

Table S7

Materials and methods*Functional divergence and functional distance analyses*

Site-specific evolutionary rates after gene duplication (type I functional divergence) may differ in the protein family, possibly due to altered functional constraints [1]. Maximum likelihood estimate for θ , the coefficient of type I functional divergence, was measured with the program DIVERGE (version 2.0) [2]. A *de novo* neighbor-joining tree was constructed with Poisson distance and re-rooted. Four clusters of the phylogenetic tree, each corresponding to one of the four CatSper groups, were then selected for likelihood ratio tests analysis.

After the coefficient of type I functional divergence for each pair of clusters was calculated, the functional branch length for each cluster, b_F , was estimated to indicate the degree of altered functional constraints [2]. The value of b_F is related to the evolutionary rates of duplicated genes compared with the ancestral gene. $b_F = 0$ suggests the site-specific evolutionary rates of the duplicated gene nearly identical to those of the ancestral gene.

Table S2. Functional divergence between CatSper protein subfamilies

* θ	CatS-2	CatS-3	CatS-4
CatS-1	0.525 ± 0.129	0.433 ± 0.116	0.483 ± 0.146
CatS-2	\	0.408 ± 0.076	0.536 ± 0.116
CatS-3	\	\	0.336 ± 0.092

* θ - Coefficients of type I functional divergence, all $p < 0.05$.

References:

- [1] X. Gu, Maximum-likelihood approach for gene family evolution under functional divergence, *Mol. Biol. Evol.* 18 (2001) 453-64.
- [2] X. Gu, K. Vander Velden, DIVERGE: phylogeny-based analysis for functional-structural divergence of a protein family, *Bioinformatics* 18 (2002) 500-1.