

# Effects of SDHB mutant genotypes on neuroendocrine tumor progression

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## Background

Pheochromocytomas (PHEOs) and paragangliomas (PGLs) are rare tumors of the neuroendocrine system with an underlying etiology that appears to be ~70% sporadic and ~30% hereditary in origin. A significant number of the hereditary PHEOs are caused by mutations in genes coding for the succinate dehydrogenase (SDH) subunits in mitochondrial electron transport chains including SDHB, SDHC, and SDHD, as well as the SDHA-associated protein SDHAF2. Interestingly PHEOs carrying SDHB gene mutations are more highly associated with extra-adrenal locations, overproduction of norepinephrine and dopamine, a high risk of malignancy, and poor therapeutic outcome. The autosomal dominant pattern of inheritance for SDHB related PHEOs suggests a potential dominant negative function of mutated SDHB proteins.

## Methods

To test this hypothesis, we used reverse transcription and PCR cloning to construct an expression plasmid for wild type human SDHB. We then engineered into it, using site directed mutagenesis, several clinically relevant SDHB mutations including a point mutation (P197R), an exon 1 deletion, and a 4 bp deletion at nt847 that results in a frame shift and truncated protein. We also cloned a full-length SDHB antisense expression construct. These constructs were transfected into HEK293 cells to verify the expression of the mutated SDHB proteins from the mammalian expression plasmids by western blotting, and to determine the effect of the mutant SDHB proteins on SDH activity in the recipient cells.

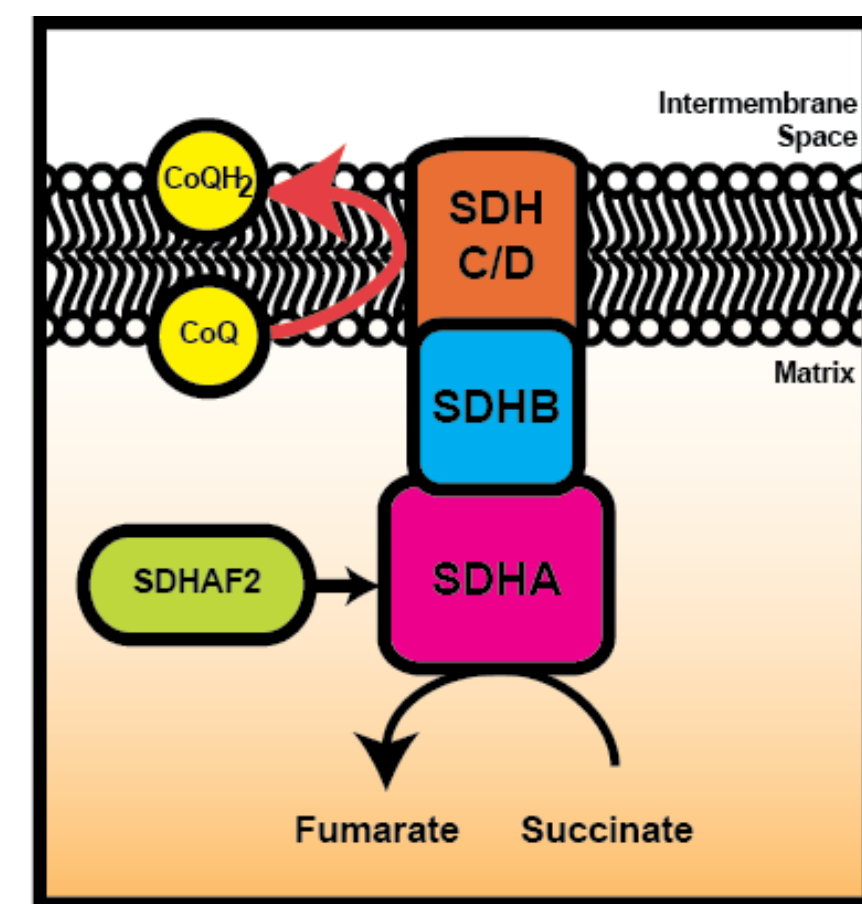
## Results and Conclusions

Clinically relevant human SDHB mutants were transiently expressed in cells and expression of the mutated cDNAs was confirmed by RT-PCR and western blotting. These constructs will be useful to generate stable transfectants of adrenal medulla cells or established PHEO cell lines to determine the effects of perturbing SDH activity on malignant progression in neuroendocrine tumors.

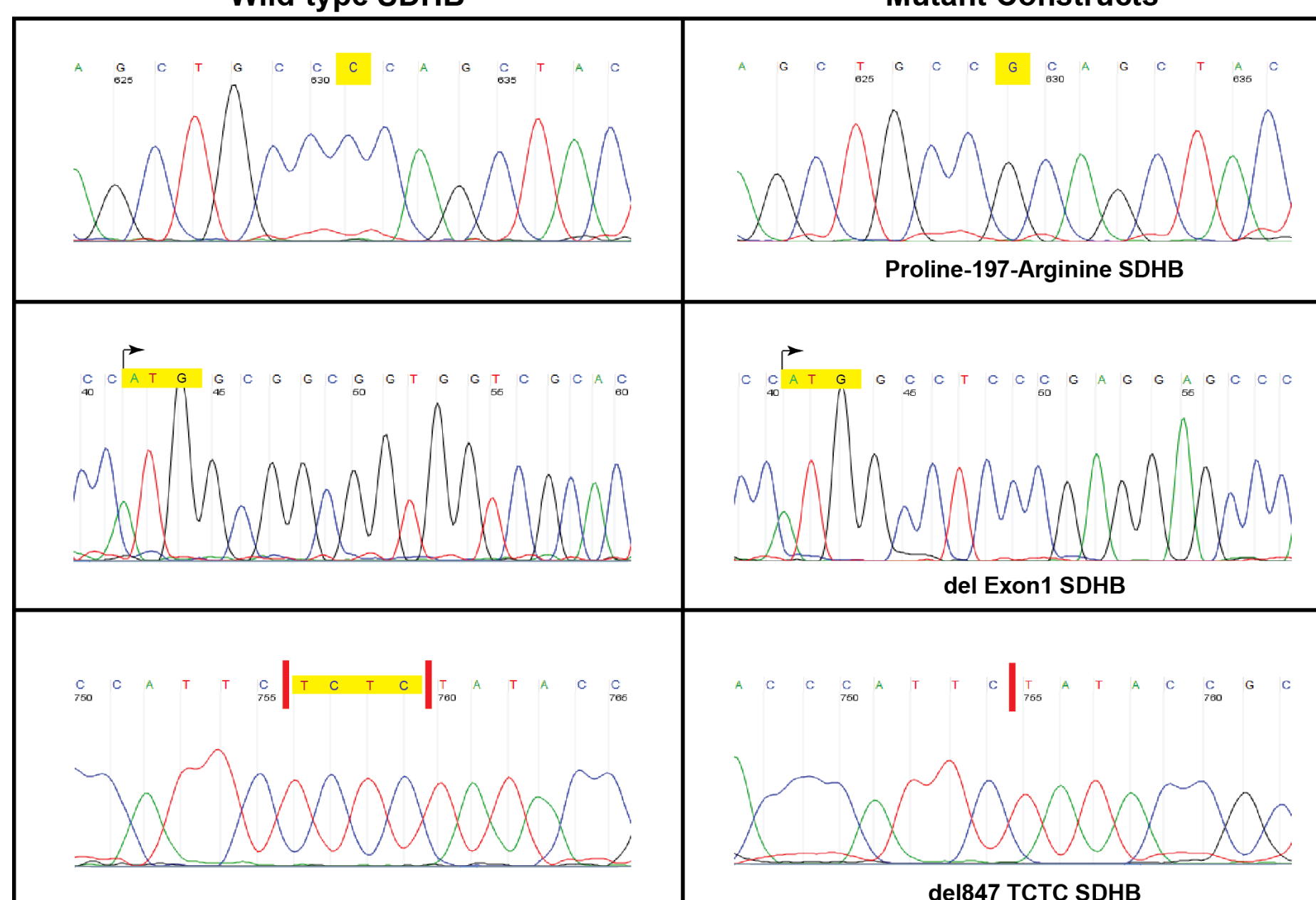
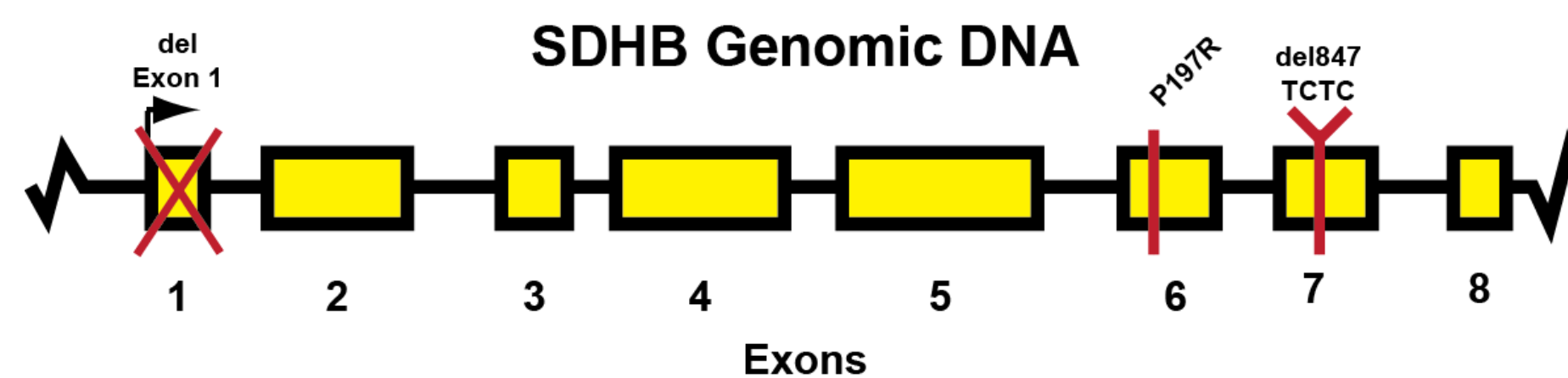
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## References:

Hensen, E.F. and Bayley, J.P. Recent advances in the genetics of SDH-related paraganglioma and pheochromocytoma. *Fam Cancer* 10(2):355-63



GENE	Disease	Number of Mutations
SDHA	Leigh Syndrome, Para/Pheo	Leigh: 7 Para/Pheo: 1
SDHAF2	PGL-2 Para / Pheo	1
SDHB	PGL-4 Sympathetic Para / Pheo	175
SDHC	PGL-3 Para / Pheo	34
SDHD	PGL-1 Head / Neck Para / Pheo	110



## Results

