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Expression of K^+ channels in normal and cancerous human breast

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Summary. Potassium (K^+) channels contribute to the regulation of cell proliferation and apoptosis and are also involved in tumor generation and malignant growth.

Using immunohistochemical analysis, we investigated the expression of four K^+ channels GIRK1 (G-Protein Inwardly Rectifying Potassium Channel 1), Ca^{2+}-activated K channel ($K_{Ca}1.1$), voltage activated K^+ channels (K_V 1.1 and K_V 1.3) and of the anti-apoptotic protein Bcl2 in normal and cancerous breast tissues and compared their expression with clinicopathological data. GIRK1 was overexpressed in carcinomatous tissues. In contrast, K_V 1.1 and K_V 1.3 were less expressed in cancerous tissue. The expression of Bcl-2 was similar in both tissues. As to the clinicopathological data, a correlation between $K_{Ca}1.1$ channel and estrogen receptor (ER) expression was observed.

GIRK1 was overexpressed in breast carcinoma suggesting its involvement in proliferation and oncogenesis and its possible use as a putative pharmaceutigal target. The correlation between $K_{Ca}1.1$ channel and ER suggests the involvement of this channel in proliferation. The loss of expression of the two channels K_V 1.1 and K_V 1.3 may correspond to their role in apoptosis.

Key words: Breast tissue, Cancer, Human, Immunostaining, Potassium channels

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Introduction

Potassium (K^+) channels are a most diverse class of ion channels in the plasma membrane. To date, no less than 20 distinct K^+ channel currents have been identified in primary tissues (Shieh et al., 2000; Wang, 2004; O'Grady and Lee, 2005). They are also widely distributed in a vast variety of tissues and cells, including both excitable and non-excitable cells, healthy and transformed cells. The diversity and expression are of paramount physiological importance, since different types of K^+ currents play different roles in regulating various cellular functions e.g. determining the membrane potential, the rate of membrane repolarization, cellular osmolarity, cell proliferation and cell death (Wonderlin and Strobl, 1996; Nerbonne and Kass, 2005). Alterations of K^+ channel function and density -channelopathies- can have profound pathophysiological consequences in a variety of diseases including cancer (Conti, 2004; Vincent and Zhang, 2005).

Studies of breast cancer cell lines have shown the involvement of different K^+ channels, particularly hEAG, hIK1, $K_{Ca}1.1$, HERG, and K_V 1.1 in proliferation, apoptosis or oncogenesis (Ouadid-Ahidouch et al., 2000, 2001, 2004a,b; Wang, 2002; Chen et al, 2005). Using a Real Time-Polymerase Chain Reaction (RT-PCR) technique, an over-expression of G-Protein Inwardly Rectifying Potassium Channel 1 (GIRK1) and two-pore domain (TWIK) related acid-sensitive (TASK) channels in breast tumor tissues was revealed (Stringer et al., 2001; Mu et al., 2003). The expression of GIRK1 was correlated with lymph-node metastasis (Stringer et al., 2001) and TASK may contribute to tumorigenesis by promoting cancer cell survival in the poorly oxygenated areas of solid tumors

(Mu et al., 2003). Apart from the study of Abdul et al., (Abdul and Hoosein, 2002, 2006; Abdul et al., 2003) who studied KV1.3 by immunohistochemistry in both breast and prostate tissues and from the study of Hemmerlein et al. (Hemmerlein et al., 2006) who studied Eag1 by immunohistochemistry on different cancers, the studies on K^+ channels on breast tissues were based solely on mRNA expression (Stringer et al, 2001; Mu et al, 2003). K^+ channels are also involved in regulating cell death (Shieh et al., 2000; Wang, 2004; O'Grady and Lee, 2005). Indeed, several studies have reported that K_V channels are involved in the initiation and regulation of the apoptotic volume decrease (Brevnova et al., 2004; Wang, 2004; O'Grady and Lee, 2005) and it has been reported that the anti-apoptotic protein Bcl-2 down-regulates the K_V expression, inducing an inhibition of apoptosis (Wang, 2004).

The aim of this study was to compare the expression of four potassium channels (GIRK1, $K_{Ca}1.1$, $K_V1.1$ and $K_V1.3$) in normal and cancerous human breast tissue by immunohistochemistry. GIRK1 mRNA was already observed in breast tissue with a difference between metastatic and non metastatic carcinoma but the protein has never been studied by immunohistochemistry. In previous studies, we characterized $K_{Ca}1.1$ channels on MCF-7 cell line but protein expression of this channel in breast tissue was never observed. $K_V1.1$ and $K_V1.3$, voltage gated K^+ channels, were selected because of their involvement in apoptosis and maybe in tumorigenesis. Finally, a correlation between the expression of these channels and the clinicopathological data was searched.

Materials and methods

Tissues specimens and patients characteristics

Normal and cancerous breast tissue was obtained from surgical specimens from women having undergone operations at the Amiens University Hospital, France. Tumor tissue was obtained from patients with primary carcinoma of the breast. Normal breast specimens were taken from mastectomy specimens, away from the tumor.

Thirty-three primary invasive ductal breast carcinomas and 31 normal human breast specimens were obtained. Mean age was 59.7 and 58.4 years in cancer and normal breast tissue respectively. In 29 cases, tumor tissue and normal tissue came from the same mastectomy specimens. Tumor grade was determined according to the Scarff, Bloom and Richardson grade. In the 33 human breast cancer specimens, 5 were of Grade I (well differentiated), 17 were of Grade II (moderately differentiated) and 11 were of Grade III (poorly differentiated). In situ carcinoma was never observed in any specimen. At the diagnosis, 14 tumors were associated with lymph-node metastasis, without distant metastasis (clinical observation, bone scintigraphy and liver scan).

Informed consent to use a portion of the tissue for research purposes (form approved by local ethic committee) was obtained from the patients before surgery.

Immunochemistry

Three-micrometers-thick sections of formalin-fixed and paraffin-embedded tissue samples were realized. The immunohistochemical staining was performed on a Ventana XT immunostainer, using antibodies against potassium channels (anti-$K_{ir}3.1$ (GIRK1) (dilution 1/300), anti-$K_{Ca}1.1$ (dilution1/300), anti- $K_V1.1$ (Kcna1) (dilution 1/50) and anti- $K_V1.3$ (Kcna3) (dilution 1/50), rabbit polyclonal, Alomone Labs, Ltd, Jerusalem, Israel). Other antibodies were used: anti-Bcl2 (DAKO clone 124, dilution 1/40, Dako, Trappes, France), anti-Ki67 (proliferation index) (DAKO, clone KiS5, dilution 1/20, Dako, Trappes, France), anti-estrogen receptor (ER) (DAKO, clone 6F11, dilution 1/20, Dako, Trappes, France) and anti-ErbB2 (Herceptin receptor) (DAKO, A485, dilution 1/1500, Dako, Trappes, France). Then the avidin-biotin-peroxydase complex technique was used. Reactions were developed using a chromogenic reaction in DAB (diamino-3,3'benzidine tetrachlorhydrate) substrate solution (DAB, Sigma Fast). The counterstain was performed with hematoxylin solution. All channels antibodies were certified for immunohistochemistry by Alomone Labs. Negative control was realized doing the same technique without primary antibody.

Immunostaining levels were determined by subjective visual scoring of the brown stain. Two operators independently evaluated antigen expression. For K^+ channels and Bcl2 staining, four intensities were determined (-, +, ++, +++) depending of the intensity and the distribution of the staining (no staining (-); <30% with low intensity (+); 30-60% with low intensity (++); >60% with low intensity or >30% with high intensity (+++); high intensity <30% was never observed). For the quantitative analysis, two groups were combined, high/medium (+++/++) and low/no stain (+/-). The histograms report the percentage of cases with high/medium stain obtained in each group.

In the table 2, Ki67, ER and ErbB2 are expressed depending of the percentage of positive nuclear staining (Ki67 and ER) or membranous staining (ErbB2). For the quantitative analysis, expression was considering positive when ≥ 50% of epithelial cells presented a strong staining.

Statistics

Immunostaining in the epithelial compartment was scored as described above. Fisher tests (Two tailed) were used in GraphPad software to estimate the correlation between K^+ channel expression in the normal and carcinoma tissues as well as to estimate the correlation between their expression and clinicopathological data. A correlation was considered significant when $p<0.05$.

Results

K^+ channel expression and clinicopathological data of the specimens in normal and malignant breast tissue

Immunohistochemistry was performed on each specimen with the different antibodies as described in Material and methods section. For the K^+ channels expression, we observed a staining of the epithelial compartment in most of the cases of normal and cancer breast tissues. The distribution of the staining was often heterogeneous. Fibroblasts of the stromal compartment were stained with GIRK1 and Bcl2 antibodies (Fig. 1). In the epithelial compartment, localization of the staining was cytoplasmic for $K_{Ca}1.1$, $K_V1.1$, $K_V1.3$ and Bcl2 antibodies but was particular for GIRK1 with a cytoplasmic apical localization in cancerous and normal breast tissue.

Results are reported on tables 1, 2 and 3. GIRK1 expression, in particular cytoplasmic apical localization, was increased in cancer tissue as compared to normal tissue (Fig. 1), while that of $K_{Ca}1.1$ was decreased in cancerous tissue (Fig. 1). $K_V1.1$ and $K_V1.3$ channels expression was decreased in carcinoma tissues (Fig. 1). The quantitative analysis of the results obtained is reported in Figure 2. GIRK1 K^+ channels were expressed in 87.9% of the breast carcinomas compared with only 45.1% in normal breast tissue (Fig. 2). $K_{Ca}1.1$ channel was highly expressed in 93.5% of normal tissue

Table 1. Normal breast specimens.

N°	Age	GIRK1	$K_{Ca}1.1$	$K_V1.1$	$K_V1.3$	Bcl2
1	49	+	++	+	-	+++
2	56	+	+++	+	-	++
3	84	++	+++	++	-	+
4	44	++	+++	++	+	+
5	76	++	+++	++	+	+
6	59	++	+++	++	+	++
7	50	++	++	++	+	+
8	60	++	+++	+++	+	+
9	60	+	++	++	-	++
10	64	+	+++	+	+	-
11	29	+	+++	+	-	+++
12	68	+	+	++	++	+
13	41	+++	++	+++	+	+++
14	59	+	++	+	++	+++
15	66	+	+++	+++	+	+
16	40	+++	+++	+++	+	+++
17	63	+++	+++	+++	+++	+++
18	81	+	++	++	+	+++
19	67	+++	+++	++	+	++
20	52	++	+++	+	+	+++
21	62	++	++	+	+	+
22	71	+	+++	+++	++	+
23	60	+	+++	++	-	++
24	75	+++	+++	+	+	-
25	46	++	+++	+++	+++	+++
26	63	+	+++	+++	+++	+++
27	55	+	+++	++	++	+++
28	41	+	+++	+++	++	++
29	43	+	-	++	+++	++
30	50	+	++	++	++	++
31	78	+	+++	++	++	+++

N°: Number; (-): no staining; (+): < 30% with low intensity; (++): 30-60% with low intensity; (+++): > 60% with low intensity or >30% with high intensity (high intensity < 30% was never observed).

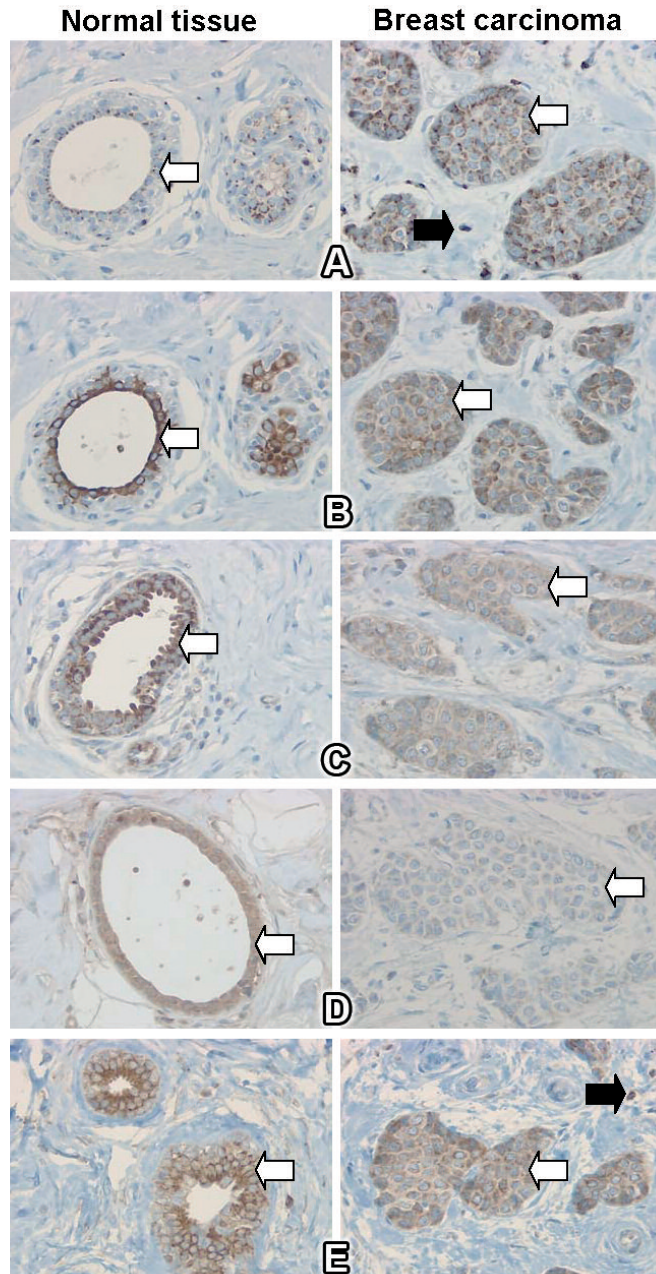

Fig. 1. Immunohistochemical detection of GIRK1, $K_{Ca}1.1$, K_V 1.1, K_V 1.3 K^+ channels and Bcl2 proteins in normal tissue and breast carcinomas. **A**) GIRK1, **B**) $K_{Ca}1.1$, **C**) K_V 1.1, **D**) K_V 1.3, **E**) Bcl2 (magnification x 200). Staining of the epithelial compartment (white arrow) and of the stromal compartment (black arrow). x 200

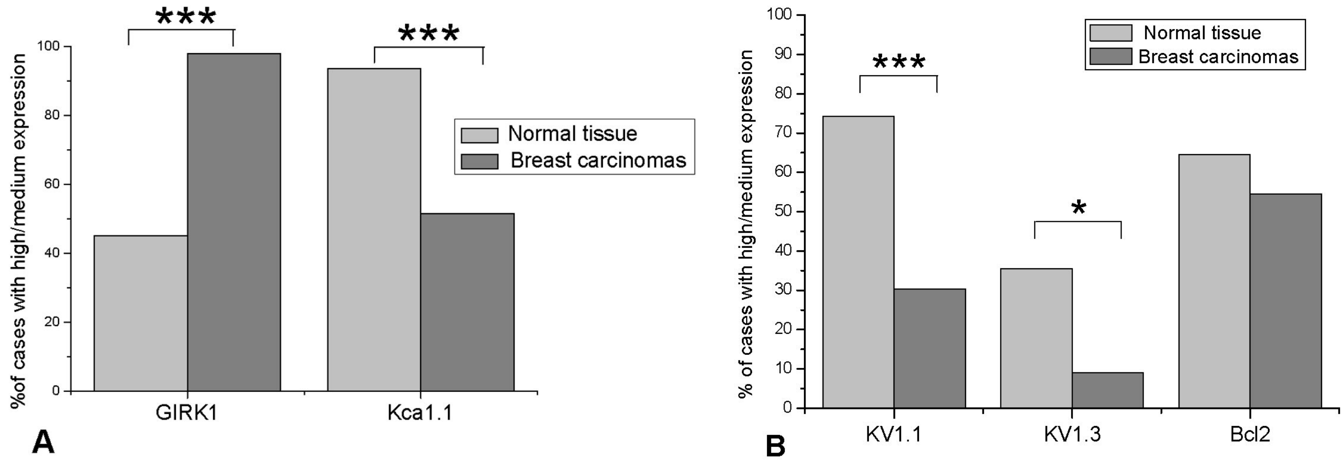

Fig. 2. Variation of the K^+ channels expression in normal and cancer breast tissues. Immunohistochemical expression of K^+ channels related to the specimen collected. 31 samples of normal breast tissue and 33 samples of breast carcinoma tissue were used. **A**) Expression of GIRK 1 and $K_{Ca}1.1$ in normal and cancerous breast tissues. **B**) $K_V1.1$, $K_V1.3$ and Bcl2 expression in normal and cancerous breast tissues.*: $p<0.05$, **: $p<0.005$, ***: $p<0.0005$

Table 2. Cancer specimens.

N°	Age	Size	SBR	ER	ErbB2	Ki67	M +	GIRK1	$K_{Ca}1.1$	K_V 1.1	K_V 1.3	Bcl2
1	59	6	1	+	-	-	-	+++	++	++	+	+++
2	66	25	1	-	-	-	-	+++	+	+	-	+++
3	55	32	1	+	-	-	-	++	+	-	-	-
4	41	50	1	+	-	-	-	++	+	+	-	++
5	43	45	1	-	-	-	+	+	+	-	-	-
6	84	12	2	+	-	-	-	++	+++	+	+	+++
7	56	20	2	+	-	-	+	+++	++	-	-	+++
8	84	35	2	+	-	-	+	+++	++	-	-	++
9	63	15	2	+	-	-	+	+++	+++	+	-	+++
10	50	10	2	+	-	-	+	+++	++	+	+	+++
11	60	12	2	+	-	-	-	+++	+	+	-	+++
12	56	8	2	+	-	-	-	+++	++	+	-	+++
13	64	20	2	-	+	-	+	++	-	+	-	-
14	68	30	2	+	-	+	-	+++	+++	++	+	++
15	63	40	2	-	+	-	-	++	+	-	-	-
16	52	18	2	-	+	-	-	+++	+	-	-	-
17	81	23	2	-	+	-	+	++	+++	+++	++	-
18	40	15	2	+	-	+	-	+++	++	++	++	++
19	62	20	2	-	-	-	-	+++	+	+	-	-
20	71	20	2	+	-	-	-	+++	+++	+++	++	+++
21	84	40	2	-	-	-	+	+++	++	+	-	+++
22	78	35	2	+	-	-	-	+++	+++	++	+	+++
23	44	40	3	+	-	-	+	++	+++	-	-	+
24	76	42	3	-	+	-	+	+++	+	-	-	-
25	43	14	3	-	+	-	+	++	-	-	-	++
26	60	45	3	+	-	-	+	+	+	+	-	+
27	29	27	3	-	+	-	-	+++	-	-	-	-
28	41	20	3	-	-	-	-	++	+++	++	-	+++
29	59	40	3	-	-	+	+	+++	++	++	+	+
30	67	40	3	-	+	-	-	+++	+	-	-	-
31	60	3	3	-	-	-	-	+	+++	++	-	+++
32	63	32	3	+	-	-	+	+	+	+	-	-
33	50	30	3	+	-	-	-	+++	+	++	-	+

N°: Number; (-): no staining; (+): <30% with low intensity; (++): 30-60% with low intensity; (+++): >60% with low intensity or >30% with high intensity (high intensity <30% was never observed); Size in millimeter; SBR: Scarff Bloom and Richardson grade; ER: Estrogen receptor expression; For ER, Ki67 and ErbB2, (+) ≥ 50% positive cells (-) <50% positive cells. M: Metastasis, (+) presence of metastasis, (-) absence of metastasis.

Table 3. Summary of the statistical results obtained for the comparison between K^+ channels, Bcl2 protein expression, ER, Ki67, ErbB2 expression, metastatic status and tumor grade.

	Cancer(C)/Normal tissue(N)	Bcl2	ER	ErbB2	Ki67	Metastasis status	Tumor grade
GIRK1	C>N ***	ns	ns	ns	ns	ns	G2>G3*
$K_{Ca}1.1$	C<N***	ns	*	ns	ns	ns	ns
$K_V1.1$	C<N***	ns	ns	ns	ns	ns	ns
$K_V1.3$	C<N*	ns	ns	ns	ns	ns	ns
Bcl2	ns	/	ns	ns	ns	ns	G2>G3*

Significant statistical results are observed if we compare GIRK1, $K_{Ca}1.1$, $K_V1.1$ and $K_V1.3$ expression between normal and cancer tissue with a higher expression of GIRK1 in cancer tissue and a decrease of the expression of $K_{Ca}1.1$, $K_V1.1$ and $K_V1.3$ in normal tissue. There is a significant correlation between ER (estrogen receptor) and $K_{Ca}1.1$ and a higher expression of GIRK1 and of $K_{Ca}1.1$ in grade II tumor compare with grade III tumor groups. *: $p<0.05$, **: $p<0.005$, ***: $p<0.0005$. ns: no significance.

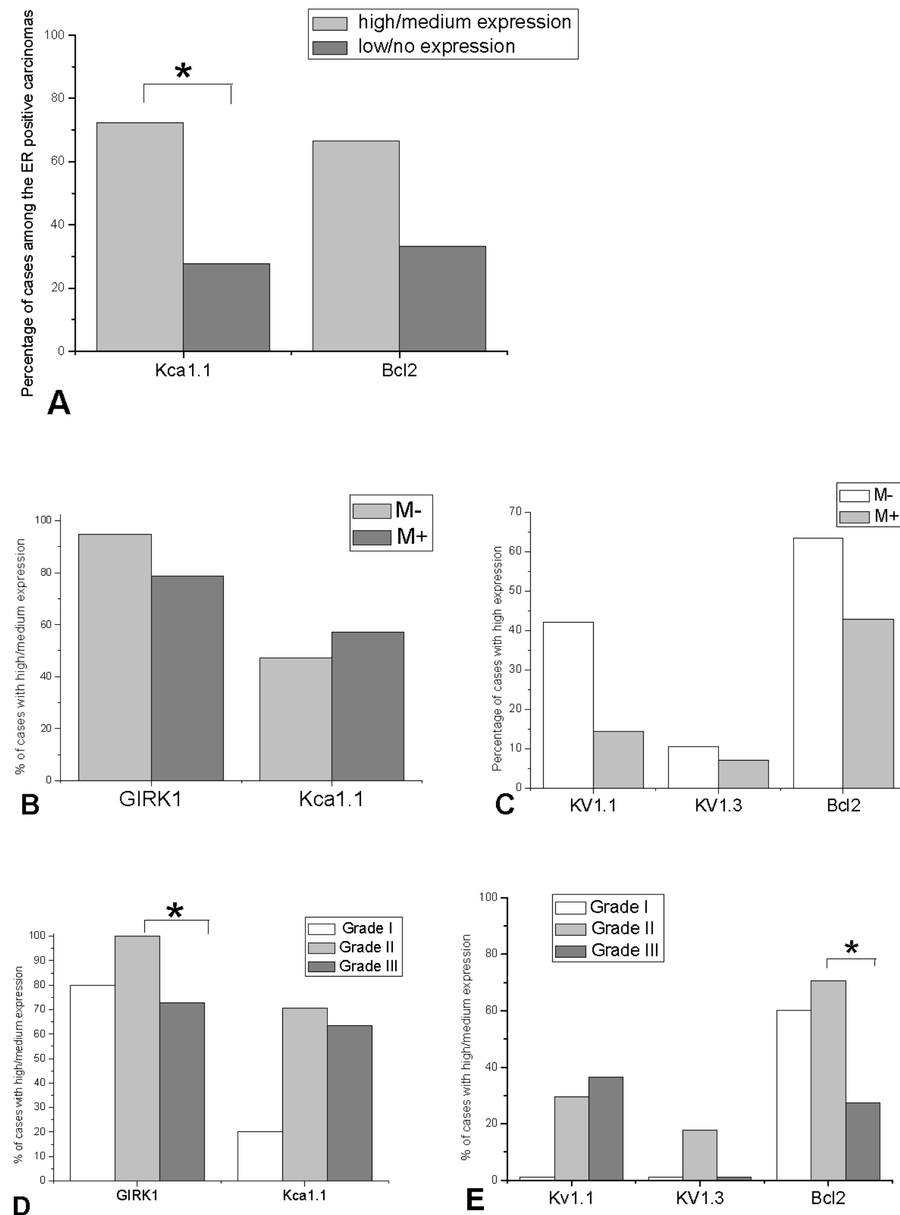

Fig. 3. K^+ channel expression, metastatic status and tumor grades in the 33 invasive ductal carcinomas. **A)** $K_{Ca}1.1$ and Bcl2 expression depending on ER (estrogen receptor) expression. **B-C**) Variation of GIRK1, $K_{Ca}1.1$ **(B)**, K_V 1.1, K_V 1.3 and Bcl2 **(C)** expression in lymph node metastasis (M+) or non metastasis (M-) tumors. **D-E)** Expression of GIRK 1, $K_{Ca}1.1$ **(D)**, K_V 1.1, K_V 1.3 and Bcl2 **(E)** depending of tumor grades. *: p< 0.05.

samples (Fig. 2) and decreased in breast carcinomas (51.5%, Fig. 2). Regarding the K_V channels, the expression of K_V1.1 was lower in breast carcinomas with only 30.3%, comparatively with normal tissue where high staining was observed in 74.2% of the 31 samples (Fig. 2). K_V1.3 was highly expressed in 35.5% of normal tissue, decreased in cancer (9%). Furthermore, no statistical difference of Bcl2 expression between normal and cancer tissue was observed (64.5 and 54.5% respectively, Fig. 2) and no correlation was observed with K_V1.1 or K_V1.3 expression (Table 3).

We next compared the expression of K^+ channels with estrogen receptor expression, ErbB2 and Ki67 expression, lymph node metastatic status and tumor grade. ER were highly expressed in 54.5% (18 samples). Among these 18 ER+ samples, 13 samples (72.2%) showed a high/medium expression for K_{Ca}1.1 (correlation statistically significant) (Fig. 3A, Table 3). There is no statistical significance between ER and Bcl2 expression even though 12 of the 18 ER+ samples (66.6%) showed a high/medium expression for Bcl2 (Fig. 3A, Table 3). Because ErbB2 is overexpressed in about 30% of human breast cancer patients (Yarden and Sliwkowski, 2001) and Ki67, an index of proliferation, is considered as prognostic factor (Ahlin et al., 2007), we investigated whether there was a correlation between these two factors and the K^+ channels expression. But any correlation was observed between ErbB2 and K^+ channels expression (Table 3) and even though GIRK1 had a high expression for the 3 samples where Ki67 was $\geq$ 50%, this is not statistically significant (Table 3). Furthermore, we found that the expression of GIRK1, K_{Ca}1.1, K_V1.1, and K_V1.3 were not correlated with lymph node positive tumours (Fig. 3B and C, Table 3).

Finally, we compared the expression of K^+ channels and Bcl2 with grade tumour. Of 33 invasive ductal breast carcinomas, 5 were Grade I, 17 were Grade II and 11 were Grade III. Statistical significant results were obtained for GIRK1 with a lower expression in grade III compared with grade II and for Bcl2 with a lower expression in grade III compared with grade II (Fig. 3E, F and Table 3). In this comparison, any statistical result was observed for the K_{Ca}1.1 and the K_V channels (Table 3).

Discussion

In cell line models including breast cancer cell, it was demonstrated that K^+ channels have a role in proliferation and/or apoptosis and may have an oncogenic potential (Pardo et al, 1999; Ouadid-Ahidouch et al., 2001, 2004a,b; Wang, 2004). The studies were mainly carried out on breast cancer cells lines or on primary breast cancer culture (Ouadid-Ahidouch et al., 2000, 2001, 2004a,b; Wang, 2002; Chen et al., 2005) and K^+ channel functionality or their mRNA levels were studied. Only a few papers report the localization of these channels by immunochemistry in normal and cancerous breast tissues.

In the present study, the epithelial expression of four K^+ channel types, GIRK1, K_{Ca}1.1, K_V1.1 and K_V1.3 in human breast tissue was demonstrated by immunohistochemistry. These four types of K^+ channels were observed in the epithelial compartment. The overexpression of GIRK1 in cancerous tissue as compared with the normal tissue supports the hypothesis that these channels may have an important role in proliferation and oncogenesis. Moreover, these channels have a particular cytoplasmic apical stain when compared with other channels which have diffuse cytoplasmic staining. This staining may also be explained by the cytoplasmic docking of the protein before its translocation to the membrane. Expression of the GIRK1 channel by the fibroblasts of the stromal compartment will be a problem for molecular studies on minced tissue. Other channels have a diffuse cytoplasmic staining which could be explained by an important production of the proteins.

K_{Ca}1.1 expression falls dramatically in cancerous tissue comparing with the normal tissue with a statistically significant difference (Fig. 2, Table 3). K_{Ca}1.1 channels play a controversial role in cell proliferation. Whereas we observed that K_{Ca}1.1 channels probably don't affect MCF-7 cell proliferation because of their minor role in MCF-7 cell membrane hyperpolarization (Ouadid-Ahidouch et al., 2004a), several studies suggest a correlation between K_{Ca}1.1 channels expression and proliferation (Bringmann et al., 2000). Furthermore, we have reported that K_{Ca}1.1 channels are involved in the proliferative effect of 17ß, estradiol (Coiret et al., 2005) and in this study, we observe that 72.2% of the ER+ specimens have a high or medium expression for K_{Ca}1.1 with a statistical significance. All these data suggest that K_{Ca}1.1 channels are more active in cell proliferation than in oncogenesis. Because of the low number of samples showing a high expression of Ki67, GIRK1 and K_{Ca}1.1 channels cannot be correlated to this proliferation index.

Decreases in intracellular K^+ concentration appear to promote critical events during the early phases of cell death including proteolytic cleavage of pro-caspase-3 and enhanced endonuclear activity (Yu, 2003; Wang, 2004). The specific K^+ channels involved in regulating apoptosis are diverse and include certain K_V channel subtypes like K_V 1.1 and K_V 1.3 (Yu and Choi, 2000; Wang, 2004). Our data show a decrease in the expression of both K_V 1.1 and K_V 1.3 in breast carcinomas in comparison with normal tissue. Indeed Bock et al. (Bock et al., 2002) have demonstrated that K_V 1.3-deficient CTLL-2 T lymphocytes are resistant to apoptosis initiated by the cytostatic drug actinomycin D and retransfection of K_V 1.3 restores sensitivity of the cells to actinomycin D, indicating a central role of K_V 1.3 in actinomycin D-triggered apoptosis. Concerning the K_V1.1 channel, previous studies showed that it was expressed, functional and involved in MCF-7 cell proliferation (Ouadid-Ahidouch et al., 2000) and its inhibition by a-dendrotoxin reduced cell proliferation.

Surprisingly our results don't confirm this data; on other part, $K_V1.3$ was never described in MCF-7 cell line whereas this channel is expressed both in normal and cancerous breast tissue both in our study and in Abdul et al. study (Abdul et al 2003). Probably the heterogeneity of cells in a human tissue and the influence of fibroblasts and inflammatory cells leads to different channel expression compare with monoclonal cell line. Moreover, the expression of K_V channels could be cell cycle dependent. Indeed, it was known that the expression of K^+ channels varied with the cell cycle in breast cancer cells (Ouadid-Ahidouch et al., 2004b).

Our results are in agreement with those of Abdul and Hoosein (Abdul and Hoosein, 2002, 2006) who reported a high expression of $K_V1.3$ channels in the normal prostate tissue and a decrease in 50% of the prostate carcinomas. However, our data are in opposition to those reported by the same authors on breast tissue (Abdul et al., 2003), where they observed an overexpression of $K_V1.3$ in carcinomas. Moreover, it has been reported that potassium current–density is inversely correlated with the metastatic capacity of human prostate cancer cells, being significantly larger in weakly metastatic LNCaP than in strongly metastasis PC-3 cells (Laniado et al., 2001). However, the molecular nature of these channels was not determined for these cell lines. Finally, Fraser et al. showed that $K_V1.3$ expression was reduced in Mat-LyLu in comparison to AT-2 rat prostate cells (Fraser et al., 2003).

Furthermore, interaction between Bcl2 protein and K^+ channels has been studied in vascular smooth muscle cells (Ekhterae et al., 2001). The authors observed that the overexpression of the antiapoptotic protein Bcl-2 decreases K_V channel activity by accelerating the current inactivation and down-regulation of the mRNA expression of the pore forming K_V channels in pulmonary artery smooth muscle cells. Our results did not show significant differences in the expression of Bcl2 between normal tissue and carcinoma suggesting that the down-regulation of both $K_V1.1$ and $K_V1.3$ might not be due to the overexpression of Bcl2.

Stringer et al. (2001) found an over-expression of GIRK1 in breast tumors with metastatic lymph nodes. Unfortunately, we failed to obtain statistical significance probably due to the low number of samples. Regarding the grade of the tumor, Bcl2 was less expressed in grade III tumors suggesting a higher apoptosis phenomenon in this tumor grade but this result is not correlated with the K_V channels expression.

To conclude, these results indicate that GIRK1 K^+ channels are intensively expressed in cancerous breast tissue significantly more than in normal breast tissue and with an apical cytoplasmic localization; these data confirm the strong implication of this channel in tumorigenesis. This channel might be used as a putative pharmaceutical target. Our data suggest a correlation between $K_{Ca}1.1$ channels and ER expression and an involvement of this channel in proliferation more than in oncogenesis. Finally, the cessation of $K_V1.1$ and $K_V1.3$ expression in cancerous tissue may correspond to their role in apoptosis and may be useful as a biomarker for the diagnosis of carcinogenesis.

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