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Letter to the Editor NOT referring to a recent journal article

Lack of *KRAS* and *BRAF* Mutation in Renal Cell Carcinoma

The epidermal growth factor receptor (EGFR) is a tyrosine kinase receptor belonging to the ErbB family, which is abnormally activated in many epithelial tumors. In renal cell carcinoma (RCC), the EGFR is strongly overexpressed in most primary tumors (83%) as well as in a large proportion of metastases (73%) [1] and has been shown to be associated with the development and progression of metastatic disease [2]. As almost half of RCC patients experience distant metastasis during the course of their disease and therapies for advanced/metastatic RCC still remain unsatisfactory, the investigation of potential therapeutic targets such as the EGFR is of utmost importance. In multicenter trials evaluating the antitumor activity of the anti-EGFR antibody panitumumab, an objective response rate of approximately 10% of RCC patients was achieved [2]. As it has recently been shown that activating mutations in the *KRAS/BRAF* genes are associated with poor response to anti-EGFR therapies in colorectal carcinomas, it was suggested that *KRAS/BRAF* mutation analysis may be a promising strategy in the selection of RCC patients for EGFR-targeted therapy [3].

We investigated 121 RCCs of low-grade (pT1/2, no metastases; $n = 50$) and advanced/metastatic (pT3, blood vessel infiltration V2, nodal or hematogenic metastases; $n = 71$) subtypes (Table 1) for *KRAS/BRAF* mutation status. Specifically, two independent samples from each tumor manifestation were enriched to a tumor tissue content of >90% by microdissection and subsequently analyzed using recently described polymerase chain reaction (PCR) DNA sequencing protocols and allele-specific PCRs detecting the *KRAS* mutations exon 2, Glycin12, and Glycin 13 as well as the *BRAF* mutation exon 15, V600E [4]. As shown in Table 1, all conventional RCCs presented unmutated *wild-type KRAS* and *BRAF* sequences, whereas in two different cases of urothelial carcinomas of the renal pelvis, the *KRAS* mutation G12D and the *BRAF* mutation V600E, respectively, were detectable (Fig. 1).

To the best of our knowledge, the data presented in this paper represent the first study of *KRAS/BRAF* mutation status in a large and well-documented cohort of primary and metastatic RCCs.

The finding that *KRAS/BRAF* are unmutated in all conventional RCCs investigated implies that, in contrast to metastatic colorectal carcinoma, the known activating *KRAS/BRAF* mutations are not responsible for the resistance of RCCs to anti-EGFR targeted therapies. None of the established parameters responsible for resistance to anti-EGFR therapies, such as lack of EGFR overexpression,

Table 1 – Histologic diagnosis, tumor stadium, and *KRAS/BRAF* mutation status of all analyzed RCCs ($n = 121$)

Histologic diagnosis	Tumor stadium	KRAS G12/G13	BRAF V600E
Clear cell carcinoma ($n = 63$)	pT1/2, N0, M0, VO ($n = 25$)	0/25	0/25
	pT3, V2orN1/2orM1 ($n = 38$)	0/38	0/38
Papillary carcinoma ($n = 22$)	pT1/2, N0, M0, VO ($n = 9$)	0/9	0/9
	pT3, V2orN1/2orM1 ($n = 13$)	0/13	0/13
Chromophobe carcinoma ($n = 14$)	pT1/2, N0, M0, VO ($n = 6$)	0/6	0/6
	pT3, V2 or N1/2 or M1 ($n = 8$)	0/8	0/8
Urothelial carcinoma ($n = 22$)	pT1/2, N0, M0, VO ($n = 10$)	0/10	1/10
	pT3, V2 or N1/2 or M1 ($n = 12$)	1/12	0/12

pT1/2 = tumor <7 cm or >7 cm in greatest dimension, limited to the kidney; N0 = no regional lymph node metastasis; M0 = no distant metastasis; pT3 = tumor extends into major veins or directly invades adrenal gland or perinephric tissues but not beyond Gerota's fascia; V2 = gross vein infiltration; N1/2 = metastasis in a single/more than one regional lymph node; M1 = distant metastasis; *KRAS* G12/G13 = KRAS mutation exon 2 Glycin 12 and Glycin 13; *BRAF* V600E = BRAF mutation exon 15 Valin 600 glutamine acid.

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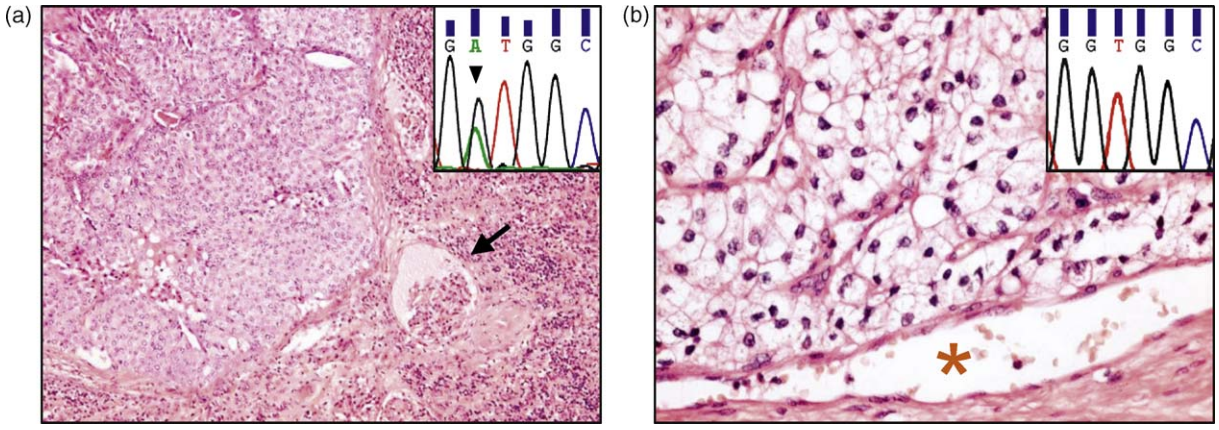

Fig. 1 – Morphologic changes and results of *KRAS* mutation analyses in a case of a urothelial carcinoma of the renal pelvis (a; arrow indicates a glomerulum of the renal tissue adjacent to the infiltrating carcinoma) with detection of the *KRAS* mutation G12D (inset, arrowhead), whereas in a classic clear cell carcinoma (b; asterisk indicates blood vessel infiltration), representative of all conventional renal cell carcinomas investigated, no *KRAS* mutation was found (inset). Images were produced with a BX50 microscope (Olympus, Hamburg, Germany) and a DP50 digital camera with DP-Soft 5.0 software (Olympus, Hamburg, Germany).

presence of activating *KRAS/BRAF* mutations (as we demonstrated), and loss of the tumor suppressor gene *PTEN*, appears manifest in RCCs. Consequently, *KRAS/BRAF* mutations beyond the known gene loci on exons 2 and 15 may have to be investigated ultimately to clarify the role of these mutations in the resistance of RCCs to anti-EGFR therapies. Nevertheless, it was shown that knockdown of Akt-1 and MEK-1 had to be overcome for continued tumor cell growth in RCC, while continuing EGFR signaling cascade suppression appeared not to inhibit cell proliferation [5]. Thus, genetic analyses of alternative members of the EGFR signaling cascade, such as Akt-1 and MEK-1, may also contribute to elucidating the reasons for the almost-complete resistance of RCCs to anti-EGFR targeted therapies.

Conflicts of interest: The authors have nothing to disclose.

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