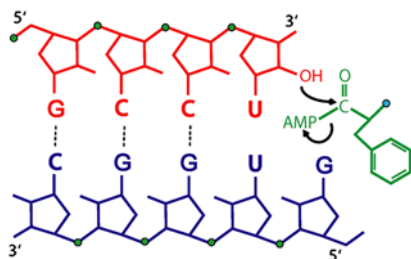


# Supporting Information

Turk et al. 10.1073/pnas.0912895107



**Fig. S1.** Schematic of reaction. Ribozyme GUGGC reacts with PheAMP and RNA substrate GCCU to form a bond between phenylalanine and the 2'-OH of the substrate RNA.

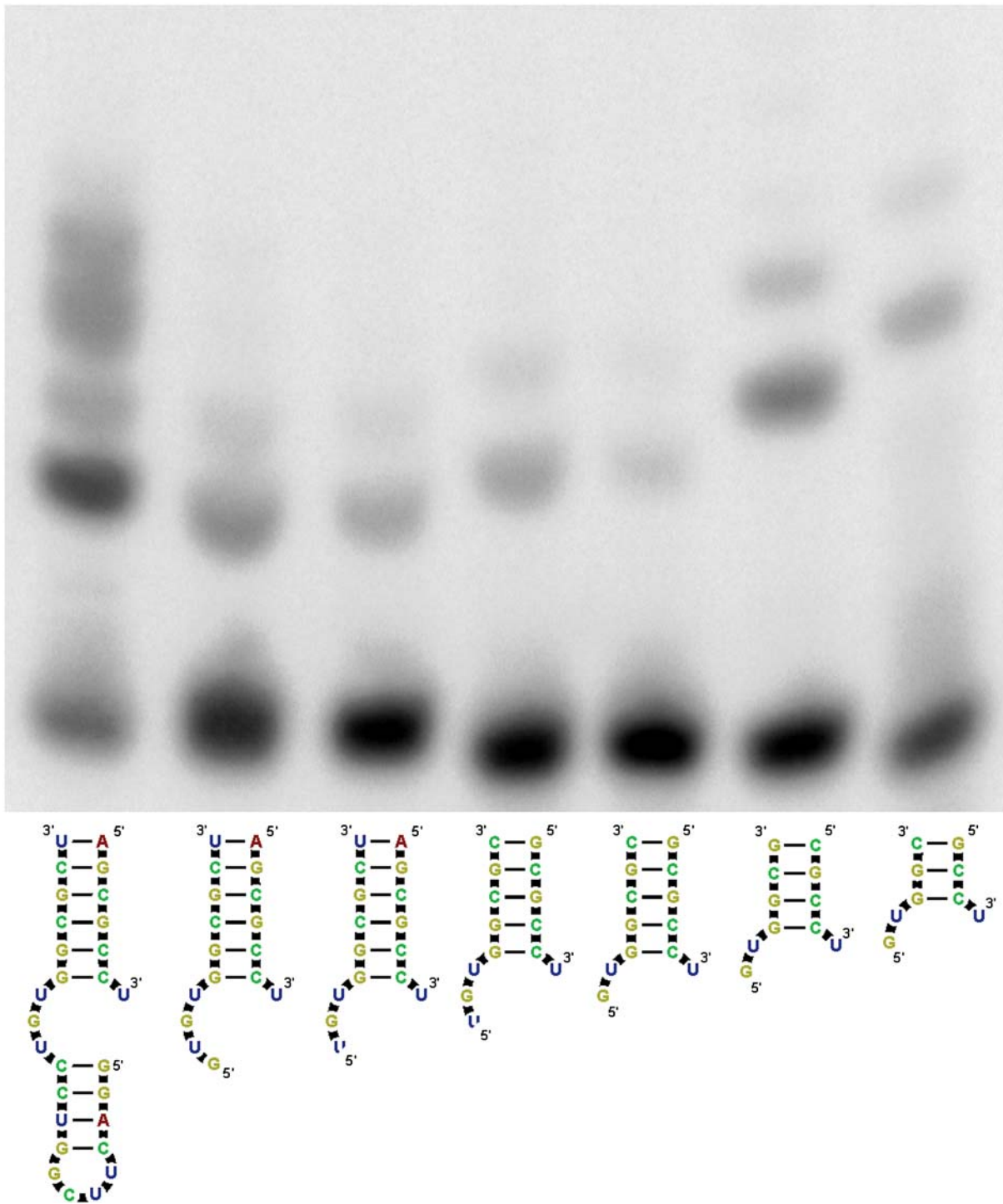
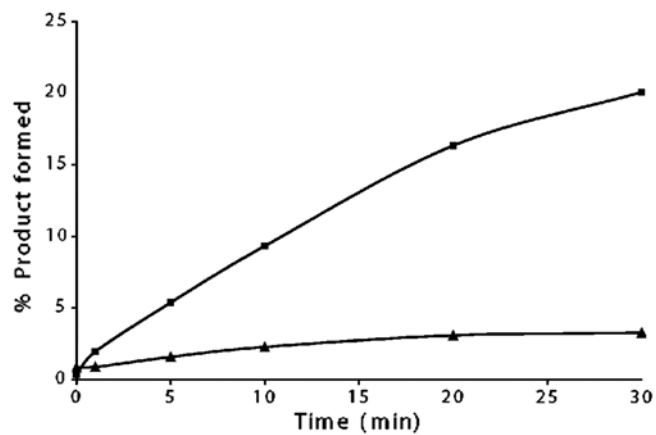
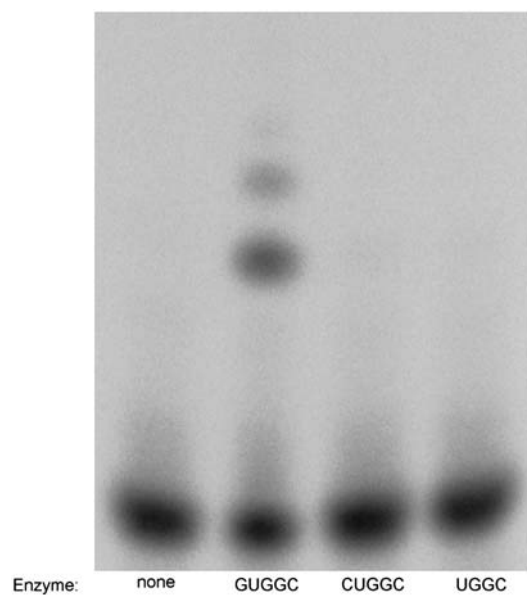


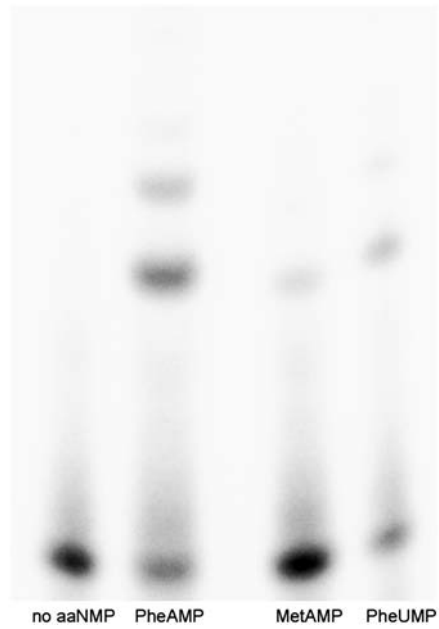
Fig. S2. Intermediate *trans* complexes react with PheAMP. Reaction conditions: 100  $\mu$ M enzyme RNA, 37  $\mu$ M substrate RNA, 100 mM KCl, 5 mM MgCl<sub>2</sub>, 100 mM HEPES pH 7.0, 2.6 mM PheAMP, 4 °C for 10 min.



**Fig. S3.** A 3'-dU substrate retains reactivity. ■, GCCU;  $k_2 = 6.54 \text{ M}^{-1} \text{ min}^{-1}$ ; ▲, GCCU3'dU;  $k_2 = 1.16 \text{ M}^{-1} \text{ min}^{-1}$ . For simplicity, these rates were calculated assuming all GUGGC participates, though the ribozyme oligonucleotide may not be saturated with GCCU. Under these conditions, PheAMP hydrolyzes at  $0.0327 \text{ min}^{-1}$ . Reaction conditions:  $10 \mu\text{M}$  ribozyme GUGGC,  $20 \mu\text{M}$  RNA substrate,  $100 \text{ mM}$  KCl,  $5 \text{ mM}$   $\text{MgCl}_2$ ,  $100 \text{ mM}$  Hepes pH 7.0,  $1.8 \text{ mM}$  PheAMP,  $4^\circ\text{C}$ .



**Fig. S4.** The 5'-G of GUGGC is required for reaction. Reaction conditions:  $25 \mu\text{M}$  enzyme RNA,  $50 \mu\text{M}$  GCCU,  $100 \text{ mM}$  KCl,  $5 \text{ mM}$   $\text{MgCl}_2$ ,  $100 \text{ mM}$  Hepes pH 7.0,  $2.6 \text{ mM}$  PheAMP,  $4^\circ\text{C}$  for 30 min.



**Fig. S5.** GUGGC/GCCU reacts with varying aminoacyl substrates. Reaction conditions: 20  $\mu$ M GUGGC, 10  $\mu$ M GCCU, 100 mM KCl, 5 mM  $MgCl_2$ , 100 mM Hepes pH 7.0, 4  $^{\circ}C$  for 30 min; 11.8 mM PheAMP, 14.5 mM MetAMP, or 12.6 mM PheUMP where indicated.

