

Supporting Information

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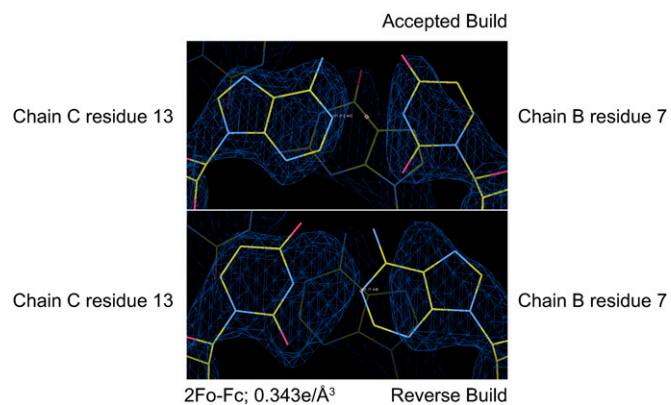


Fig. S1. Assignment of the dsRNA orientation in the hOAS3.DI•dsRNA19 crystal structure. The orientation of the dsRNA was determined by refining the data against build 1 (accepted) and build 2 (reverse). Biased electron density maps are consistent only with build 1.

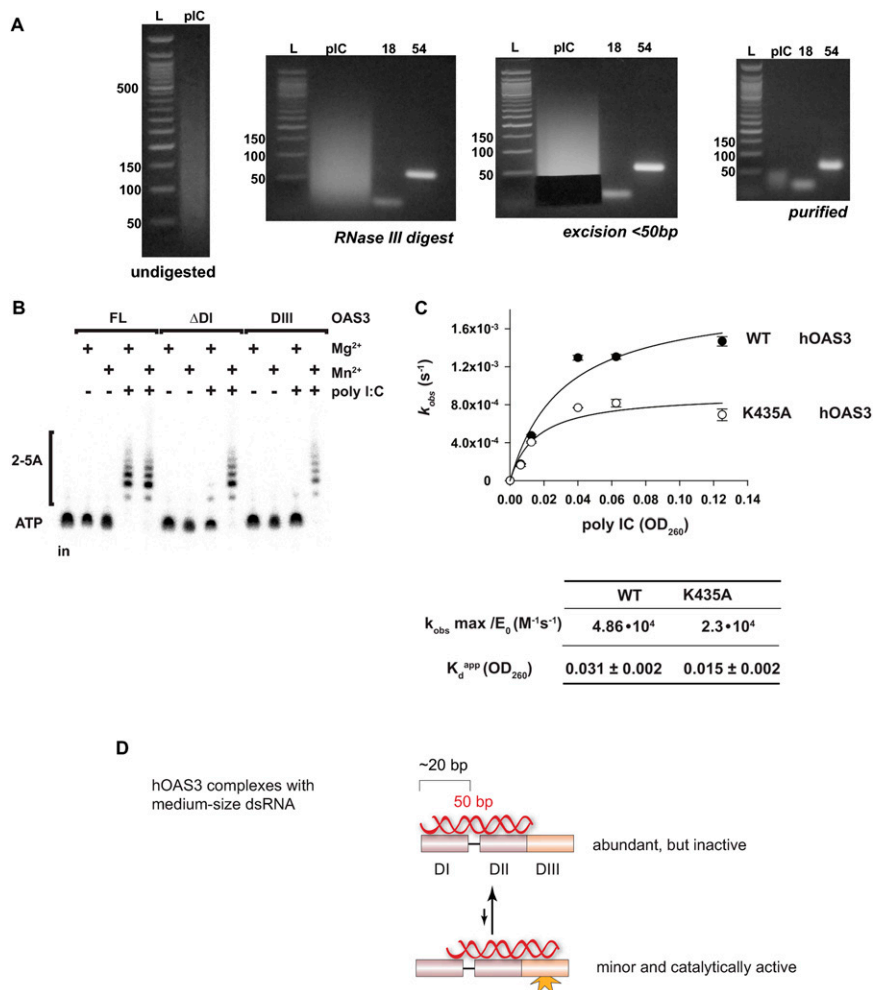


Fig. S5. Preparation of <50-bp poly I:C, and a model for its recognition by hOAS3. (A) Agarose gels (4%) of poly I:C (Left), poly I:C digested with RNase III (Center), and purified digested poly I:C (Right). Long (undigested) poly I:C used in this study ranges from less than 50 bp to ~500 bp, with a median size ~120 bp. Gel markers: L, 50-bp DNA ladder (New England Biolabs); 18, dsRNA18 (Methods); 54, a 54 bp in vitro transcribed dsRNA size control. (B) Two-hour end point activity assays with full length (FL) and truncated hOAS3 in the presence of Mg²⁺ and Mn²⁺. We find that Δ DI has robust activity in the presence of Mn²⁺, confirming that it is properly folded and well behaved. (C) Mutagenesis of a predicted dsRNA-binding contact in domain DII of hOAS3 (see also Fig. S3). Reactions were conducted as in Fig. 4B. (D) A proposed model for recognition of medium-size poly I:C by hOAS3, which explains the low specific activity with shorter dsRNA.

Table S1. Data collection and refinement statistics

Human OAS3.DI•dsRNA19

Data collection	
Space group	P2 ₁
Cell dimensions	
<i>a</i> , <i>b</i> , <i>c</i> , Å	48.7, 104.7, 63.8
α , β , γ , °	90, 98, 90
Resolution, Å	43.82–2.0 (2.1–2.0)*
R-meas, %	6.6 (67.4)
CC(1/2)	99.9 (88.9)
<i>I</i> / σ <i>I</i>	21.33 (3.25)
Completeness, %	99.8 (100)
Redundancy	6.27 (6.25)
Refinement	
Resolution, Å	2.0
No. unique reflections	42,802 (5,835)
<i>R</i> _{work} / <i>R</i> _{free}	0.1818/0.2113
No. atoms	
Protein	2,707
dsRNA19	799
Water	876
B-factors	
Protein	48.06
dsRNA19	84.87
r.m.s. deviations	
Bond lengths, Å	0.007
Bond angles, °	0.997
Ramachandran plot, %	
Favored	92
Allowed	8
Disallowed	0.0

A single crystal was used for data collection.

*Highest resolution shell is shown in parenthesis.