

| Helix | Base pair | Sequence | Fold reduction in initial rate |
|-------|--------------|------------|--------------------------------|
| P1 | 3-95 4-94 | G-U A-U | 1 (no reduction) |
| | | G G A G | 5 |
| | | C U C U | 13 |
| | | C-G C-G | 2 |
| P2 | 14-50 | U-A | 1 |
| | | U C | > 20 |
| | | GA | > 20 |
| | | G-C | 1 |
| Р5 | 66-86 | U-A | 1 |
| | | UU | 1.4 |
| | | AA | 10 |
| | | A-U | 1 |

Supplementary Figure 2 Testing the secondary structure model of kinase ribozyme 5-16 by site-directed mutagenesis. (a) Identification of the minimized catalytic core of kinase 5-16 by deletion analysis. Initial rates were measured in 1 mM GTP γ S. Time points were taken within the first 60 minutes of the reaction, and analyzed by APM polyacrylamide gel elec trophoresis. (b) Testing proposed base pairs with compensatory mutations. Initial rates were measured as described. Mutations in P1 and P5 were tested in the context of the full length ribozyme, while mutations in P2 were tested in the context of the ribozyme's minimized catalytic core. Numbering of base pairs acc ording to alignment in Fig. 4.

b