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Questionable usefulness of Braf-V600E mutation to distinguish between sporadic and hereditary colorectal tumors with microsatellite instability in young patients

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Braf-V600E mutations occur in most sporadic tumors with microsatellite instability (MSI) but have never been observed in HNPCC tumors with MSI. Based on these findings, it has been proposed to use the Braf mutation as screening method for non-Amsterdam MSI positive cases for simplifying HNPCC genetic testing. The aim of this study was to assess the value of this mutation for the clinical practice of suspected HNPCC families in the Comunidad Valenciana (Spain).

A total of 16 colorectal tumors with MSI from probands with suspected Lynch syndrome (non-Amsterdam II, Bethesda guidelines families) were tested for Braf-V600E mutation. Another eight tumors from probands fulfilling Amsterdam II criteria were also tested for Braf mutations. Germline pathogenic mutations at MLH1, MSH2 and MSH6 genes were analyzed in all these individuals. Besides, 119 colorectal tumors from sporadic origin were screened for the Braf mutation. MSI status was assessed using the five Bethesda microsatellite markers by PCR and capillary electrophoresis. Mutation analysis was performed by PCR and direct sequencing.

Four of the probands from non-Amsterdam families, and two from Amsterdam II families, showed a pathogenic germline mutation in one of the repair genes analyzed (25%). As expected, none of these patients had Braf mutation. We did not find any Braf mutated case among the 12 probands MSI positive from non-Amsterdam, Bethesda guidelines families; who were negative for mutations in the repair genes. Median of age of these patients was 41 years. We found eigth Braf-V600E mutant cases in the series of sporadic colorectal tumors analyzed (6.7%). The association between Braf mutation and MSI was statistically significant ($p < 0.001$).

Recently it has been reported that Braf mutation frequency of colorectal tumors with MSI increase with patient age (frequency of MSI tumors with mutation aged <55 years was 7% and 61% in those aged over 55 years). The results presented here, shows that the Braf mutation did not detect any sporadic case among the 11 tumors from patients fulfilling Bethesda criteria with MSI and no pathogenic mutations in the repair genes.

The detection of Braf-V600E mutation may represent an exclusion criterion for Lynch syndrome with a high specificity; but the sensibility of this marker is very low, mainly for patients aged <55 years. These patients represent the most frequent uncertain situation to be classified as sporadic or familial tumors in clinical practice.

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Relationships between HER2 and the leptin (Ob/ObR) system in breast cancer

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Background: The obesity hormone leptin (Ob) has been implicated in tumorigenesis, especially in the development of breast cancer (BC). The mitogenic, angiogenic, and antiapoptotic activities of Ob are mediated through the leptin receptor (ObR). Data obtained in HEK293T cells engineered to coexpress ObR and the oncoreceptor HER2 suggested that Ob can transactivate HER2 via ObR. To address this putative interaction, we studied whether simultaneous expression of Ob, ObR and HER2 can occur in human BC.

Material and Methods: The expression of Ob and ObR was evaluated by immunohistochemistry in 59 BCs (31 HER2-positive, 28 HER2-negative). Ob and ObR were classified as positive (at least +) or negative (below +). The relationships among Ob and ObR and the clinicopathological features, i.e., grading (G1, G2, G3), tumor size (diameter in mm), node involvement (positive or negative), vascular invasion (positive or negative), ER and PgR expression (positive or negative) were analyzed using the χ^2 test.

Results: Ob and ObR were coexpressed in 78% of BC and were correlated in all BCs ($p < 0.001$), in HER2-positive ($p \leqslant 0.001$) and HER2-negative ($P = 0.045$) subgroups. In all BCs, the expression of Ob and/or ObR was associated with tumor size (Ob: $p = 0.04$; ObR: $p = 0.05$) and node positivity (ObR: $p = 0.04$; Ob/ObR: $p = 0.02$). Furthermore, the expression of Ob occurred more frequently in large (>10 mm), node-positive tumors (trends $p = 0.06$ and $p = 0.08$). The simultaneous expression of Ob/ObR and HER2 was found in 39% of BCs, but the Ob/ObR system was also frequent in HER2-negative BCs. Ob, ObR and combined Ob/ObR did not correlate with HER2, grading, VI, and ER/PgR.

Conclusions: Ob and ObR are often coexpressed and a subset of Ob/ObR-positive tumors exhibits concomitant expression of HER2. Thus, interactions between Ob/ObR and HER2 might occur in breast cancer, where activation of Ob/ObR could impede anti-HER2 treatments.

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No genomic changes in mammographically dense breast

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Background: Increased mammographic density gives a 4−6 times higher breast cancer risk and is the strongest risk factor after BRCA-mutations and age (Boyd et al. 886−94;Couzin 1664−66). Little is known of the biological basis for mammographic density. As early diagnosis improves prognosis, it would be of interest to find biological markers that identify the earliest stages of breast cancer development. Genomic alterations are extensively studied in breast cancer, but hardly in a population with dense mammograms. Representational Oligonucleotide Microarray Analysis (ROMA) has shown to be a reliable tool for detecting genomic alterations (Sebat et al. 525−28). In order to look for early genomic alterations in high-risk women, ROMA was performed on DNA from healthy individuals with increased mammographic density and on breast cancer patients.

Methods: Women referred to hospitals for “second look” after mammographic screening were included in the study. Healthy women with a certain degree of density and women with newly diagnosed breast cancers were included. Tru-cut biopsies were taken and DNA isolated. ROMA was performed on DNA from 16 healthy individuals and 18 breast cancer patients. The microarray data were processed in GenePix Pro 4.0 and analysed using CGH explorer.

Results: ROMA performed on DNA from breast tumors showed classical alterations previously described by groups using ROMA (Hicks et al. 51−63) and conventional CGH-techniques (Kerlikowske et al. 386−95). However, when performing ROMA on DNA from dense breasts we did not find such alterations.

Discussion/Conclusion: We identified typical DNA changes in breast carcinomas, but this pilot study did not show evidence of genomic alteration in women with increased mammographic density. The increased risk for breast cancer development among women with increased mammographic density could be exerted by epigenetic changes. If the increased risk is caused by DNA changes in dense breasts, we may need more samples to detect them.

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