Heterologous prion-forming proteins interact to cross-seed aggregation in *Saccharomyces cerevisiae*

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Supplementary Information

Table S1. Strains used in this study.

<table>
<thead>
<tr>
<th>Strain</th>
<th>Description</th>
<th>Reference</th>
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<tr>
<td>2261</td>
<td>74D-694; [psi-], high [RNQ+], MATA, ade1-14 his3Δ-200 trp1-289 ura3-52 leu2-3,112, sup35Δ::HysBMX4, RNQ1</td>
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<td>2040</td>
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Table S2. Plasmids used in this study.

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<th>Description</th>
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<td>5049</td>
<td>pYK810</td>
<td>Cover SUP35 deletion with SUP35</td>
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<td>5281</td>
<td>p416TEF-SP5</td>
<td>Template for screen</td>
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<td>5787</td>
<td>p415TEF-SP5</td>
<td>Backbone for screen candidate expression</td>
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<td>6244</td>
<td>pEMBL-SUP35</td>
<td>[PSI+] induction – WT</td>
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<td>6257</td>
<td>pRS315-sup35-N5Y</td>
<td>Cover SUP35 deletion with sup35-N5Y</td>
<td>This study</td>
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<td>5074</td>
<td>pPROEX-Htb-RNQ1</td>
<td>WT Rnq1 purification</td>
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<td>SL7111</td>
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<td>WT Sup35 bait for Rnq1-trap on resin</td>
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<td>6771</td>
<td>pAED4-SCNM-G7C-his7</td>
<td>Purification for crosslinking reagent</td>
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<td>6772</td>
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<td>Control bait for Rnq1-trap on resin</td>
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<td>pRS313-rnq1-Q298R</td>
<td>Mitotic stability testing of Rnq1-Q298R</td>
<td>This study</td>
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<td>6312</td>
<td>pRS313-RNQ1</td>
<td>Mitotic stability testing of WT Rnq1</td>
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Table S3. Antibodies used in this study.

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<th>Source</th>
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<td>Lindquist lab</td>
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<td>Rnq1</td>
<td>Full-length Rnq1</td>
<td>Rabbit polyclonal</td>
<td>1:1,000</td>
<td>True lab</td>
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<tr>
<td>Rnq1</td>
<td>Full-length Rnq1</td>
<td>Rat polyclonal</td>
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<td>True lab</td>
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<td>Rabbit</td>
<td>Rabbit IgG</td>
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<tr>
<td>Rat</td>
<td>Rat IgG</td>
<td>Rabbit polyclonal</td>
<td>1:10,000</td>
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</table>
Figure S1. Rnq1-Q298R aggregates have similar properties to WT Rnq1 aggregates
Representative western blots from thermal stability experiments quantified in Figure 3B. Cell lysates were subjected to a temperature gradient before SDS-PAGE and western blotting. Insoluble material, such as prion aggregates, requires treatment at high temperatures in order to enter the resolving gel. There were no observable differences between WT and Rnq1-Q298R aggregates.
Figure S2. Mutants isolated from SP5 suppressor screen
(A) Full spottings of screen candidates from Figure 4A. (B) SDD-AGE analysis demonstrates that the rescuing mutants Sup35-N5Y, Sup35-Q6R, and Sup35-G116V can all propagate [PSI*].
Figure S3. Boiled gel assay with additional [rnr] controls

As in Figure 5B, the indicated yeast strains we subjected to a boiled gel protocol to assess the aggregated vs monomeric Sup35. No aggregated Sup35 was detectable in any of the [rnr] strains, regardless of genetic background.
Figure S4. Full crosslinking blots
(A) SDD-AGE from Figure 6, probed with rat anti-Rnq1 primary and rabbit anti-rat secondary antibodies. (B) Complete SDS-PAGE blot from Figure 6. Two exposures are shown to allow for clear viewing of the recombinant Rnq1 in lanes 1 and 3.