FILE S1

Alignments of *Ntf-2* and *ran* proteins and Drosophila retroduplicates highlighting amino acid residues of known function

A. Alignments of Dntf-2 retrogenes with their parental and with rat Ntf2. Da_Ntf-2r is shown as Dntf-2_Retana in the alingment and Dg_Ntf-2r is shown as Dntf-2_Retgrim in the alignment.

D.melanogaster

```
Dntf2_D.sim              MSLNPQYEEIGKGFVQQYYAIFDDPANRANVVNFYSATDSFMTFFGHQIQ 50
Dntf2_D.sec              MSLNPQYEEIGKGFVQQYYAIFDDPANRANVVNFYSATDSFMTFFGHQIQ 50
Dntf2_PA_D.mel           MSLNPQYEDIGKGFVQQYYAIFDDPANRANVVNFYSATDSFMTFFGHQIQ 50
Dntf2r_D.mel             MSLNLQYEDIGKEFVQQYYAIFDDPANRENVINFYNATDSFMTFFGNQIQ 50
Dntf2r_D.sim             MSLNPQYEEIGKGFVQQYYAIFDDPVNRENVVHFYSATDSFMTFFGRQIQ 50
Dntf2r_D.sec             MSLNPQYEEIGKGFVQQYYAILDDLANRENAVNFYSVTDSFMTFFGHQIQ 50
Ntf2_R.norvegicus        MGDKPIWEQIGSSFIQHYYQLFDN--DRTQLGAIY-IDASCLTWEGQQFQ 47
                         *. :  :*:**. *:*:** ::*:  :* :   :*    * :*:**.*:*

Dntf2_D.sim              GAPKILEKVQSLSFQFLTRVITTVDSQPTFDGGVLINVLGRICCIDIPFH 100
Dntf2_D.sec              GATKILEKVQSLSFQFLTRVITTVDSQPTFDGGVLINVLGRICCIDIPFH 100
Dntf2_PA_D.mel           GAPKILEKVQSLSFQFLTRVITTVDSQPTFDGGVLINVLGRICCIDIPFH 100
Dntf2r_D.mel             GAPKILEKVQSLSFQFLARVITTVDSQPTSDGGVLIIVLGRIKCIDIPFH 100
Dntf2r_D.sim             GAPKILEKVQSLSFQFLSIVITTVDSQPTFDGGVLISVLGRIKCIDIPFH 100
Dntf2r_D.sec             GAPKILEKVQSLSFQFLSIVITTVDSQPTFDGGVLIFVLGRIKCIDIPFH 100
Ntf2_R.norvegicus        GKAAIVEKLSSLSFQFLQHSITAQDHQPTPDSCIISMVVGQLKADLPIM 97
                         * . *:**:.** ****   **: * *** *. ::  *:*:*:.*:**

Dntf2_D.sim              AFSQVFFLKANAGTFFVAHDIFRLNIHNSA 130
Dntf2_D.sec              AFSQVFVLKANAGTFFVAHDIFRLNIHNSA 130
Dntf2_PA_D.mel           AFSQVFFLKANAGTFFVAHDIFRLNIHNSA 130
Dntf2r_D.mel             AFSQIFLLKPNGGSLFVAHDIFRLNIHNSA 130
Dntf2r_D.sim             SFSQIFLLKPNGGSFLVAHDIFRLNIHNSA 130
Dntf2r_D.sec             SFSQIFLLKPNGGSFLVAHDIFRLNIHNSA 130
Ntf2_R.norvegicus        GFHQMFLLKNINDAWVCTNDMFRLALHNFG 127
                         .* *:*.**   .: . ::*:*** :** .
```

Red box - Interacting interface with RanGDP.

Orange – Amino acids interacting with FxFG repeats of nucleoporins.

D.ananassae

```
Dntf2_Retana             MPLNPHYEPMGQEFVKQYYVIFDNPATRALTATFFSHNDSFMTFFGEQVLGYYKIFEKVK 60
Dntf2_D.ana              MSLNPQYEDIGKGFVQQYYAIFDDPANRANVVNFYSATDSFMTFFGHQIQGAPKILEKVQ 60
Ntf2_R.norvegicus        MGDKPIWEQIGSSFIQHYYQLFDN--DRTQLGAIY-IDASCLTWEGQQFQGKAAIVEKLS 57
                         *  :* :* :*. *:::** :**:   *:    ::    * :*:**.*. *   *.**:.

Dntf2_Retana             SIFFQKVNRTLTNVDCQPTGDGGILMSVLGRICCIDIPSLSFSQIFVLKPDTSPNAYYLS 120
Dntf2_D.ana              SISFQKITRVITTVDSQPTFDGGVLINVLGRICCIDIPPHAFSQVFVLKANAG--TFFVA 118
Ntf2_R.norvegicus        SIFFQKIQHSITAQDHQPTPDSCIISMVVGQLKADLPIMGFHQMFLLKNIND--AWVCT 115
                         **.***: : :*  * *** *. ::  *:*:*:.*:**  .* *:*:**   .  ::  :

Dntf2_Retana             HDIFRLNIHDTE 132
Dntf2_D.ana              HDIFRLNIHNSA 130
Ntf2_R.norvegicus        NDMFRLALHNFG 127
                         :*:*** :*:
```

D.grimshawi

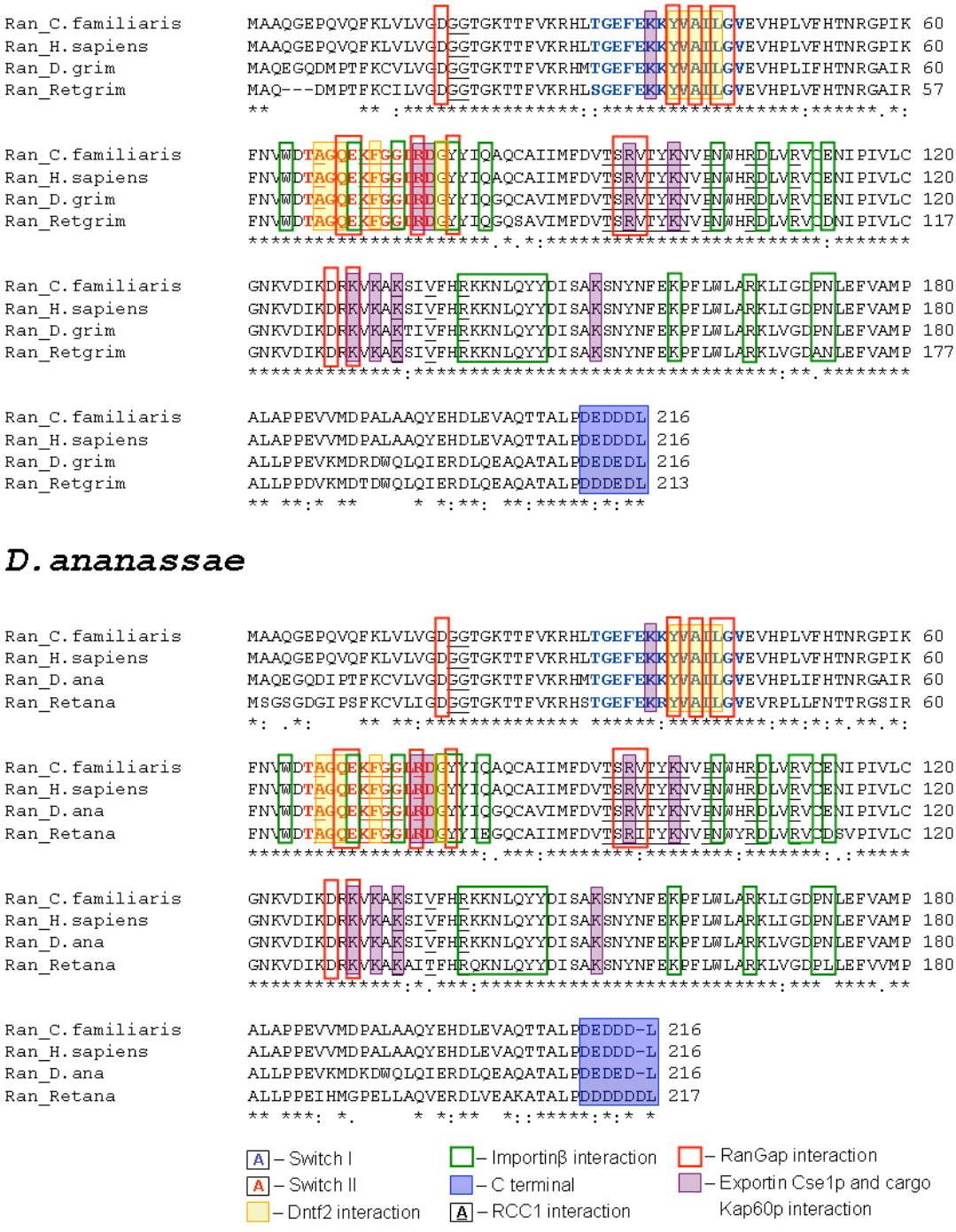
```
Dntf2_Retgrim            MAINPQYEAVGKGFVQQYYAIFDDPANRANVVNFYSTTDSFMTFFGHQIQGAPKILEKVQ 60
Dntf2_D.grim             MALNPQYEDIGKGFVQQYYAIFDDPANRANVVNFYSATDSFMTFFGHQIQGAPKILEKVQ 60
Ntf2_R.norvegicus        MGDKPIWEQIGSSFIQHYYQLFDN--DRTQLGAIY-IDASCLTWEGQQFQGKAAIVEKLS 57
                         *. :* :* :*..*:*:** :**:  :*:::  :*    * :*:**:*:** . *:**:.

Dntf2_Retgrim            SISFQKINRIITTIDSQPTFDGGVLINVLGRIQCIRIPPHSYSQVFLLKANAGSFFVAHD 120
Dntf2_D.grim             SISFQKISRVITTVDSQPTFDGGVLINVLGRVQCIDIPPLAFSQVFVLKANAGTYYVAHD 120
Ntf2_R.norvegicus        SIFFQKIQHSITAQDHQPTPDSCIISMVVGQLKADLPIMGFHQMFLLKNINDAWVCTND 117
                         **.****.: **: * *** *. ::  *:*:::.*:**  .: *:*:**   .::  ::*

Dntf2_Retgrim            IFRLNIHNSA 130
Dntf2_D.grim             IFRLNIHNSA 130
Ntf2_R.norvegicus        MFRLALHNFG 127
                         :*** :** .
```

B. Alignments of Da_Ran-like (Ran_Retana in the alingment) and Dg_Ran-like (RanRetgrim in the alingment) retrogenes with their parental and with dog Ran are shown below. Residues that were inferred to be of major functional importance from published crystallographic results (see text for references) are outlined.

D.grimshawi



D.ananassae