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Relationships between the concentrations of epidermal growth factor, insulin-like growth factor-I, estrogens and progesterone receptors in women breast cancer, and the histologic type of tumor and its grade of malignancy

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Introduction. Receptors of epidermal growth factor (EGF-R), insulin-like growth factor-I (IGF-I-R), estrogens (ER) and progesterone (PR) are believed to be predictive and prognostic indices in breast cancer in women. The objective of the study was to explain the relationships between receptors of EGF-R, IGF-I-R, ERc and PRc in breast carcinoma and the morphological features of these tumours.

Material and methods. 408 women with breast carcinoma (incl. 304 ductal carcinomas, among them 16 in situ carcinomas and 288 invasive carcinomas). Other types were: lobular, medullary, mucinous and mixed carcinomas. The grade of histological malignancy of ductal carcinoma was determined by Bloom-Richardson method and its modified version. The concentrations of EGF-R, IGF-I-R, ERc and PRc were determined by biochemical radiocompetitive methods.

Results. In ductal carcinomas in situ the concentrations of ERc and PRc were evidently higher than in infiltrating ductal carcinomas. On the other hand, EGF-R concentrations in in situ carcinomas were only half the value of that in infiltrating ductal carcinomas. In analogous situations IGF-I-R concentrations were higher but were not differing statistically significant between them.

In ductal carcinomas ERc and PRc concentrations correlated inversely with the grade of histological malignancy. Between EGF-R concentrations of ductal carcinomas and the grade of histological malignancy of tumours a statistically significant positive correlation was found in premenopausal women only, while in postmenopausal women such a correlation did not exist. There was a tendency towards inverse correlation between IGF-R concentration and grade of malignancy of ductal carcinomas but it did not reach statistical signifficance.

Lobular and mucinous carcinomas contained high concentrations of ERc and PRc. Medullary carcinomas had very low ERc concentrations and high concentrations of PRc, EGF-R and IGF-I-R. Mucinous carcinomas contained high concentrations of IGF-I-R but low concentrations of EGF-R.

Conclusions. The data indicates that ductal carcinomas of lower grade of malignancy have higher ERc and PRc concentrations but lower EGF-R concentration. In general higher malignancy tumours are characterised by a decrease in ERc and PRc concentrations and an increaes in EGF-R concentration. IGF-I-R concentration in ductal carcinomas has a tendency to decrease with the grade of malignancy but this fails to reach statistical significance.

Zależności między stężeniami receptorów naskórkowego czynnika wzrostu, insulinopodobnego czynnika wzrostu I, estrogenów i progesteronu w rakach piersi u kobiet, a typem nowotworu i stopniem jego złośliwości histologicznej

Wprowadzenie. Receptory naskórkowego czynnika wzrostu (EGF-R), insulinopodobnego czynnika wzrostu I (IGF-I-R), estrogenów (ER) i progesteronu (PR) są uważane za rokownicze i predykcyjne wskaźniki raka piersi u kobiet. Celem pracy było zbadanie relacji między receptorami EGF-R, IGF-I-R, ERc i PRc występującymi w rakach piersi, a ich morfologicznymi właściwościami.

Materiały i metody. Badania przeprowadzono na 408 rakach piersi kobiet, wśród których było 304 raków przewodowych, 16 raków in situ, a 288 raków inwazyjnych. Inne typy raków to: raki zrazikowe, rdzeniaste, śluzotwórcze i mieszane. Stopień złośliwości histologicznej przewodowych raków piersi określano metodą Blooma i Richardsona oraz jej zmodyfikowaną wersją. Stężenia receptorów EGF-R, IGF-I-R, ERc i PRc oznaczano biochemicznie metodami radiokompetycyjnymi.

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Wyniki. W przewodowych rakach piersi in situ stężenia ERc i PRc były wyraźnie większe, niż w przewodowych rakach naciekających. Natomiast stężenia EGF-R w rakach in situ były o połowę mniejsze niż w przewodowych rakach naciekających. W analogicznych sytuacjach stężenia IGF-I-R były znaczące, lecz statystycznie nie różniły się między sobą.
W przewodowych rakach stężenia ERc i PRc były odwrotnie skorelowane ze stopniem ich histologicznej złośliwości. Między stężeniami EGF-R, a stopniem histologicznej złośliwości przewodowych raków piersi u kobiet młodszych istnieje statystycznie znamienna dodatnia korelacja, natomiast w rakach kobiet starszych korelacja ta nie osiągnęła poziomu znamienności. Między stężeniami IGF-R i stopniem złośliwości histologicznej raków przewodowych była tendencja do odwrotnej korelacji, ale nie osiągała ona statystycznej znamienności. Raki zrazikowe i śluzotwórcze cechowały się przede wszystkim dużymi stężeniami ERc i PRc. Raki rdzeniaste miały bardzo małe stężenia ERc i duże stężenia PRc, EGF-R i IGF-R. Raki śluzotwórcze charakteryzowały się dużymi stężeniami IGF-I-R, a małymi stężeniami EGF-R.
Wnioski. Przedstawione w obecnej pracy dane świadczą o tym, że przewodowe raki piersi, o mniejszej złośliwości histologicznej, posiadają większe stężenia ERc i PRc, natomiast mniejsze stężenia EGF-R. Generalnie, w miarę złośliwienia nowotworu stężenia ERc i PRc maleją, a stężenia EGF-R narastają. Stężenia IGF-I-R w rakach przewodowych, wraz ze wzrostem złośliwości histologicznej, mają tendencje do zmniejszania się, ale różnica nie osiągała znamienności statystycznej.

Key words: Epidermal growth factor receptor, insulin-like growth factor receptor, estrogen receptor, progesterone receptor, histological grade of malignancy, breast cancer
Słowa kluczowe: receptor naskórkowego czynnika wzrostu, receptor insulinopodobnego czynnika wzrostu I, receptor estrogenów, receptor progesteronu, stopień złośliwości histologicznej, rak piersi

Introduction

Receptors of epidermal growth factor (EGF-R), insulin-like growth factor-I (IGF-I-R), estrogens (ER) and progesterone (PR) are believed to be predictive and prognostic indices in breast cancer in women. Among these biochemical indices cytosol receptors of estrogens (ERc) and progesterone (PRc) are most common. Low concentrations or absence of these receptors in tumours are thought to be an unfavourable, while high concentrations are considered a favourable prognostic factor for endocrine therapy and general prognosis [1, 2]. Concomitant presence of ERc and PRc in a tumour is regarded as a more favourable therapeutic and prognostic prediction, while the absence of both receptors is unfavourable prognostic [3, 4]. The cellular functions of ER and PR are closely connected with receptors of growth factors, such as EGF-R and IGF-I-R, the concentrations of which are regulated by estrogens and progesterone [5, 6]. Moreover, EGF-R concentrations correlate inversely and those of IGF-I-R correlate positively with ERc and PRc [7--11]. Generally, EGF-R is considered as an unfavourable and IGF-I-R – prognostically favourable. Recently, however, we have demonstrated that breast cancers containing high concentrations of EGF-R have unfavourable prognosis only if the tumour contains no ERc and PRc. Prognosis is better if ERc, PRc and EGF-R are present simultaneously in the tumour [3, 12].

Biochemical prognostics of a functional character are connected with the morphologic pattern of breast cancer. Among morphologic parameters the most important are: histologic type and grade of malignancy evaluated mainly acc. to the method of Bloom and Richardson [13].

The diagnostic and prognostic significance of the morphologic features of breast cancer and concentrations of ERc, PRc, EGF-R, IGF-I-R have always raised queries as to the relations between them. As to ERc and PRc the prevailing view is that there exists a negative correlation between their concentrations in breast cancer and the grade of malignancy [14, 15]. Controversies exist, however, as to the relations between the concentrations of the receptors of growth factors (EGF-R, IGF-I-R) and histologic grade. Some authors describe such a correlation [7, 15-17] while others have failed to find it [8, 18-21].

The objective of the present study was to search for a relation between four receptors (EGF-R, IGF-I-R, ERc and PRc) found in breast cancer and the morphologic features of the tumours. These relationships are not yet sufficiently known.

Material and methods

The material consisted of 408 specimens of breast cancer (maximal number of tumours) obtained from women treated in the Oncology Center in Warsaw. The concentrations of epidermal (EGF-R) and insulin-like-I (IGF-I-R) growth factors receptors, and cytosol receptors of estrogens (ERc) and progesterone (PRc) were determined in the cancer tissues. It was attempted, whenever possible, to evaluate all four receptors in the same tumour.

Preparation of membrane and cytosol fractions from breast cancers

Tumours obtained during surgery and frozen in liquid nitrogen were powdered in a vibrating mill (Micro-Dismembrator II). The obtained powder was suspended in TEDG buffer (10 mM Tris-HCl, 1.5 mM EDTA, 0.5 mM DTT, 10% glycerol, pH = 7.4), homogenised by means of Polytron and left in ice for 20 minutes, stirring several times. The homogenate was spun at 800 x g for 10 minutes, the sediment (crude nuclear fraction) was discarded, and supernatant was centrifuged again at 220 000 x g for 30 minutes. The obtained sediment (crude membrane fraction) was washed with phosphate buffer from the remaining cytosol protein and was taken for the determination of epidermal growth factor receptor (EGF-R) and insulin-like growth factor receptor-I (IGF-I-R). In the supernatant obtained by ultracentrifugation (cytosol fraction) estrogen and progesterone receptors were determined. In the membrane fraction and cytosol fraction protein concentration was determined according to the method of Lowry [22].

Determination of estrogens and progesterone receptors in cytosol fraction

The estrogens and progesterone receptors were determined in the cytosol fraction by the radiocompetitive charcoal-dextran method in version of one point assay analysis at maximal saturating concentrations of labelled hormones. Briefly, triplicate samples of cytosol (0.1 ml) from human breast cancer were incubated overnight at 4°C with labelled estradiol (7nM $^{3}[H]E_2$,) or with labelled progestagen (10 nM ^{3}[H]ORG2058) in presence or absence of 100-fold molar excess of non-labelled competitor – diethylstilbestrol (DES) or ORG2058. After that to the samples 0.2 ml of dextran-coated charcoal suspension (0.5 % and 5% respectively) was added. Incubation at 4°C with shaking was prolonged for 10 min. and specimens were then centrifuged at 9000 x g for 15 min. at 4°C. 150 µl of supernatant was poured into vials containing the scintillation mixture and radioactivity was counted in a Beckman scintillation counter. The specific binding of estradiol (E_2) or progestagen (ORG2058) in the cytosol was calculated from the difference between the total binding and the non-specific binding. Specific binding sites were expressed as fmols/mg protein and were assumed to be as appropriate receptor – ERc or PRc. Detailed description can be found in the publication of Paszko et al. [23, 24].

Determination of epidermal growth factor receptors

For EGF receptor determination we applied the radiocompetitive method. The EGF receptor was determined in the membrane fraction using one-point assay analysis at a maximal saturating concentration (4nM ^{125}I-EGF) using triplicate samples for total and unspecific bindings. 100 µl of the suspension containing an equivalent of 100 µg of membrane protein was incubated with ^{125}I-EGF in the presence or absence of a100 times higher concentration of non-labelled EGF (in relation to ^{125}I-EGF) for 1 h at room temperature. To terminate the reaction 2 ml of cold phosphate buffer (0.025 M, pH – 7.0) were added to the samples and the suspension was spun at 6000 x g for 45 minutes. The supernatant containing ^{125}I-EGF not bound to cell membranes was discarded and radioactivity of ^{125}I-EGF bound to membranes was measured in the sediment. The results of EGF-R determinations were expressed in fmols/mg of membrane protein. Specific binding of ^{125}I-EGF to cell membranes was calculated from the difference between total and non-specific bonds. The details of EGF-R determinations were described earlier [9, 11].

Determination of insulin-like growth factor-I receptors

The IGF-I receptor was determined in the membrane fraction of breast cancers using one-point assay analysis at the maximal saturating concentration (0,35 – 0,40 nM ^{125}I IGF-I) using triplicate samples for total and unspecific bindings. 100 µl of the suspension containing an equivalent of 100 µg of membrane protein were incubated with ^{125}I-IGF-I in the presence or absence of 150 times higher concentration of non-labelled IGF-I (in relation to ^{125}I-IGF-I) for 18 hours at 4°C. To terminate the reaction 2 ml of cold phosphate buffer (0.025 M, pH – 7.0) were added. The obtained suspension was spun at 6000 x g for 45 minutes. The supernatant containing ^{125}I-IGF-I not bound with membrane fraction was discarded and radioactivity of membrane-bound ^{125}I-IGF-I was measured in the sediment. Specific binding of ^{125}I-IGF-I to cell membranes was calculated from the difference between total and non-specific bonds. The results of receptor determinations were expressed in fmols/mg of membrane protein.

Tumour tissues were assumed to be positive for EGF-R, ERc and PRc when the concentrations of the respective receptors were equal to or greater than 10 fmol/mg of cytosol or cell membrane protein. For IGF-I-R the borderline was 4 fmol/mg membrane protein.

Histologic evaluation

The morphological studies included determination of the histological type of the tumours according to the present WHO classification [25]. In ductal carcinoma (non-specific) the histological grade of malignancy was determined by the method of Bloom-Richardson designated by us as B [13] and its modified version designated by us as SBR [26]. The results of the biochemical studies were compared with those of morphological examinations by means of statistical tests, such as the Wilcoxon and Kendall test using the Statgraphics statistical pack.

Results

In the material of 408 breast cancers 304 were ductal carcinomas, among them 16 preinvasive and 288 invasive carcinomas. Lobular carcinomas were found in 76 specimens, medullar carcinomas in 9, mucinous carcinomas in 6 and mixed (ductal-lobular) carcinomas in 13 cases. In these cancer groups the concentrations of EGF-R, IGF-I--R, ERc and PRc were determined. The results presented in Table I, II and Figure 1 show the values of concentra-

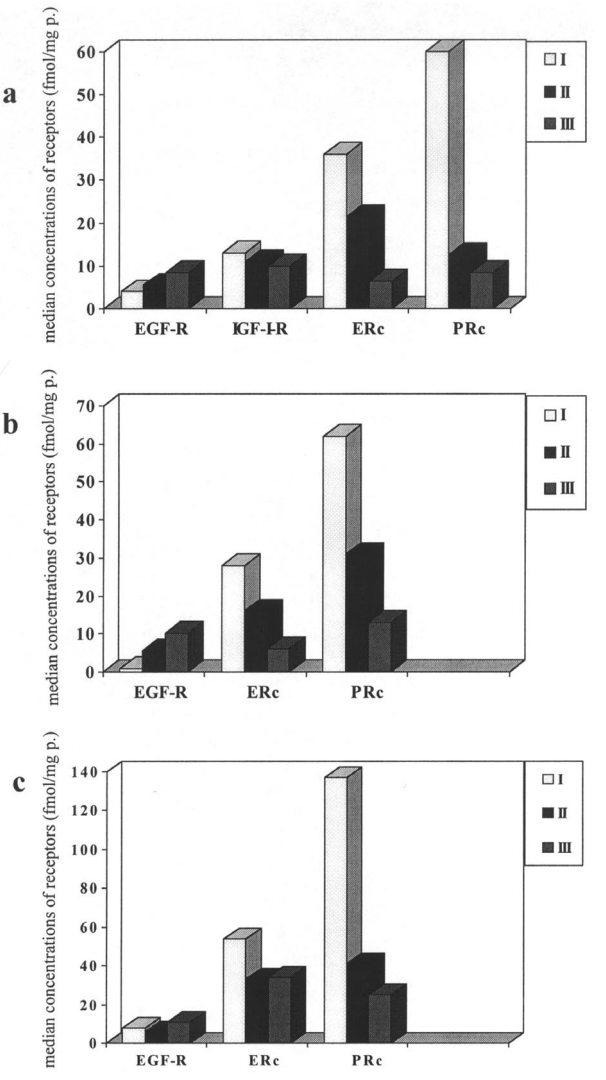

Figure 1. Concentrations of EGF-R, IGF-I-R, ERc, PRc and the degree of histological malignancy (I, II, III) of ductal breast carcinoma.
a – breast carcinomas of women aged 20-86 years
b – breast carcinomas of premenopausal women
c – breast carcinomas of postmenopausal women

Table I. Concentrations of estrogen (ERc), progesterone (PRc), epidermal growth factor (EGF-R) and insulin-like growth factor receptors-I (IGF-I-R) in various histologic types of breast carcinoma

Type of carcinoma		Receptors*			
		ERc	PRc	EGF-R	IGF-I-R
	**				
Ductal	A	132.2 ±65.8	126.9 ±45.4	4.1 ±3.4	14.4 ±2.5
carcinoma	M	25.5	54	0	13
in situ	R	0 – 1026	0 – 594	0 – 27	0 – 36
	n	16	16	8	16
Ductal	A	44.2 ±5.1	71.9 ±10.3	8.1 ±0.6	15.7 ±1.0
carcionoma	M	3	12	5	11
inv.	R	0 – 539	0 – 1939	0 – 58	0 – 134
	n	270	270	288	270
Statistic between groups of ductal carcinoma ***		z = 0.1	z = 0.05	z = 0.03	z = 0.7
Lobular	A	36.7 ±5.1	122.9 ±34.3	5.7 ±0.9	16.3 ±2.5
carcinoma	M	20	20	4	12
	R	0 – 213	0 – 2047	0 – 24	0 – 103
	n	76	76	48	61
Medullary	A	10.3 ±3.9	71.6 ±64.6	14.3 ±3.1	15.0 ±7.0
carcinoma	M	11	6	14	12
	R	0 – 34	0 – 588	6 – 29	2 – 34
	n	9	9	7	4
Mucinous	A	81.0 ±33.2	37.2 ±25.1	1.8 ±1.0	31.3 ±13.4
carcinoma	M	71	7	1.5	26
	R	0 -202	0 -155	0 – 4	7 -66
	n	6	6	4	4
Mixed	A	71.4 ±30.9	112.5 ±48.9	5.1 ±1.8	11.4 ±2.2
carcinoma	M	33	41	5	9
	R	0 – 380	0 – 537	0 – 15	5 – 24
	n	13	13	8	8

* Concentrations of steroid hormone receptors-fmol /mg c. pr.; growth factor receptors-fmol /mg m.pr.
** A – arithmetic mean; M – median; R – range; n – number of cases
*** differences between concentrations estimated by Wilcoxon test, z – probability

Table II. The concentrations of estrogen (±ERc), progesterone (±PRc), insulin-like growth factor-I and epidermal growth factors receptors in breast ductal carcinoma

Grade of histological malignancy*		Receptors**			
		ERc	PRc	EGF-R	IGF-I-R
	***				
	A	48.0 ±16.6	133±26.6	7.9 ±1.6	20.4 ±5.2
	M	36	60	4	13
I	R	0 – 213	0 – 839	0 – 46	3 – 134
	n	45	45	45	25
	A	31.3 ±4.5	41.1 ±10.4	6.7 ±0.85	15.5 ±1.1
	M	21.5	12.5	5.5	11
II	R	0 – 178	0 – 468	0 – 33	0 – 89
	n	54	54	54	186
	A	24.6 ±11.2	30.5 ±11.6	11.9 ±2.1	14.6 ±2.1
III	M	6.5	8.5	8.5	10
	R	0 – 360	0 – 319	0 – 58	0 – 95
	n	32	32	32	59
		I i II – 0.0336	I i II – 0.0009	I i II – 0.4628	I i II – 0.2
Statistic between groups Probability****		I i III – 0.0584	I i III – 0.0027	I i III – 0.1353	I i III – 0.2
		II i III – 0.5185	II i III – 0.5158	II i III – 0.0079	II i III – 0.6

* according to Bloom and Richardson [13].
** concentrations of steroid hormone receptors – fmol/mg cytosol protein; growth factor receptors – fmol/mg membrane protein
*** A – arithmetic mean; M – median; R – range; n – number of cases
**** differences between groups estimated by Wilcoxon test – probability

tions of all four receptors corresponding to histological grade of malignancies of ductal breast carcinoma. Besides, Table II contains the results of statistical comparisons of these values.

In ductal carcinomas correlations were calculated between the grade of histological malignancy and the concentrations of EGF-R, IGF-I-R, ERc and PRc. Correlations were determined in the entire group of ductal carcinomas of women aged 20-86 years, in the group of younger women (considered as premenopausal) aged 20-50 years and in the group of older women (considered as postmenopausal) aged 51-86 years. The results are presented in Table III. We have divided the tumours into two groups: first one, with cancers from premenopausal women and the second with cancers from postmenopausal women. After that, the material included a small group of cases, so the concentrations of IGF-I-R could not be compared with the grade of histological malignancy of breast ductal carcinoma.

Discussion

In ductal carcinomas *in situ* the concentrations of ERc and PRc were significantly higher than in invasive ductal carcinomas (132.2 and 126.9 fmol/mg cytosol protein, versus 44.2 and 71.9 fmol/mg cytosol protein). On the other hand, EGF-R concentrations in *in situ* carcinomas were only half of those in infiltrating ductal carcinomas (4.1 versus 8.1 fmol/mg m.p.). In analogous situations IGF-I-R concentrations were higher but were not different between themselves (14.4 versus 15.7 fmol/mg membrane protein).

In our studies lobular carcinomas had, in the first place, high PRc concentrations (122.9 fmol/mg c. p.) and fairly high concentrations of ERc (36.7 fmol/mg c. p.). The mean EGF-R concentration in lobular carcinoma was at the detectability level (5.7 fmol/mg m. p.), while IGF-I-R was at average level, similar to that in other types of carcinomas (16.3 fmol/mg m. p.). In the studies of Lasota et al. concentrations of ERc and PRc were high, similarly as in our previous study [14].

The material included only 9 medullar carcinomas. They had, in the first place, very low ERc concentrations (mean 10.3 fmol/mg c. p.) and relatively high PRc concentrations (71.6 fmol/mg c. p.), considerable EGF-R and IGF-I-R concentrations (14.3 fmol/mg m. p. and 15.0 fmol/mg m. p.).

Only 6 mucinous carcinomas were found in our material. They had very low EGF-R concentrations (mean 1.8 fmol/mg) and relatively high concentrations of ERc, PRc and IGF-I-R (respective mean values were: 81.0, 37.2 c. p. and 31.3 fmol/mg m. p.). Lasota et al. al-

Table III. Correlations between the concentrations of estrogen (ERc), progesterone (PRc), epidermal growth factor (EGFR) and insulin-like growth factor receptors-I (IGF-I-R) and the grade of histological malignancy of breast ductal carcinoma

Age of patients	Compared parameters Receptor		Method estimating of malignancy	Number of cases	Correlation coefficient***	p****
	ERc	vs	B*	131	-0.3251	0.0000
	ERc	vs	SBR**	131	-0.3628	0.0000
	PRc	vs	B*	131	-0.3195	0.0000
	PRc	vs	SBR**	131	-0.2450	0.0002
20-86 years	EGF-R	vs	B*	131	0.1764	0.0124
	EGF-R	vs	SBR**	131	0.1919	0.0043
	IGF-I-R	vs	B*	270	-0.0594	0.2280
	ERc	vs	B	46	-0.4092	0.0005
	ERc	vs	SBR**	46	-0.3897	0.0005
20-50 years	PRc	vs	B*	46	-0.2599	0.0291
	PRc	vs	SBR**	46	-0.1603	0.1579
	EGF-R	vs	B*	46	0.2380	0.0502
	EGF-R	vs	SBR**	46	0.3074	0.0079
	ERc	vs	B*	85	-0.2801	0.0010
	ERc	vs	SBR**	85	-0.3623	0.0000
51-86 years	PRc	vs	B*	85	-0.3536	0.0000
	PRc	vs	SBR**	85	-0.2885	0.0005
	EGF-R	vs	B*	85	0.1488	0.0907
	EGF-R	vs	SBR**	85	0.1354	0.1057

* B – the grade of histological malignancy estimated by Bloom and Richardson method [13]
** SBR – the grade of histological malignancy estimated by SBR method [26]
*** estimated by rank Kendall method
**** probability

so found relatively high concentrations of ERc and PRc in five mucinous carcinomas (mean 188.5 and 40.5 fmol/mg c. p.).

Mixed, that is ductal-lobular carcinomas, were present in 13 cases in our material. They had fairly high concentrations of ERc and PRc, and moderate EGF-R and IGF-I-R concentrations. In view of high variability of these tumours it was difficult to determine the relationship between the concentrations of the studied receptors and the degree of their differentiation.

In our study we demonstrated that in ductal carcinomas the concentrations of ERc and PRc were inversely correlated with the grade of malignancy. This was observed in the group of all women as well as postmenopausal women. In the group of carcinomas from women aged 20-50 years (premenopausal), the correlation between PRc concentration and the histological grade of malignancy was statistically significant only when it was assayed by the Bloom-Richardson method ($p = 0.029$). In the analysis by the modified Bloom-Richardson method (SBR) the correlation was inverse, but did not reach significance level ($p=0.15$). This data confirms earlier studies of Lasota et al. on the relationships between ERc and PRc concentrations, on one hand, and the histological grade of malignancy of carcinoma on the other hand [14]. Similarly as in our study, the negative correlation between ERc and PRc and the grade of malignancy was highly significant, but in the selected groups or study methods the significance of this correlation for PRc was doubtful.

A statistically significant positive correlation exists between EGF-R concentrations and the histological grade of malignancy of ductal carcinoma [15]. However, in the group of postmenopausal women this correlation failed to reach significance level ($p= 0.09$ for the B method and 0.105 for the SBR method). High controversies are present in the problem of the relationship between EGF-R concentration and the grade of morphological differentiation of breast carcinoma. Skoog et al. [27] and John A. Foekens et al. [8], Pirinen et al. [16], Torregrosa et al. [28, 29] found no correlation between tumour differentiation and EGF-R concentration, while Sainsbury et al. [17], Gupta et al. [20] and Pekonen et al. [30] found a correlation between EGF-R concentration and the histological grade of malignancy of breast tumours. These divergences of opinions may be caused by the type of cases in which these correlations were studied. This is visible in our study. When correlations were calculated in the entire material of breast cancers in women aged 20-86 years, a positive statistically significant correlation was found between EGF-R level and the grade of morphological differentiation of ductal carcinoma. When, however, the tumours were divided into two groups, one group of cancers from premenopausal women (20-50 years) and the other from postmenopausal women (51-85 years), this correlation remained in the younger group but was not present in the older group.

In our material IGF-I-R concentration in ductal carcinoma showed no correlation with the grade of malignancy, although in numerical data a tendency for inverse correlation was noted – although insufficient for significance (Table II). This is unexplained because IGF-I-R concentrations correlate positively with these of ERc and PRc, which on the other hand, correlated inversely with the grade of malignancy of breast cancer. Similarly, Foekens et al. [8], Pekonen et al. [30] and Peyrat et al. [21] and Kluska et al. [31] failed to find correlations between IGF-I-R concentration and the grade of malignancy of carcinoma. However, Bonneterre et al. [7] demonstrated also a correlation between IGF-I-R and the grade of malignancy.

Conclusions

The data presented here indicates that ductal carcinomas of lower grade of malignancy had higher ERc and PRc concentrations but lower EGF-R concentration. In general with progressing malignancy of the tumour the concentrations of ERc and PRc were decreasing, and the concentration of EGF-R increased. However, the correlation between EGF-R concentration and the grade of malignancy carcinoma was significant in the tumours of younger women only, but not in the older women. IGF-I-R concentration in ductal carcinomas showed a tendency to decrease with the grade of malignancy, but failed to reach statistical significance.

Lobular carcinomas had, in the first place, high PRc and moderately high ERc concentrations. The mean EGF-R concentrations in these tumours were at a detectability threshold, and that of IGF-I-R was at the level found in average types of breast cancer.

Medullary carcinomas had a relatively low ERc concentrations but high concentrations of PRc, EGF-R and IGF-I-R.

Mucinous carcinomas had very low EGF-R concentrations and relatively high concentrations of ERc, PRc and IGF-I-R.

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