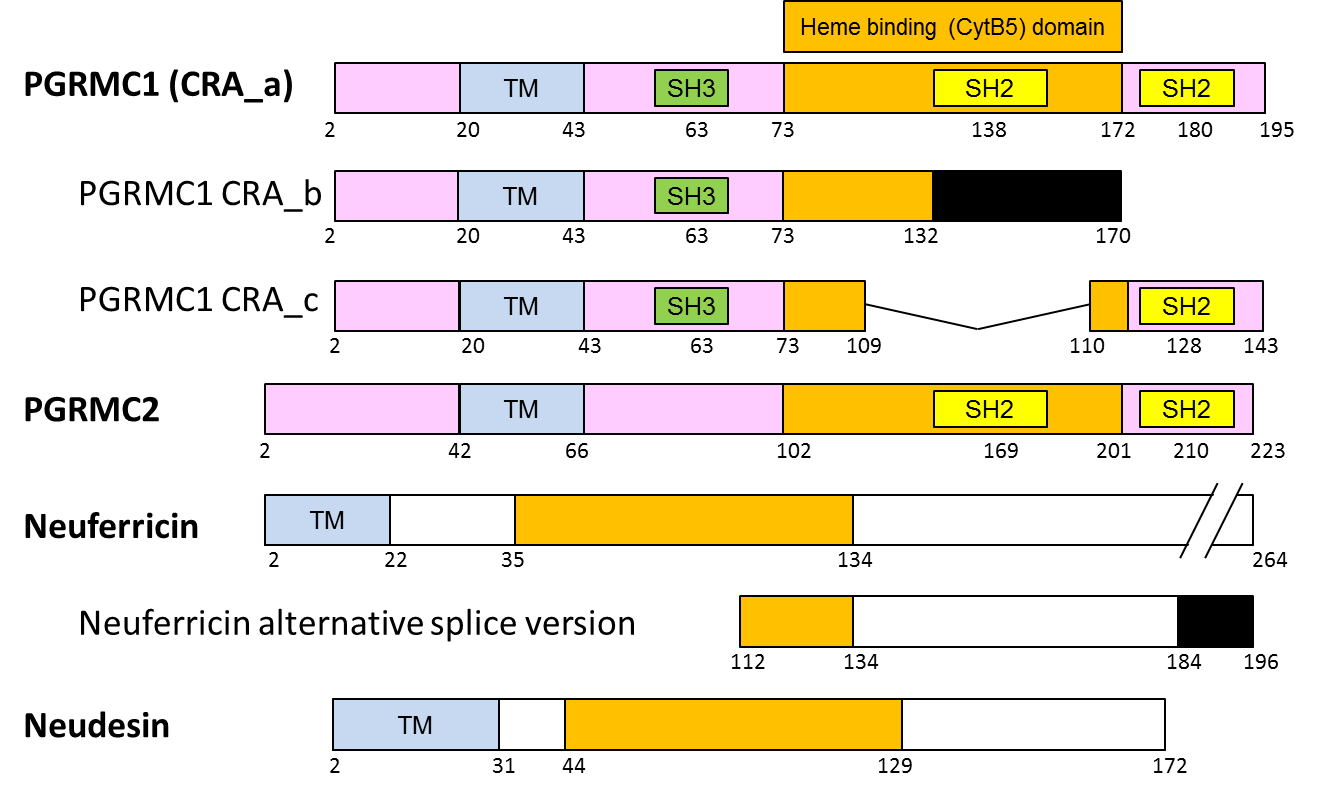
**A) PGRMC1 alignment with human MAPR proteins**



**B) PGRMC1 vertebrate species alignment**

O00264 PGRC1\_HUMAN 1 MAAEDVVATGADPSDLESGGLLHEIFTSPLNLLLLGLCIFLLYKIVRGDQPAASGDSDDD 60

K6ZK25 K6ZK25\_PANTR 1 MAAEDVVATGADPSDLESGGLLHEIFTSPLNLLLLGLCIFLLYKIVRGDQPAASGDSDDD 60

F7BA07 F7BA07\_MACMU 1 MAAEDAVATGADPSELESGGLLHEIFTSPLNLLLLGLCIFLLYKIVRGDQPAASGDSDDD 60

O55022 PGRC1\_MOUSE 1 MAAEDVVATGADPSELEGGGLLHEIFTSPLNLLLLGLCIFLLYKIVRGDQPGASGDNDDD 60

P70580 PGRC1\_RAT 1 MAAEDVVATGADPSELEGGGLLQEIFTSPLNLLLLGLCIFLLYKIVRGDQPGASGDNDDD 60

Q17QC0 PGRC1\_BOVIN 1 MAAEDVAATGADTSELESGGLLQEIFTSPLNLLLLGLCIFLLYKIVRGDQPAAS-DSDDD 59

E2RJW8 E2RJW8\_CANFA 1 MAAEDVAATGADPSELEGGGLLHEIFTSPLNLLLLGLCVFLLYKIVRGDQPAAGGDSDDD 60

Q5ZKN2 PGRC1\_CHICK 1 MAAEEPAMAGEEAVATEGGGLLLEIVGSPLNLSLLGLCLFLLYQILRGERPAAQ--PGEA 58

Q6GQL5 Q6GQL5\_XENLA 1 MA-EE--------------GILQEIFTSPLNICLLCLCLYLLYKILRGDKPQSNENT--- 42

Q5U3G0 Q5U3G0\_DANRE 1 MA-EEA-------VE-QTSGILQEIFTSPLNISLLCLCLFLLYKIIRGDKPADYGPV--- 48

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O00264 PGRC1\_HUMAN 61 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

K6ZK25 K6ZK25\_PANTR 61 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

F7BA07 F7BA07\_MACMU 61 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

O55022 PGRC1\_MOUSE 61 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

P70580 PGRC1\_RAT 61 EPPPLPRLKPRDFTPAELRRYDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

Q17QC0 PGRC1\_BOVIN 60 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 119

E2RJW8 E2RJW8\_CANFA 61 EPPPLPRLKRRDFTPAELRRFDGVQDPRILMAINGKVFDVTKGRKFYGPEGPYGVFAGRD 120

Q5ZKN2 PGRC1\_CHICK 59 GPPPLPKMKRRDFTLEQLRPYDGVRDPRILMAVNGKVFDVTRASKFYGPDGPYGIFAGRD 118

Q6GQL5 Q6GQL5\_XENLA 43 -EEQLPKMKKRDFTRAELQEYDGVQNPRILMAISNKVFDVTRGKKFYGPDGPYGIFAGRD 101

Q5U3G0 Q5U3G0\_DANRE 49 -EEPLPKLKKRDFTLADLQEYDGLKNPRILMAVNGKVFDVTRGKKFYGPEGPYGVFAGKD 107

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O00264 PGRC1\_HUMAN 121 ASRGLATFCLDKEALKDEYDDLSDLTAAQQETLSDWESQFTFKYHHVGKLLKEGEEPTVY 180

K6ZK25 K6ZK25\_PANTR 121 ASRGLATFCLDKEALKDEYDDLSDLTAAQQETLSDWESQFTFKYHHVGKLLKEGEEPTVY 180

F7BA07 F7BA07\_MACMU 121 ASRGLATFCLDKEALKDEYDDLSDLTAAQQETLSDWESQFTFKYHHVGKLLKEGEEPTVY 180

O55022 PGRC1\_MOUSE 121 ASRGLATFCLDKEALKDEYDDLSDLTPAQQETLSDWDSQFTFKYHHVGKLLKEGEEPTVY 180

P70580 PGRC1\_RAT 121 ASRGLATFCLDKEALKDEYDDLSDLTPAQQETLNDWDSQFTFKYHHVGKLLKEGEEPTVY 180

Q17QC0 PGRC1\_BOVIN 120 ASRGLATFCLDKEALKDEYDDLSDLTPAQQETLSDWDSQFTFKYHHVGKLLKDGEEPTVY 179

E2RJW8 E2RJW8\_CANFA 121 ASRGLATFCLDKEALKEEYDDLSDLTPAQQETLSDWDSQFTFKYHHVGKLLKEGEEPTVY 180

Q5ZKN2 PGRC1\_CHICK 119 ASRGLATFCLDKEALRDDYDDLSDLNATQQETLRDWESQFTFKYHHVGKLLKDGEEPTVY 178

Q6GQL5 Q6GQL5\_XENLA 102 ASRGLATFCLDKDALKDTDDDLSDLTATQKETLNDWEEQFTFKYHHVGKLLKAGEEPTEY 161

Q5U3G0 Q5U3G0\_DANRE 108 ASRGLATFCLEKEALKDTHDDLSDLNAMQQESLSEWETQFTQKYDYIGKLLKPGEEPTEY 167

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O00264 PGRC1\_HUMAN 181 SDEEEPKDESARKND- 195

K6ZK25 K6ZK25\_PANTR 181 SDEEEPKDESARKND- 195

F7BA07 F7BA07\_MACMU 181 SDEEEPKDESARKND- 195

O55022 PGRC1\_MOUSE 181 SDDEEPKDETARKNE- 195

P70580 PGRC1\_RAT 181 SDDEEPKDEAARKSD- 195

Q17QC0 PGRC1\_BOVIN 180 SDKEEPKDESTRKND- 194

E2RJW8 E2RJW8\_CANFA 181 SDEEEAKDENARKND- 195

Q5ZKN2 PGRC1\_CHICK 179 SDEEEKDAQDAKKE-- 192

Q6GQL5 Q6GQL5\_XENLA 162 TDDEDAKDSSDSKKKN 177

Q5U3G0 Q5U3G0\_DANRE 168 TDDEEVKDKKKD---- 179

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**C) PGRMC2 vertebrate species alignment**

O15173 PGRC2\_HUMAN 1 ------------------------MAAGDGDVKLGTLGSGSESSNDGGSESPGDAGAAAE 36

H2QQ58 H2QQ58\_PANTR 1 MGGAARGVGEGRGRGGGGRRWRAVMAAGDGDVKLGTLGSGSESSNDGGSESPGGAGAAAE 60

F7GRJ8 F7GRJ8\_MACMU 1 ------------------------MAAGDGDVKLGTLGSGSESSSDGGSESPGGAGAAAE 36

Q80UU9 PGRC2\_MOUSE 1 ------------------------MAAGDGDVKLSTLGSGGESGGD---GSPGGAGATAA 33

Q5XIU9 PGRC2\_RAT 1 ------------------------MAAGDGDVKLSTLGSGGERGGD---GSPGGAGATAA 33

A5PJQ6 A5PJQ6\_BOVIN 1 ------------------------MAAGDGDVNLGTLGSGSESSSDGSSESPGGAGAAAE 36

F1PN69 F1PN69\_CANFA 1 ------------------------MAAGDGDVKLGTLGSGSESSSDGSSESPSGVGAAAE 36

Q5ZLX0 Q5ZLX0\_CHICK 1 -----------------------MADGGDG--RLRT----AESGGDGGAGE--------- 22

Q66IX1 Q66IX1\_XENLA 1 ----------------MSESWE--------------------VEEQLAGTSPPGREGEPS 24

Q7SZC7 Q7SZC7\_DANRE 1 -----------------------MADDGDGRSAVA------DTSGDQGTTEEL------- 24

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O15173 PGRC2\_HUMAN 37 GGGWAAAALALLTGGGEMLLNVALVALVLLGAYRLWVRWGRRGLGAGAGAGEESPATSLP 96

H2QQ58 H2QQ58\_PANTR 61 GGGWAAAALALLTGGGEMLLNVALVALMLLGAYRLWVRWGRRGLGAGAGAGEESPAASLP 120

F7GRJ8 F7GRJ8\_MACMU 37 GGGWAAAALALLTGGGEMLLNVALVALVLLGAYRLWVRWGRRGLGTGAGAGEESPAASLP 96

Q80UU9 PGRC2\_MOUSE 34 RSSWVAA---LLATGGEMLLNVALVALVLLGAYRLWVRWGRRGLCSGPGAGEESPAATLP 90

Q5XIU9 PGRC2\_RAT 34 RSSWVAA---LLATGGEMLLNVALVALVLLGAYRLWVRWGRRGLCSGPGAGEESPAATLP 90

A5PJQ6 A5PJQ6\_BOVIN 37 GGSCLATALALLTGGGEMLLNVALVALVLLGAYRLWVRWGRRGLGAGAGAGEESPAASLP 96

F1PN69 F1PN69\_CANFA 37 GGGWAAAALALLTGGGEMLLNVALVALVLLGAYRLWVRWGRRGLGTGAGAGEESPAASLP 96

Q5ZLX0 Q5ZLX0\_CHICK 23 ------------PAGGGMLLHVGLLALVLLAAYRLYLRWRKRSALG--GAAQQSQAALLP 68

Q66IX1 Q66IX1\_XENLA 25 GLGW--------LFVGELLLNAALVLVLIYGAFRIYQRWKGSGSG---------GAASLP 67

Q7SZC7 Q7SZC7\_DANRE 25 -------DPGAGLGLGGMLLNLSVLVLVLAACYVLYARWWRRA---GADLGRGSEASPLP 74

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O15173 PGRC2\_HUMAN 97 RMKKRDFSLEQLRQYDGSRNPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 156

H2QQ58 H2QQ58\_PANTR 121 RMKKRDFSLEQLRQYDGSRNPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 180

F7GRJ8 F7GRJ8\_MACMU 97 RMKKRDFSLEQLRQYDGSRNPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 156

Q80UU9 PGRC2\_MOUSE 91 RMKKRDFSLEQLRQYDGARTPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 150

Q5XIU9 PGRC2\_RAT 91 RMKKRDFSLEQLRQYDGARTPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 150

A5PJQ6 A5PJQ6\_BOVIN 97 RMKKRDFSLEQLRQYDGSRNPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 156

F1PN69 F1PN69\_CANFA 97 RMKKRDFSLEQLRQYDGSRTPRILLAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLA 156

Q5ZLX0 Q5ZLX0\_CHICK 69 RMKRRDFSLEQLREFDGTRNPRILLAVNGKVFDVTKGSKFYGPEGPYGIFAGRDASRGLA 128

Q66IX1 Q66IX1\_XENLA 68 RMKRRDFTLQQLREYDGTHKPRILLAVNGKVFDVTQGSKSYGPDGPYGLFAGRDASRGLA 127

Q7SZC7 Q7SZC7\_DANRE 75 KMRRRDFTLQQLRDYDGVQNPRILMAVNTKVFDVTSGKKFYGREGPYGIFAGRDASRGLA 134

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O15173 PGRC2\_HUMAN 157 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 216

H2QQ58 H2QQ58\_PANTR 181 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 240

F7GRJ8 F7GRJ8\_MACMU 157 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 216

Q80UU9 PGRC2\_MOUSE 151 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 210

Q5XIU9 PGRC2\_RAT 151 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 210

A5PJQ6 A5PJQ6\_BOVIN 157 TFCLDKDALKDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 216

F1PN69 F1PN69\_CANFA 157 TFCLDKDALKDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 216

Q5ZLX0 Q5ZLX0\_CHICK 129 TFCLDKDALRDEYDDLSDLNAVQMESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDT 188

Q66IX1 Q66IX1\_XENLA 128 TFCLDKEALRDEYDDLSDLNAVQMESVREWEMQFKDKYEYVGRLLKPGEEPSEYTDEEDV 187

Q7SZC7 Q7SZC7\_DANRE 135 TFCLEKDALRDEYDDLSDLNAVQMESVREWEMQFMEKYDYVGRLLKPGDEPSEYTDEEDM 194

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O15173 PGRC2\_HUMAN 217 KDHNKQD 223

H2QQ58 H2QQ58\_PANTR 241 KDHNKQD 247

F7GRJ8 F7GRJ8\_MACMU 217 KDHNKQD 223

Q80UU9 PGRC2\_MOUSE 211 KDHSKQD 217

Q5XIU9 PGRC2\_RAT 211 KDHSKQD 217

A5PJQ6 A5PJQ6\_BOVIN 217 KDHNKQD 223

F1PN69 F1PN69\_CANFA 217 KDHNKQD 223

Q5ZLX0 Q5ZLX0\_CHICK 189 KDHTKQE 195

Q66IX1 Q66IX1\_XENLA 188 RDHTKQD 194

Q7SZC7 Q7SZC7\_DANRE 195 KDHQKHE 201

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**D) PGRMC1 v. PGRMC2 human alignment**

O00264 PGRC1\_HUMAN 1 MAAED-------------------VVATGADPSDLESGGLLHEI---F----TSPLNLLL 34 O15173 PGRC2\_HUMAN 1 MAAGDGDVKLGTLGSGSESSNDGGSESPGDAGAAAEGGGWAAAALALLTGGGEMLLNVAL 60

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O00264 PGRC1\_HUMAN 35 LGLCIFLLYK----IVRGDQPAASGDSDDDEPPPLPRLKRRDFTPAELRRFDGVQDPRIL 90 O15173 PGRC2\_HUMAN 61 VALVLLGAYRLWVRWGRRGLGAGAGAGEESPATSLPRMKKRDFSLEQLRQYDGSRNPRIL 120

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O00264 PGRC1\_HUMAN 91 MAINGKVFDVTKGRKFYGPEGPYGVFAGRDASRGLATFCLDKEALKDEYDDLSDLTAAQQ 150 O15173 PGRC2\_HUMAN 121 LAVNGKVFDVTKGSKFYGPAGPYGIFAGRDASRGLATFCLDKDALRDEYDDLSDLNAVQM 180

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O00264 PGRC1\_HUMAN 151 ETLSDWESQFTFKYHHVGKLLKEGEEPTVYSDEEEPKDESARKND 195

O15173 PGRC2\_HUMAN 181 ESVREWEMQFKEKYDYVGRLLKPGEEPSEYTDEEDTKDHN--KQD 223

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**Figure S3.** Primary structure of PGRMC1-related proteins, and sequence conservation of vertebrate PGRMC1. **A.** The schematic primary isoform of human PGRMC1 (CRA\_a) is aligned with predicted PGRMC1 isoforms CRA\_b and CRA\_C, as well as the other Membrane Associated Progesterone Receptor (MAPR) family members including PGRMC2, two predicted splice variants of Neuferricin, and Neudesin. PGRMC1 CRA\_b and CRA\_c protein sequences are conceptual translations from nucleotide sequences. There is currently no report of their presence as actual proteins. Pink shading of PGRMC1 and PGRMC2 indicates their closely shared protein structure not shared with other MAPR proteins. Black boxes for PGRMC1 CRA\_b and Neuferricin alternative splice version represent predicted amino acids that are not contained in an alternative isoform of the same protein. The conserved heme binding domain (related to cytochrome b5) common to all MAPR family members2 is boxed orange. Blue boxes correspond to the predicted transmembrane helix.

Other abbreviations are based on consensus predicted sequences only and have not been experimentally verified: “SH3” (green boxes) and “SH2” (yellow boxes) denote target sequences for binding of SH3 and SH2 domain proteins to PGRMC11 and corresponding conserved sequences in PGRMC2. Numbers below the proteins depict the amino acid numbers from the following protein sequences: PGRMC1 CRA\_a (UniProt O00264, Genbank NP\_006658, CCDS ID 14576.1); PGRMC1 CRA\_b (Genbank EAW89880.1); PGRMC1 CRA\_c (Genbank EAW89881); PGRMC2 (UniProt O15173); Neuferricin and Neuferricin alternative splice version (UniProt Q8WUJ1); Neudesin (UniProt Q9UMX5). The amino acid number associated with predicted SH2 target sequences for PGRMC1 and PGRMC2 denote the tyrosine residue that would have to be phosphorylated for interaction with SH2 domain proteins. Similarly, proline 63 is at the center of the predicted SH3 target sequence in PGRMC11 that is not present in PGRMC2. A short amino acid loop inserted between helices 3 and 4 of the heme binding domain defines the MAPR family within the larger MAPR family.1,2 In PGRMC11 and PGRMC2 this loop contains a tyrosine as the predicted target for SH2 proteins, absent in Neudesin and Neuferricin.

**B.** Protein sequence alignment of PGRMC1 from (*species;* with UniProt IDs) Human (*Homo sapiens;* O00264), Chimpanzee (*Pan Troglodytes;* K6ZK25), Rhesus Macaque (*Macaca mulatta;* F7BA07), Mouse (*Mus musculus;* O55022), Rat (*Rattus norvegicus;* P70580), Cow, (*Bos Taurus;* Q17QC0), Dog (*Canis familiaris;* E2RJW8), Chicken (*Gallus gallus;* Q5ZKN2), Frog (*Xenopus laevis;* Q6GQL5Z), and Zebrafish (*Danio rerio;* Q5U3G0). PGRMC1 amino acid sequences across species were aligned with the Clustal Omega3 multiple alignment tool on UniProt (http://www.uniprot.org/align/). The N-terminal alignments produced by Clustal Omega for Frog and Zebrafish were corrected manually to align the MA-EE motifs with the N-terminus. Amino acid numbers are given to the right of each line. **Alignment** **Key:** “–”amino acid not present in that species. “\*” identical amino acid across all species. “: ” chemically similar amino acids across species. “.” conserved in all species except one.   
**C.** Alignment of PGRMC2 protein sequences from the same species as **B**, using the same methods with the indicated Protein IDs from UniProt. Based upon the conserved PGRMC1 N-terminus in all other mammals, M25 is the most likely true initiator methionine of the Chimpanzee sequence. **D.** Alignment of human PGRMC1 and PGRMC2 sequences, as above. The Clustal Omega alignment was manually corrected to align D5 across both species.

**References**

1 Cahill, M. A. Progesterone receptor membrane component 1: an integrative review. *J Steroid Biochem Mol Biol* **105**, 16-36, (2007).

2 Mifsud, W. & Bateman, A. Membrane-bound progesterone receptors contain a cytochrome b5-like ligand-binding domain. *Genome Biol* **3**, RESEARCH0068, (2002).

3 Sievers, F. *et al.* Fast, scalable generation of high-quality protein multiple sequence alignments using Clustal Omega. *Molecular systems biology* **7**, 539, (2011).