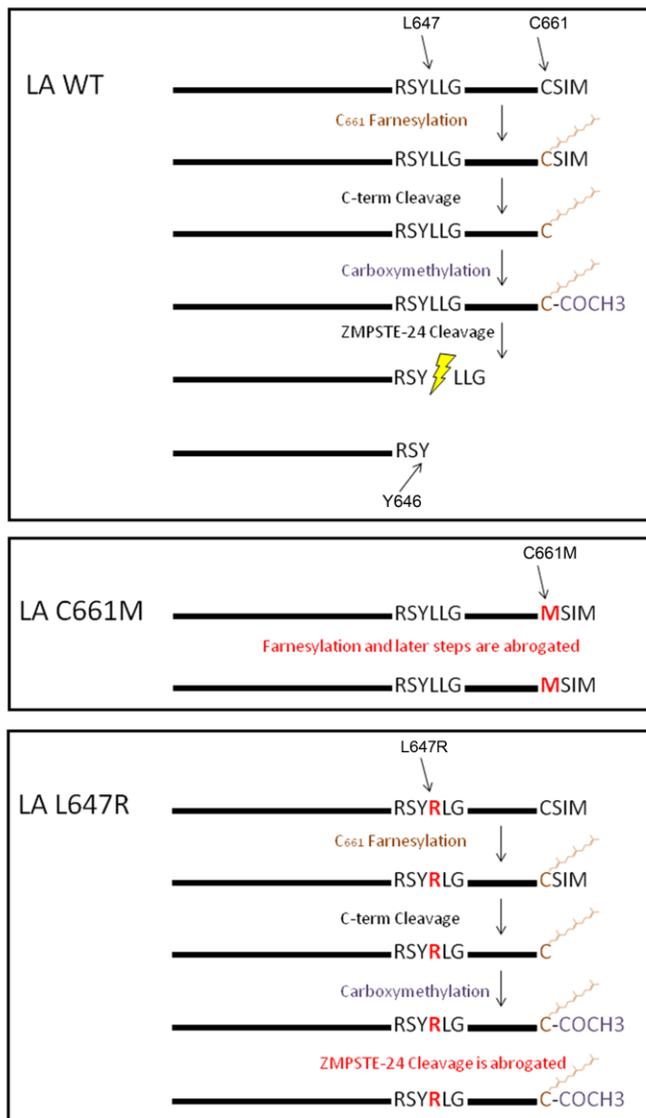


**TABLE S1**

Mutation	Forward Primer	Reverse Primer
C661M	5'- CAGAGCTCCCAGAACATGAGCATCATGTAATC- 3'	5'- GATTACATGATGCTCATGTTCTGGGAGCTCTG- 3'
L647R	5'-CCGCTCCTACCGCCTGGGCAACTC-3'	5'-GAGTTGCCCAGGCGGTAGGAGCGG-3'
R401C	5'-CCATGCCTACCGAAAGCTGC-3'	5'-CCGCTAGTGCGAGCATGCTG-3'
H506P	5'-GAGCTGGGGCCACCCCTAGCCCCCTAC-3'	5'-GTAGGGGGGCTAGGGGTGGCCCCAGCTC-3
R527P	5'-GGAGCAGCCTTCCCACGGCTCTCATC-3'	5'-GATGAGAGCCGTGGGAAGGCTGCTCC-3'

**Supplemental Table S1: List of mutagenic primers used for the production of lamin A and prelamin A mutants.**

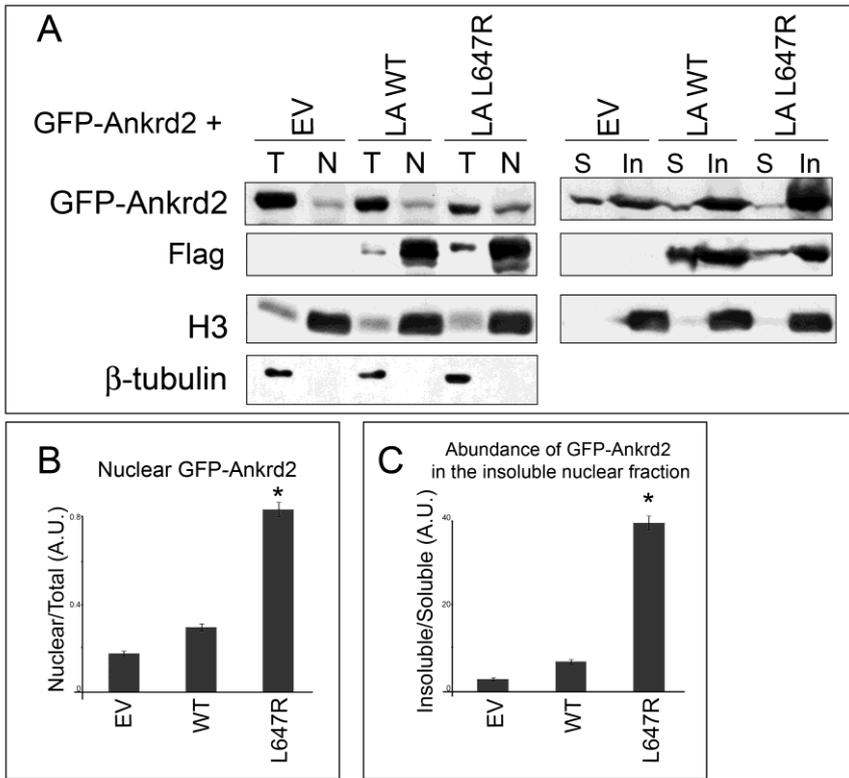
## Supplemental Figure S1



### Supplemental Figure S1: Scheme of lamin A maturation steps, and lamin A unprocessable mutants used in this work.

**LA WT:** Through a multi-step maturation process [29], prelamins A is converted into mature lamin A which is 15 amino acids shorter. In **LA C661M** mutant, maturation is blocked, as Cysteine (C) 661 is mutated into a Methionine (M) and thus no longer recognized by the farnesyltransferase for farnesylation. The final product is a nonfarnesylated prelamins A. Processing of the **LA L647R** mutant is abrogated as the Leucine (L) 647, recognized by the endoprotease ZMPSTE-24, has been mutated into an Arginine (R). As a result, the released product is a farnesylated prelamins A.

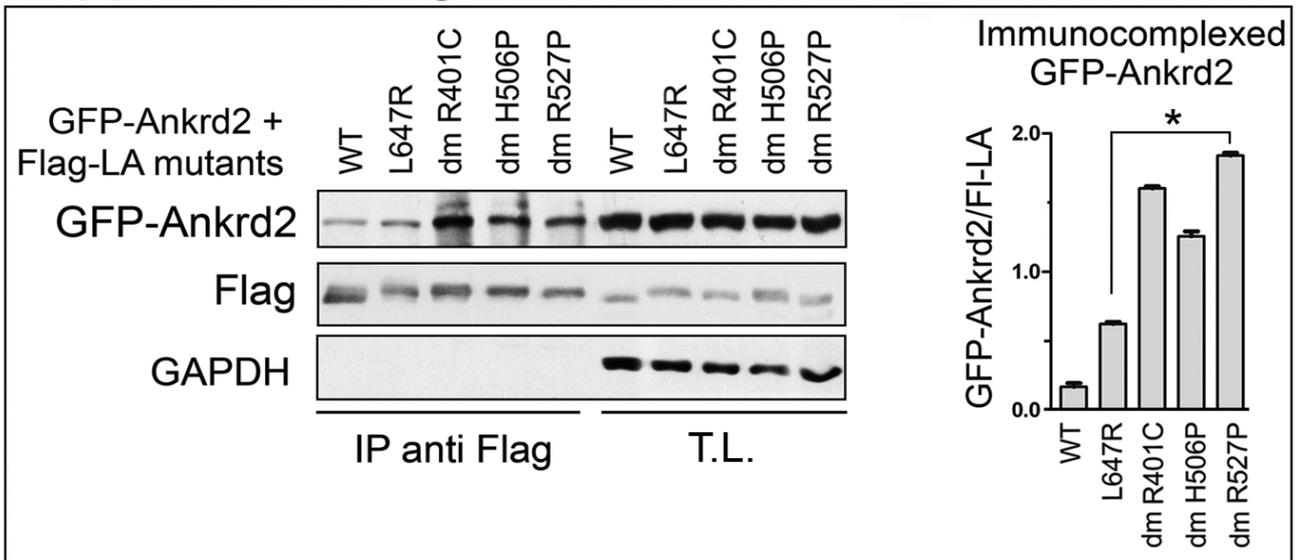
## Supplemental Figure S2



**Supplemental Figure S2: GFP-Ankrd2 is recruited to the insoluble nuclear fraction following the overexpression of farnesylated prelamin A.** **A)** HEK 293T cells were co-transfected with vectors encoding GFP-Ankrd2 and Fl-lamin A wild type (WT), farnesylated prelamin A (L647R) or, as a control, empty vector (EV). After 48 hours, cells were collected and nuclear fractions obtained according to the protocol described in “Methods”. Before the extraction, total extracts (T) were sampled for Western blot analysis. Nuclear fractions (N) were washed twice with ice-cold nuclear extraction buffer and lysed in 3 mM EDTA, 0.2 mM EGTA, 1 mM DTT, and protease inhibitors for 5 min on ice as described by Krishnan and coll. [17]. This fraction was equally divided in two parts: the first one was entirely loaded next to the total one (T) and resolved on the SDS-PAGE; the second one was further fractionated by centrifugation at  $1700 \times g$  for 4 min at  $4^\circ\text{C}$  to give the insoluble pellet (In) containing the chromatin and nuclear matrix proteins and the supernatant, consisting of nucleoplasmic soluble proteins (S). Also these fractions were dissolved in loading buffer and resolved on the SDS-PAGE. Expression of GFP-Ankrd2, Flag,  $\beta$ -tubulin as a control of cytosolic contamination, and Histone 3 (H3, from Santa-Cruz) as control of the insoluble nuclear fraction, were analyzed by Western blotting.

**B)** Immunoblot signals of nuclear GFP-Ankrd2 were quantified by densitometry and the mean data were normalized to those from total GFP-Ankrd2 of the same sample. Graphs are presented as mean  $\pm$ SD of three independent experiments ( $n = 3$ ), \*  $p < 0.05$ . **C)** Immunoblot signals of GFP-Ankrd2 of the insoluble fraction normalized to that found in the soluble one of the same sample. Graphs are presented as mean  $\pm$ SD of three independent experiments ( $n = 3$ ), \* $p < 0.05$  vs. Ankrd2 ratio obtained in Fl-LA WT – GFP-Ankrd2 overexpressing cells.

## Supplemental Figure S3



**Supplemental Figure S3: Ankrd2 tightly interacts with EDMD2-mutated forms of prelamins A.** Co-immunoprecipitation analysis of HEK 293T cells transfected with GFP-Ankrd2 and Flag-lamin A constructs encoding wild type lamin A (LA WT), LA L647R and double mutants L647R/R401C, L647R/H506P and L647R/R527P (dm R401C, dm H506P and dm R527P). After 18-24 hours from transfection, cells were lysed and lamin A species immunoprecipitated from total lysates (T.L.) by anti-Flag antibody. GFP-Ankrd2 amount was then quantified in the Flag-immunocomplexes. GAPDH antibody was used to confirm equal loading. Representative western blots are shown. Bar graph were obtained by normalizing GFP-Ankrd2 to Flag densitometric values. Plots are mean  $\pm$  SD ( $n = 3$ ). \* $p < 0.05$  vs. Ankrd2 amount obtained from Fl-LA WT-transfected cells.