

Supplementary Materials:

Supplementary Figure S1 — Amino acid sequence of the hP2X₅^{FL} subunit compared with best hits in the NCBI protein database. The complete protein sequence of the hP2X₅^{FL} clone used in the present study was aligned with the NCBI database using the BLAST webpage. Available online: <https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE=Proteins> (accessed on 10 January, 2022). The best hit is the exon 10-deleted sequence ID NM_002561 (equal to UniProt Q93086.4) [1,2]. Our hP2X₅^{FL} sequence differs from NM_002561/Q93086.4 by having one leucine residue each at positions 216 and 232 (highlighted in green) instead of a phenylalanine. The mutagenically inserted 22 codons of human exon 10 (GenBank ID AF168787 [2]) are highlighted in yellow-green; within this 22 residues, the 3 residues in bold italics are conservatively different in rat P2X₅. The bolt-blue underlined stretches of residues mark the transmembrane domains TM1 and TM2. The residue stretches highlighted in red and salmon refer to the head domain and the left flipper, which are shown in Supplementary Figure S2 with the same color code. Also shown are individual residues involved in ATP binding in lime-green.

Supplementary Figure S2 — 3D structure of a hP2X₅^{FL} protomer modelled with AlphaFold 2. The 3D structure of the hP2X₅^{FL} subunit was modelled by using AlphaFold 2 [3] and visualized using Pymol version 2.5.2 (<http://www.pymol.org/>). Residues encoded by exon 10 are highlighted in light green and include the pre-TM2 region and the following two-thirds of TM2. For better orientation, the head domain and the left flipper are highlighted in red and salmon, respectively. The two leucine residues, which are phenylalanines in the NCBI sequence NM_002561 [2], are indicated in magenta in the stick presentation. In addition, conserved residues involved in ATP binding as green sticks.

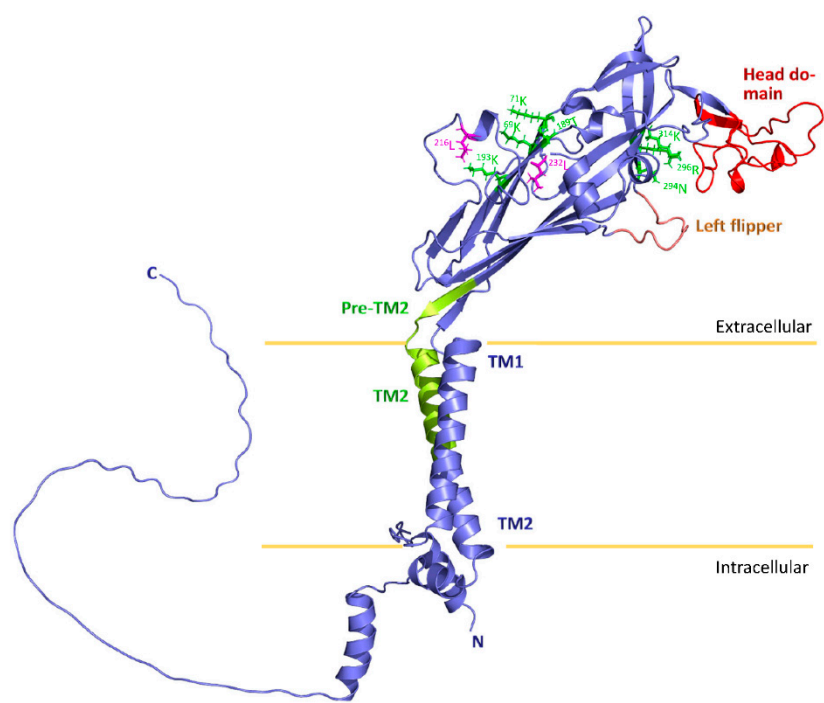
References:

1. Le K.T.; Paquet M.; Nouel D.; Babinski K.; Seguela P. Primary structure and expression of a naturally truncated human P2X₅ ATP receptor subunit from brain and immune system. *FEBS Lett.* **1997**, *418*, 195-199; [https://doi.org/10.1016/s0014-5793\(97\)01380-x](https://doi.org/10.1016/s0014-5793(97)01380-x).
2. Bo X.N.; Jiang L.H.; Wilson H.L.; Kim M.; Burnstock G.; Surprenant A.; North R.A. Pharmacological and biophysical properties of the human P2X₅ receptor. *Mol. Pharmacol.* **2003**, *63*, 1407-1416; <https://doi.org/10.1124/mol.63.6.1407>.
3. Jumper J.; Evans R.; Pritzel A.; Green T.; Figurnov M.; Ronneberger O.; Tunyasuvunakool K.; Bates R.; Zidek A.; Potapenko A.; Bridgland A.; Meyer C.; Kohl S.A.A.; Ballard A.J.; Cowie A.; Romera-Paredes B.; Nikolov S.; Jain R.; Adler J.; Back T.; Petersen S.; Reiman D.; Clancy E.; Zielinski M.; Steinegger M.; Pacholska M.; Berghammer T.; Bodenstein S.; Silver D.; Vinyals O.; Senior A.W.; Kavukcuoglu K.; Kohli P.; Hassabis D. Highly accurate protein structure prediction with AlphaFold. *Nature* **2021**, *596*, 583-589; <https://doi.org/10.1038/s41586-021-03819-2>.

Our hP2X5 ^{FL}	1	MGQAGCKGLCLSLFDYKTEKYVIAKNKKVGLLYRLLQASILAYLVVWVFLIKKGYQDVDT	60
Q93086.4	1	MGQAGCKGLCLSLFDYKTEKYVIAKNKKVGLLYRLLQASILAYLVVWVFLIKKGYQDVDT	60
Our hP2X5 ^{FL}	61	SLQSAVITKVGVAFTNTSDLGQRIWDVADYVIPAQGENVFFVVTNLIVTPNQQRNVCAE	120
Q93086.4	61	SLQSAVITKVGVAFTNTSDLGQRIWDVADYVIPAQGENVFFVVTNLIVTPNQQRNVCAE	120
Our hP2X5 ^{FL}	121	NEGIPDGACSKDSDCHAGEAVTAGNGVKTGRCLRRENLAGTCEIFAWCPLETSSRPEEP	180
Q93086.4	121	NEGIPDGACSKDSDCHAGEAVTAGNGVKTGRCLRRENLAGTCEIFAWCPLETSSRPEEP	180
Our hP2X5 ^{FL}	181	FLKEAEDFTIFIKNHIRFPKFNFSKSNVMDVKDRSLKSCHFPGKNHYCPIFRLGSVIRW	240
Q93086.4	181	FLKEAEDFTIFIKNHIRFPKFNFSKSNVMDVKDRSLKSCHFPGKNHYCPIFRLGSVIRW	240
Our hP2X5 ^{FL}	241	AGSDFQDIALEGGVIGINIEWNCDLKAASECHPHYSFSRLDNKLSKSVSSGYNFRFARY	300
Q93086.4	241	AGSDFQDIALEGGVIGINIEWNCDLKAASECHPHYSFSRLDNKLSKSVSSGYNFRFARY	300
Our hP2X5 ^{FL}	301	YRDAAGVEFRTLMAKYGIRFDVMVNGKAGKFSIPTINVGSGVALMGAFAFFCDLVLIY	360
Q93086.4	301	YRDAAGVEFRTLMAKYGIRFDVMVNGK-----GAFFCDLVLIY	338
Our hP2X5 ^{FL}	361	LIKKREFYRDKKYEEVRGLEDSSQEADEASGLGLSEQLTSGPGLLMPEQQELQEPPEA	420
Q93086.4	339	LIKKREFYRDKKYEEVRGLEDSSQEADEASGLGLSEQLTSGPGLLMPEQQELQEPPEA	398
Our hP2X5 ^{FL}	421	KRGSSSQKNGSVCPQLLEPHRST	444
Q93086.4	399	KRGSSSQKNGSVCPQLLEPHRST	422

Schiller et al. Suppl. Fig. 1

Figure S1. Amino acid sequence of the hP2X5^{FL} subunit compared with best hits in the NCBI protein database.



Schiller et al. Suppl. Fig. 2

Figure S2. 3D structure of a hP2X5^{FL} protomer modelled with AlphaFold 2. .