have colon and lung cancer. The same data as in the main group have been collected in all of them. Circulating levels of PRL before any therapy were measured in all of them, and later again at regular time intervals during the observation period.

Estimation of primary tumor parameters

Histological diagnosis was performed on fresh specimens obtained by means of needle biopsy, or on specimens taken at surgery. Degree of differentiation was estimated according to well established criteria (15). Size of primary tumor was determined first by palpation, and then measured with a caliper always in two right angle perpendicular diameters. The estimated size was

in most cases replaced by data given in written pathological reports from actual surgical operations.

Determination of blood levels of PRL

Blood samples were drawn under sterile conditions at eight hours in the morning each time, centrifuged at 3000 rpm for 10 minutes under room temperature, and serum was stored at –20°C until processed. The circulating levels of PRL were determined by means of radioimmunoassay (RIA) method using commercially avaliable kits (Serono). Range of normal values was 52-520 mU/L.

Statistical analysis of results

The nature of distribution of quantitive variables was checked using distribution histograms with P-P (probability plots), and detrended probability plots. The results were evaluated using nonparametric Mann-Whitney test, and Spearman's correlation coefficient using two sided significance of p values, with calculated mean values and standard errors. The p values less than 0.05 were considered as statistical significant. All results were processed in SPSS version 10.

RESULTS

The circulating levels of PRL

Baseline levels of PRL were significantly higher (U=765, p<0.04) in breast cancer patients than in healthy women, and they also were higher (U=768, p<0.02) than in patients with other locations and histologic types of cancer (table I). None of women from healthy control group was hyperprolactinemic. There was also no correlation between age and PRL levels and between menopausal status and levels of PRL in this control group.

The circulating levels of PRL and the parameters of primary tumor

Thirty five patients (76.08%) had intraductal invasive carcinoma, four (8.69%) scirrhous, two (4.37%) medulary, two (4.37%) lobular, one (2.17%) scirrhous infiltrative, and one (2.17%) anaplastic carcinoma. One patient (2.17%) had breast fibrosarcoma.

The average size of primary tumor in hyperprolactinemic breast cancer patients was significantly greater (U=125.5, p<0.01) in comparison to normoprolactinemic ones (table II).

The calculated correlation coefficient between PRL levels before treatment and the size of primary tumor was r=0.37 with p<0.01 in the group of all breast cancer patients, and r=0.68 and p<0.0001 in hyperprolactinemic breast cancer patients (table III). It means that there was a slight positive stohastic correlation between these two variables in the group of all breast cancer patients, and a positive correlation in hyperprolactinemic ones, and calculated correlation coefficients were statistically significant. It was not the case either in normoprolactinemic (r=-0.15, p<0.5), or in patients with other types and locations of cancer (r=0.09, p<0.63) (table III).

The coefficient of correlation between circulating levels of PRL and degree of differentiation was r=0.51 with p<0.01 in normoprolactinemic breast cancer patients (table IV). It means that there was a slight positive correlation between these two variables, but calculated correlation coefficient was statistically significant. Such a correlation was not detected either in hyperprolactinemic (r=-0.33, p<0.12) or in other cancer patients (r=-0.07, p<0.73) (table IV).

Table I. The average circulating levels of prolactin (PRL) in healthy women (control group I), in patients with different locations and histologic types of cancer (control group II), and in women with breast cancer.

Group/number	PRL (mU/L) mean±S.E.	PRL<520mU/L	PRL>520mU/L	U*	p
		Number/total number (%)			
Control group I 40	442±17.7[a]	40/40 (100)	0/40 (0)	765	0.04 (c vs. a)
Control group II 33	481±51.6[b]	24/33 (73)	9/33 (27)		
Breast cancer patients 46	610±65.3[c]	23/46 (50)	23/46 (50)	768	0.02 (c vs. b)

* Mann-Whitney U value

Table II. The average size of primary tumor in breast cancer patients with hyperprolactinemia and normoprolactinemia.

Group/number	PRL (mU/L) mean±S.E.	Size of primary tumor (cm^2)	U*	p
Hyperprolactinemia 23	929±87	11.4±2.4[a]	125.5	0.01 (a vs. b)
Normoprolactinemia 23	292±25	6.1±0.7[b]		

* Mann-Whitney U value

PRL=prolactin

DISCUSSION

The average circulating levels of PRL before treatment were significantly higher in breast cancer patients than in healthy controls, and in patients with other types and locations of cancer (table I). This difference points towards the specificity of PRL for breast cancer which leads to its diagnostic and prognostic importance in this disease.

The size of primary tumor was significantly greater in

Table III. The correlation between the circulating levels of prolactin (PRL) before treatment and the size of primary tumor in breast cancer patients and in patients with other types and locations of cancer.

Group/number	PRL (mU/L) mean±S.E.	Size of primary tumor (cm^2)	r*	p
All breast cancer patients	610.5±65.3	8.8±1.3	0.37	0.01
Hyperprolactinemia	929.0±87.0	11.4±2.4	0.68	0.0001
Normoprolactinemia	292.0±16.5	6.1±0.7	-0.15	0.5
Other cancer patients	524.1+-65.2	18.3±3.8	0.09	0.63

* Spearman's correlaton coefficient

Table IV. The correlation between the circulating levels of prolactin (PRL) before treatment and the degree of differentiation of primary tumor in breast cancer patients and in patients with other types and locations of cancer.

Group/number	PRL (mU/L) mean±S.E.	Degree of differentiation**	r*	p
All breast cancer patients	689.8+-62.5	1.78±0.11	0.01	0.93
Hyperprolactinemia	927.4+-108.4	1.73±0.15	-0.33	0.12
Normoprolactinemia	488.9+-25.5	1.88±0.17	0.51	0.01
Other cancer patients	524.1+-65.2	1.70±0.16	-0.07	0.73

* Spearman's correlaton coefficient

** 1=well differentiated, 2=medium differentiated, 3=poorly differentiated

hyperprolactinemic breast cancer patients in comparison to those with normoprolactinemia (table II). There was a significant positive correlation between the baseline levels of PRL and the size of primary tumor in all breast cancer patients and, especially, in hyperprolactinemic ones, while in normoprolactinemic patients and in patients with various other cancers such a connection did not exist (table III). There was no correlation between the PRL levels and the degree of differentiation of primary tumor either in the group of all breast cancer patients or in hyperprolactinemic ones. The correlation was found only in the group of normoprolactinemic patients (table IV). These findings support the hypothesis that breast cancer cells can synthesize and secrete PRL. The circulating levels of PRL are thus directly dependable on the size of primary tumor, but this is not differentiation dependent phenomenon which means that either cells (dedifferentiated and mature) can synthesize and secrete this hormone. This finding is in accordance with other results published before (7, 9, 10, 16, 17). The results in this paper also suggest potential stimulative effect of PRL on the growth of breast cancer cells but the mechanism of action as well as the origin of circulating PRL and its effect on breast cancer cells are still unclear.

It is now recognize that both endocrine and autocrine/paracrine sources of PRL exist in mammals (17). It has already been documented that PRL plays an important role in the initiation and differentiation of breast cancer cells in rodents (2, 3, 10, 17), and in humans in vitro (10, 17), but its role as an etiologic factor in the initiation of breast cancer in humans has not been firmly documented. However, the results of other studies support the conclusion that PRL can be synthesized and secreted by breast cancer cells and that it can act as a local growth promotor in the surrounding of these cells (7, 18, 19). It remains to be clarified whether for the stimulation of growth of breast cancer cells is responsible autocrine PRL, or endocrine PRL secreted by pituitary or other tissues, or possible stress, or all of them. The molecular structure of the PRLR seems to remain intact in tumor tissue, and systemic and local production of PRL may participate in tumor cell growth and proliferation through functional receptors (20). Results in our study document a positive correlation between the size of primary tumor and the circulating levels of PRL, favoring the opinion that for the development and potentiation of the growth of breast cancer cells more responsibility lies on the side of endocrine (hypophyseal) PRL. However, the origin of circulating PRL in breast cancer patients is not clear. It is not known yet whether it originates exclusively from the pituitary or it also might be a product of breast cancer cells. Furthermore, it still remains to be clarified whether hyperprolactinemia is the result or the cause of breast cancer. The fact that antihyperprolactinemic drugs are very effective in lowering the circulating levels of PRL but, in majority of cases, ineffective in controlling the development of breast cancer (17) backs up the hypothesis that both endocrine and autocrine PRL are involved in breast carcinogenesis. Probably, the best situated explanation is that endocrine PRL is responsible for initiation, and autocrine for perpetuation of carcinogenic process.

Results of our study document positive correlation between the size of primary tumor and the circulating levels of PRL favoring the conclusion that serum levels of PRL are, probably, directly dependable on the size of primary tumor in breast cancer patients, especially in those with hyperprolactinemia, but this is not differentiation dependent phenomenon.

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