CGRP and Breast Cancer

Increased CGRP associated with increased breast density, a breast cancer risk factor

Breast density, 99mTc(V)DMSA uptake, calcitonin gene related peptide (CGRP)*, estrogen receptor and Ki-67 expression in mixed invasive associated with extensive in situ ductal carcinoma (DCIS+IDC) and pure invasive ductal carcinoma (IDC)

Purpose: The aim of our study was to assess the variation of CGRP* expression in patients with IDC and in patients with IDC+DCIS in relation with % mammographic breast density (BD), the uptake of the cell proliferation seeking radiotracer 99mTc-(V)DMSA (SMM uptake), the proliferation index (Ki -67) and the estrogen receptors (ER) status.
Methods: We studied retrospectively 24 women who referred to our department with suspicious findings on palpation and/or mammography and were evaluated preoperatively with 99mTc-(V)DMSA scintimammography. Histology revealed 12 IDC (mean age ± SD = 66.5 ± 13.1) and 12 IDC + DCIS (mean age ± SD = 58.5 ± 15.1). Immunohistochemical staining for CGRP*, Ki-67 and ER status was performed in all 24 surgical specimens. BD and SMM uptake were calculated by computer assisted methods and were correlated with CGRP* expression by linear regression analysis. CGRP* values were compared (Mann Whitney test) between patients with >25% and <25% BD. BD and SMM uptake were compared between CGPR*(-) and CGRP*(+) patients and between IDC and IDC + DCIS. Ki-67 and ER were compared between IDC and IDC + DCIS. Ki-67 was also compared between patients with BD >25% and <25%, whereas ER was correlated with CGRP*.
Results: Overall positive correlation was found between BD and CGRP* expression ($r = 0.577$ $p<0.001$) [rBD(IDC)-CGRP* = 0.765 $p<0.001$, rBD(IDC+DCIS)-CGRP* = 0.746 $p<0.001$]. Positive correlation was found between SMM uptake and CGRP* only in IDC + DCIS [rSMM(IDC+DCIS)-CGRP*= 0.634 $p<0.05$]. CGRP* expression was significantly higher in patients with >25% BD compared to the ones with <25% (23.18 ± 7.5 vs. 9 ± 5.1 ,$p=0.00008$). BD and SMM uptake was significantly higher in CGRP*(+) than in CGRP*(-) patients (30.68 ± 11.24 vs. 18.01 ± 4.5 ,$p=0.001$ and 22.63 ± 10.61 vs. 11.92 ± 4.79, p= 0,012, respectively) as well as in IDC + DCIS as compared to pure IDC (31.38 ± 8.19 vs. 21.67 ± 11.6, p= 0.044 and 27.94 ± 8.04 vs. 13.19 ± 6.84, $p=0.0002$, respectively). Ki -67 was significantly higher in IDC + DCIS than in pure IDC (27.91 ± 13.8 vs. 12.8 ± 9.44) as well as in patients with BD>25% than in patients with BD<25% (26 ± 15.44 vs. 10 ± 1.63). On the contrary, ER was significantly lower in IDC + DCIS compared to pure IDC (71.81 ± 95.45 vs. 162.17 ± 55.49) and strongly inversely correlated with CGRP* in pure IDC ($r=-0.742$,$p<0.01$).
Conclusion: BD, SMM, CGRP* and Ki-67 expression were significantly increased and ER status significantly decreased in DCIS+IDC as compared to IDC, indicating that the two groups are different entities, the DCIS+IC being more aggressive and ER independent and possibly associated with a pathway linked to stromal involvement and CGRP* activity.

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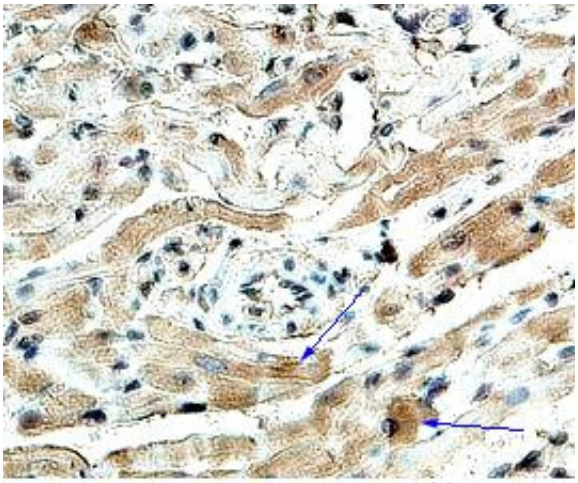
BACKGROUND: We evaluated the variation of calcitonin gene related peptide (CGRP) expression in patients with mixed invasive with extensive in situ ductal carcinomas (IDC + DCIS) and pure IDC, in relation to mammographic breast density (%BD), proliferation-seeking radiotracer (99m)Tc(V) dimercaptosuccinic acid (DMSA) uptake (scintimammographic-SMM), proliferation index Ki-67, and estrogen receptor (ER) status. We also assessed CGRP expression with histological grade. METHODS: We studied retrospectively 24 women with suspicious findings on mammography who were evaluated preoperatively with (99m)Tc(V)DMSA scintimammography. Histology revealed 12 IDC (grade II in 8, grade III in 4 patients; mean size +/- SD, 2.6 +/- 1.3 cm; mean age +/- SD, 66.5 +/- 13.1 years) and 12 IDC + DCIS (grade II in 6, grade III in 6 patients; DCIS component mean size +/- SD, 5.3 +/- 1.8 cm; IDC component mean size +/- SD, 2.5 +/- 1.1 cm; mean age +/- SD, 58.5 +/- 15.1 years). Immunohistochemistry for CGRP, Ki-67, and ER status was performed in all 24 surgical specimens. BD and SMM were calculated by computer-assisted methods and were statistically correlated with CGRP expression. BD, SMM, Ki-67, and ER were statistically compared between IDC and IDC + DCIS, whereas CGRP, Ki-67, and ER were compared between patients with BD >25 and <25%. CGRP was also compared (t test) between grade II and grade III in both groups. RESULTS: Overall positive correlation was found between BD and CGRP ($r = 0.577$, $P < 0.001$). Positive correlation was established between SMM and CGRP only in IDC + DCIS (r (SMM(IDC+DCIS)-CGRP) = 0.634, $P < 0.05$). CGRP and Ki-67 were significantly higher in patients with BD >25% compared with <25% BD patients ($P = 0.00008$ and $P = 0.014$, respectively). BD and SMM were significantly higher in CGRP(+) than in CGRP(-) patients as well as in IDC + DCIS compared with IDC. Ki-67 was significantly higher, whereas ER was significantly lower, in IDC + DCIS than in IDC. In all patients, CGRP was significantly higher in grade II as compared with grade III ($P = 0.005$). In the mixed group (IDC + DCIS), grade II cancers had also significantly higher CGRP values as compared with grade III ones ($P = 0.004$). In pure IDC, no statistical difference was found between grade II and III ($P = 0.4$). CONCLUSIONS: BetaD, SMM, CGRP, and Ki-67 were significantly increased, whereas ER was significantly decreased, in IDC + DCIS as compared with IDC, indicating that IDC + DCIS is an entity that is more aggressive, ER independent, and possibly associated with a pathway linked to stromal involvement and CGRP activity.

Papantoniou et al. Breast Cancer. 2010 Feb 9. [Epub ahead of print]

Mapping in Rat Heart (H-015-09)



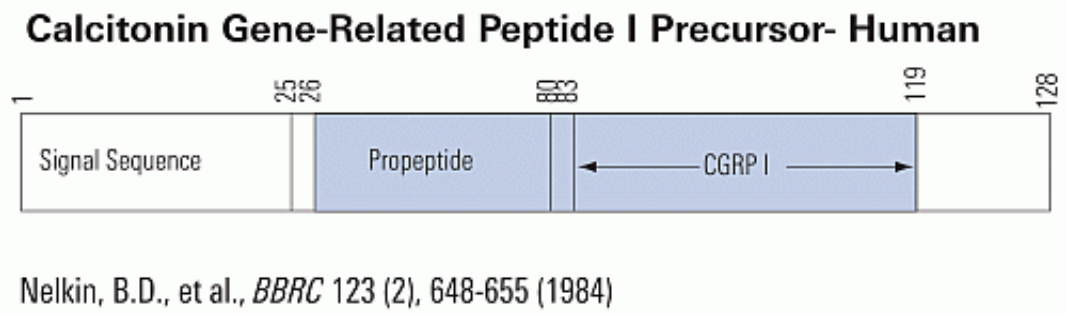
Rat heart tissue was stained by Anti-CGRP (Rat) Antibody (Catalog No.: H-015-09)

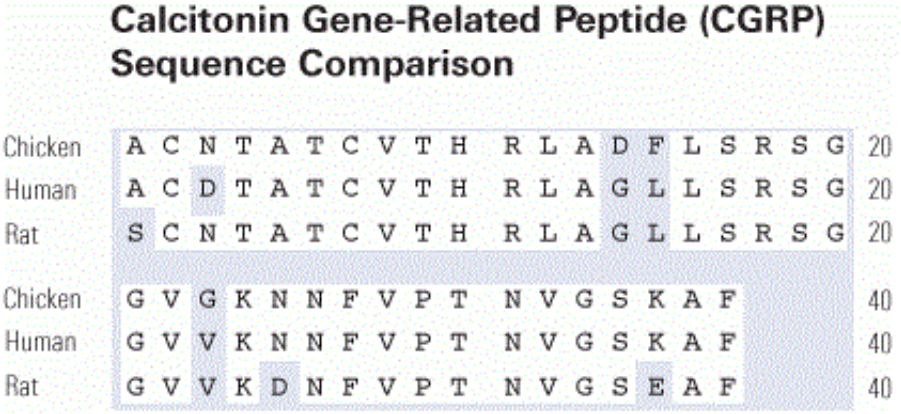
Tissue Sample	Rat Heart
Fixative	10% formalin
Embedding	Paraffin
Negative Control	No primary antibody
Pretreatment	N/A
Blocking	2% Normal Goat Serum
Primary Antibody	Rabbit Anti-CGRP (Rat) Antiserum (Catalog No.:H-015-09)
Optimal Dilution	1:200 (1hour at RT)
Secondary Antibody	Goat Anti-Rabbit IgG, Biotinylated (1:400), 30 min
Amplification	ABC (Vector) (1:400, 30 min)
Detection System	HRP
Substrate	DAB (Sigma), 3 min
Counterstained	Hematoxylin, 30 sec

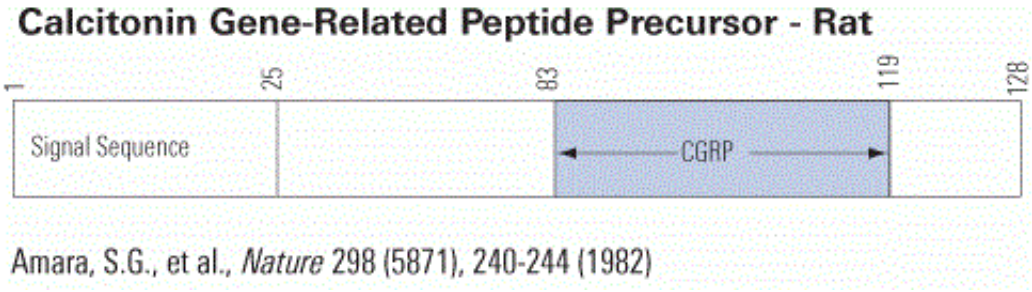
The calcitonin gene peptides: biology and clinical relevance.

The calcitonin/CGRP multigene complex encodes a family of peptides: calcitonin, its C-terminal flanking peptide, katacalcin, and a third novel peptide, calcitonin gene-related peptide (CGRP). The 32-amino acid peptide calcitonin inhibits the osteoclast, thereby conserving skeletal mass during periods of potential calcium lack, such as pregnancy, growth, and lactation. This hormonal role is emphasized by observations that lower circulating calcitonin levels are associated with bone loss and that calcitonin replacement prevents further bone loss. Structurally, CGRP resembles calcitonin and has been implicated in neuromodulation and in the physiological regulation of blood flow. Here we review the molecular genetics, structure, and function of the calcitonin-gene peptides as analyzed in the laboratory and focus on more recent clinical studies relating to disorders and therapeutics.

Zaidi et al. Crit Rev Clin Lab Sci. 1990;28(2):109-74.







CGRP (Human, Rat), CGRP II (Human), Amylin and Adrenomedullin (Human) Sequence Comparison

h-CGRP	A	C	D	T	A	T	C	V	T	H	R	L	A	G	L	L	S	R	S	G	20
h-CGRP II	A	C	N	T	A	T	C	V	T	H	R	L	A	G	L	L	S	R	S	G	20
r-CGRP	S	C	N	T	A	T	C	V	T	H	R	L	A	G	L	L	S	R	S	G	20
h-Amylin	K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	20
h-ADM (15-51)	G	C	R	F	G	T	C	T	V	Q	K	L	A	H	Q	I	Y	Q	F	T	20
											31										
h-CGRP	G	V	V	K	N	N	F	V	P	T	N	V	G	S	K	A	F				40
h-CGRP II	G	M	V	K	S	N	F	V	P	T	N	V	G	S	K	A	F				40
r-CGRP	G	V	V	K	D	N	F	V	P	T	N	V	G	S	E	A	F				40
h-Amylin	N	N	F	G	A	I	L	S	S	T	N	V	G	S	N	T	Y				40
h-ADM (15-51)	D	K	D	K	D	N	V	A	P	R	S	K	I	S	P	Q	G				40

Amylin (Feline, Rat, Human) and CGRP and Adrenomedullin (Rat) Sequence Comparison

r-Amylin	K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	R	S	S	20
f-Amylin	K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	Y	R	S	S	20
h-Amylin	K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	20
r-CGRP	A	C	D	T	A	T	C	V	T	H	R	L	A	G	L	L	S	R	S	G	20
r-ADM (15-51)	G	C	R	F	G	T	C	T	V	Q	K	L	A	H	Q	I	Y	Q	F	T	20
r-Amylin	N	N	L	G	P	V	L	P	P	T	N	V	G	S	N	T	Y				40
f-Amylin	N	N	L	G	A	I	L	S	P	T	N	V	G	S	N	T	Y				40
h-Amylin	N	N	F	G	A	I	L	S	S	T	N	V	G	S	N	T	Y				40
r-CGRP	G	V	V	K	N	N	F	V	P	T	N	V	G	S	K	A	F				40
r-ADM (15-51)	D	K	D	K	D	N	V	A	P	R	N	K	I	S	P	Q	G				40

Catalog No.	Name	Size
T-G-015-09	CGRP (Rat, Mouse) - I-125 Labeled Purified IgG	10 µCi
T-G-015-07	CGRP II (Human) - I-125 Labeled Purified IgG	10 µCi
T-G-015-02	CGRP (Human) - I-125 Labeled Purified IgG	10 µCi
T-015-09	CGRP (Rat, Mouse) - I-125 Labeled	10 µCi
T-015-07	CGRP II (Human) - I-125 Labeled	10 µCi
T-015-04	CGRP [Tyr0] (Human) - I-125 Labeled	10 µCi
RK-015-09	CGRP (Rat, Mouse) - RIA Kit	1 kit
RK-015-07	CGRP II (Human) - RIA Kit	1 kit
RK-015-02	CGRP (Human) - RIA Kit	1 kit
H-015-09	CGRP (Rat, Mouse) - Antibody for Immunohistochemistry	50 µl
H-015-07	CGRP II (Human) - Antibody for Immunohistochemistry	50 µl
H-015-02	CGRP (Human) - Antibody for Immunohistochemistry	50 µl
H-001-37	CGRP Type 1 Receptor (436-461) (Human) - Antibody for Immunohistochemistry	50 µl
G-015-09	CGRP (Rat, Mouse) - Purified IgG Antibody	400 µg

Catalog No.	Name	Size
G-015-07	CGRP II (Human) - Purified IgG Antibody	400 µg
G-015-02	CGRP (Human) - Purified IgG Antibody	400 µg
G-001-37	CGRP Type 1 Receptor (436-461) (Human) - Purified IgG Antibody	200 µg
FR-015-09	CGRP (Rat, Mouse) - Rhodamine Labeled	1 nmol
FR-015-03	CGRP (17-37) (Human) - Rhodamine Labeled	1 nmol
FR-015-02	CGRP (Human) - Rhodamine Labeled	1 nmol
FG-G-001-37A	CGRP Type 1 Receptor (436-461) (Human) - FAM Labeled Purified IgG	100 µl
FG-015-09A	CGRP (Rat, Mouse) - FAM Labeled	1 nmol
FG-015-03A	CGRP (17-37) (Human) - FAM Labeled	1 nmol
FG-015-02A	CGRP (Human) - FAM Labeled	1 nmol
FEK-015-09	CGRP (Rat, Mouse) - Fluorescent EIA Kit	1 kit
FEK-015-07	CGRP II (Human) - Fluorescent EIA Kit	1 kit
FEK-015-02	CGRP (Human) - Fluorescent EIA Kit	1 kit
EK-015-09	CGRP (Rat, Mouse) - EIA Kit	1 kit
EK-015-07	CGRP II (Human) - EIA Kit	1 kit
EK-015-02	CGRP (Human) - EIA Kit	1 kit
B-G-001-37	CGRP Type 1 Receptor (436-461) (Human) - Biotin Labeled Purified IgG	100 µl
B-015-22	CGRP [Di-Biotinyl-(Lys0)] - Biotin Labeled	100 µg
B-015-06	CGRP (8-37) (Human) - Biotin Labeled	20 µg
B-015-02	CGRP (Human) - Biotin Labeled	20 µg
B-015-01	CGRP (Chicken) - Biotin Labeled	20 µg
015-31	CGRP, Pro (1-54) (Human)	100 µg
015-30	CGRP I/II (26-37) (Human)	200 µg
015-29	CGRP (1-25) (Human)	100 µg
015-28	CGRP (1-17) (Human)	100 µg
015-21	CGRP alpha-[Cys(Et)2,7] (Human)	200 µg
015-20	CGRP II, alpha (27-37) [Tyr27, Ala31, Pro34, Phe35] (Human)	200 µg
015-19	CGRP II, beta (Rat)	200 µg
015-18	CGRP (34-37) (Rat, Mouse)	500 µg
015-17	CGRP (33-37) (Rat, Mouse)	500 µg
015-16	CGRP (32-37) (Rat, Mouse)	500 µg
015-15	CGRP (31-37) (Rat, Mouse)	500
015-14	CGRP (30-37) (Rat, Mouse)	500 µg
015-13	CGRP (29-37) (Rat, Mouse)	500 µg
015-10	CGRP (8-37) (Rat, Mouse)	200 µg
015-09	CGRP (Rat, Mouse)	200 µg
015-08	CGRP [Tyr0] (8-37) (Human)	100 µg
015-07	CGRP II (Human)	200 µg
015-06	CGRP (8-37) (Human)	200 µg
015-05	CGRP [Cys(Acm2,7)] (Human)	200 µg
015-04	CGRP [Tyr0] (Human)	200 µg
015-03	CGRP (17-37) (Human)	200 µg
015-02	CGRP (Human)	200 µg
015-01	CGRP (Chicken)	200 µg
014-41	Calcitonin, Pro / CGRP I/II (1-25) (Human)	100 µg
001-37	CGRP Type 1 Receptor (436-461) (Human)	100 µg