USP8 in pituitary corticotroph adenoma pathogenesis

Reincke et al. performed exome sequencing of 10 corticotroph adenomas. They found somatic mutations in the USP8 deubiquitinase gene in 4 of 10 adenomas. The mutations clustered in the 14-3-3 protein binding motif and enhanced the proteolysis cleavage and catalytic activity of USP8. Cleavage of USP8 led to increased deubiqutination of the Epidermal growth factor receptor, impairing its downregulation and sustaining EGFR signaling pathway , which has been implicated in ACTH hypersecretion. USP8 mutants enhanced promoter activity of the gene encoding proopiomelanocortin [1].

Inhibition of USP8 or EGFR is promising for treating USP8-mutated corticotrophin adenoma [2]

Microadenomas that strongly express POMC were common among mutated tumors, which may lead to the mechanisms by which very small adenomas secrete excess ACTH to present overt CD. While USP8 mutations were less likely to enhance tumorous ACTH hypersecretion via EGFR-mediated activation, the presence of USP8 mutations may predict favorable responses to the somatostatin analog pasireotide, which exhibits high affinity for SSTR5. In contrast, non-mutated aggressive tumors such as CCA may respond better to the alkylating agent temozolomide because of their significantly weak expression of MGMT [3].

Testing for USP8 variants can be performed from small amounts of FFPE tissue. NGS showed higher sensitivity for USP8 mutation detection than did Sanger sequencing. Assessment for USP8 mutations may complement histopathological diagnosis [4].

USP8 mutations are well known driver mutations in corticotroph adenomas. Differences in transcriptomic profiles between functioning and silent tumours or tumours with different USP8 status haven't been investigated.

48 patients (28 CD, 20 SCA) were screened for USP8 mutations with Sanger sequencing. 24 patients were included in transcriptomic profiling with Ampliseq Transcriptome Human Gene Expression Core Panel. The entire patients group was included in qRT-PCR analysis of selected genes expression. Immunohistochemistry was used for visualization of selected protein. Results We found USP8 mutation in 15 patients with CD and 4 SCAs. USP8 mutations determine molecular profile of the tumours as showed by hierarchical clustering and identification of 1648 genes differentially expressed in USP8-mutated and USP8-wild type tumours. Mutations affect many molecular pathways as observed in Gene Set Enrichment analysis. USP8-mutated adenomas showed higher level of POMC, CDC25A, MAPK4 but lower level of CCND1, CDK6, CDKN1B than USP8-wt tumours. 87 genes differentially expressed between CD-related adenomas and SCAs were found, including those involved in cell signalling (GLI2, DLC1, TBX2, RASFF6,), cell adhesion (GJA1, CDH6), ion transport (KCNN4,

KCNJ5) and GABA signalling (GABBR2, GABARD). Conclusion USP8 mutations occur in functioning and silent corticotrophinomas. They have pleiotropic effect, not limited to EGFR signalling, and affect expression levels of many genes involved in different pathways. Expression of GABA-related genes GABBR2, GNAL, GABARD and KCNJ5 correspond to functional status of the tumours [5].

Cushing's disease is almost always caused by hypersecretion of adrenocorticotropic hormone (ACTH) from a pituitary neuroendocrine tumor. A mutation in the deubiquitinase gene USP8 has been found in human ACTH-producing pituitary neuroendocrine tumor cells. This mutational hotspot hyperactivates USP8, rescuing epidermal growth factor receptor (EGFR) from lysosome degradation and ensuring its sustained signaling in Cushing's disease. An EGFR inhibitor would be an effective anti-tumor agent in EGFR-related tumors [6].

The discovered aberrant chaperoning activity of heat shock protein 90 on the one hand and the presence of USP8 mutations on the other hand partially explained the causes of their development. Corticotroph tumors arise initially as benign microadenomas but with time form invasively growing aggressive macroadenomas which can switch to corticotroph carcinomas in extremely rare cases. The mechanisms through which corticotroph tumors escape from glucocorticoid negative feedback are still poorly understood, as are the processes that trigger the progression of benign corticotroph adenomas toward aggressive and malignant phenotypes [7].

Weigand et al., demonstrated that USP8 mutations are associated with deregulation of p27/kip1, HSP90, and phosphorylated CREB. These findings suggest that these proteins are direct or indirect clients of USP8 and could therefore be potential targets for therapeutic approaches in patients with CD. [8].

Mutations of the deubiquitinase gene USP8 occur in 35-62% of corticotroph adenomas. However, the major driver mutations in USP8 wild-type tumors remain elusive.

Chen et al., reported recurrent mutations in the deubiquitinase gene USP48 (predominantly encoding p.M415I or p.M415V; 21/91 subjects) and BRAF (encoding p.V600E; 15/91 subjects) in corticotroph adenomas with wild-type USP8. Similar to USP8 mutants, both USP48 and BRAF mutants enhance the promoter activity and transcription of the gene encoding proopiomelanocortin (POMC), which is the precursor of ACTH, providing a potential mechanism for ACTH overproduction in corticotroph adenomas. Moreover, primary corticotroph tumor cells harboring BRAF V600E are sensitive to the BRAF inhibitor vemurafenib.

This study thus contributes to the understanding of the molecular mechanism of the pathogenesis of corticotroph adenoma and informs therapeutic targets for this disease [9].

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