Aberrant activation of USP22 is critical in colorectal cancer progression

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Abstract

BACKGROUND: Recent studies provided strong support for the view that ubiquitin-specific protease 22 (USP22) plays a central role in cell-cycle progression and also in pathological processes such as oncogenesis. We have recently shown that USP22 levels are elevated in colorectal carcinoma with associated increase in the expression of several cell-cycle-related genes. However, the precise mechanism for these functions of USP22 at molecular level has not been fully elucidated.

HYPOTHESIS: The present study was aimed at clarifying clinical significance and exploring molecular mechanism of USP22 in colorectal cancer progression. And we proposed the hypothesis that USP22 may act as an oncogene by activating BMI-1-mediated INK4a/ARF pathway and Akt pathway.

METHODS: USP22 expression was detected with quantitative RT-PCR, Western blot, and immunohistochemistry (IHC) in 43 CRCs and non-cancerous matched fresh tissues. Furthermore, USP22 protein expression was analyzed in 192 CRC tumors by IHC to evaluate the association with survival. With the knock-down of USP22, the changes of cellular proliferation, cell cycle, cell apoptosis, tumor xenograft change in nude mice, and the potential mechanism were investigated.

RESULTS: In 43 paired fresh tissues, the expression level of USP22 was significantly higher in primary CRCs than that in the paired non-cancerous tissues at both mRNA and protein levels ($P < 0.0001$).Nuclear USP22 expression significantly increased fromnormalmucosa through adenoma to primary carcinoma ($P < 0.0001$) and from primary carcinoma to liver metastasis ($P=0.021$). Notably, high USP22 expression was significantly associated with shorter disease-specific survival ($P < 0.0001$) and shorter disease-free survival ($P < 0.0001$). Cox regression analysis showed USP22 was an independent prognostic parameter for CRC patients. The knock-down of USP22 protein expression by miRNA resulted in the inhibition of cellular proliferation, the accumulation of cells in the G1 phase, and the reduction of apoptosis. Furthermore, with orthotopic mice as a model, tumor growth was suppressed when USP22 miRNA silencing vector was injected. RNAi-knockdown of USP22 in HCT16 cells also led to the repression of BMI-1 and was accompanied by the up-regulation of p16INK4a and p14ARF, with a consequent decrease in E2F1 and p53 levels. In addition, down-regulation of c-Myc-targeted cyclin D2 was also noticed in cells treated with USP22-siRNA. Furthermore, our results showed that USP22 deletion also caused down-regulation

of Akt/GSK3b activity, which can also contribute to the reduction of cyclin D2.
CONCLUSION: USP22 might be an independent predictive factor for CRC prognosis and aberrant expression of USP22 may play an essential role in colorectal carcinogenesis and liver metastasis. Collectively, our current results suggest that USP22 may act as an oncogene in CRC progression via regulating both BMI-1-mediated INK4a/ARF pathway and Akt signaling pathway.