

**Figure 6**

Multiple sequence alignment of PRF1 protein sequences from human (hg), mouse (mm), rat (rn), chicken (ga), zebrafish (dan), and fugu (fu). The alignment shows conserved regions across species, with some residues highlighted in blue boxes indicating specific amino acid conservation or mutations. Arrows indicate positions corresponding to the PRF1 gene structure shown in Figure 1.

The alignment displays the following sequences:

- hg: MAARLLLLLGILLLLLPVPAPCHTAARSECKRSHKFVPGAWLAGE
- PRF1\_hgA: - - - - -
- PRF1\_hgB: - - - - -

Conserved regions are indicated by arrows at positions 20, 40, 60, 80, 100, 120, 140, 160, 180, 200, 220, 240, 260, 280, 300, 320, 340, 360, 380, 400, 420, 440, 460, 480, 500, 520, and 540.

Key conserved motifs include:

- VVHTPPLHPDFKRALGDLPHHFNASTQPAYLRRLISNYGTHFIRAVE
- LGGRISSALTA LR TC EL ALEG L TDNEVEDCLTVEAQVNII--GIHG
- SISSAEAKACEEKKKKHKMTASFHQTYRE RHSEVVG GHHTSINDLLF
- G IQAGPEQYS AWVNSLPGSPGLVDY TLEPLHVLLDSQDPRREALRR
- A LS QYLTD R ARWRDCSRPCPPGRQKSPRPDCQCVCVCHGS AVTTQDCC
- P RQRGLAQL E VTFIQAWGLWG DWFTATDAYVKLFFGGQE LRTSTVW
- D NNNPIWSVR LD FGDVLLATGGPLRLQVWDQDSGRDDDLLGTCDQA
- P KSGSHEVRCNLNHGH LKFRYHARC LPHLGGGTCLDYVPQM LLGEP

The alignment ends with the sequence: hg PGNRSGAVW, PRF1\_hgA - - - - -, PRF1\_hgB - - - - -.