

Supplemental figure legends.

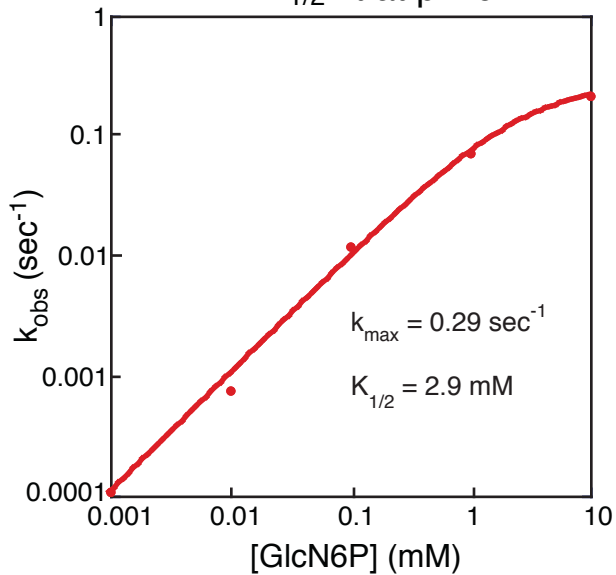
S1. Representative data from a single trial used to do kinetic analysis of dependence of cleavage rate on concentration of GlcN6P. Each panel is at a different, labeled pH. Each point is the calculated rate of the reaction for a stated pH and GlcN6P concentration for a single experiment. Three independent experiments were done to calculate the final reported k_{\max} and $K_{1/2}$ values. In all panels, the fit is shown in a solid red line and the values of k_{\max} and $K_{1/2}$ obtained for this experiment are displayed.

S2. Titration of GlcN6P monitored by ^{31}P NMR. A. Overlay of the ^{31}P spectra collected at various pHs. B. Plot of pH versus ^{31}P chemical shift of GlcN6P. Fit is to equation (4) in the main text.

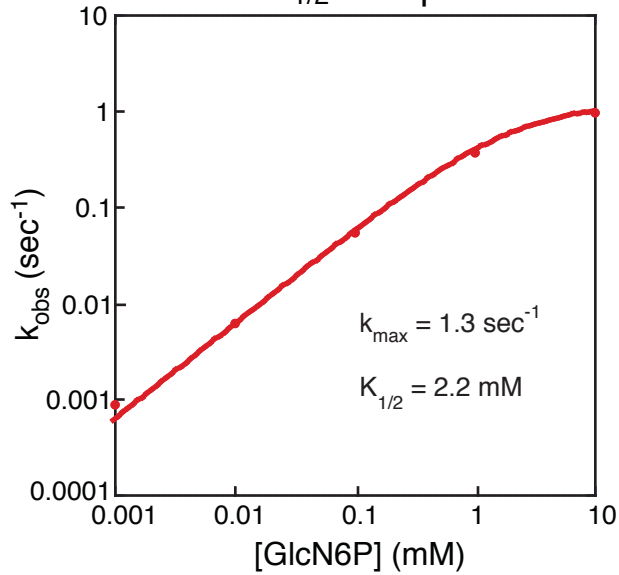
S3. Representative data from a single trial used to do kinetic analysis of dependence of cleavage rate on concentration of MaN6P. Each panel is at a different, labeled pH. Each point is the calculated rate of the reaction for a stated pH and MaN6P concentration for a single experiment. Three independent experiments were done to calculate the final reported k_{\max} and $K_{1/2}$ values. In all panels, the fit is shown in a solid red line and the values of k_{\max} and $K_{1/2}$ obtained for this experiment are displayed.

S1. Kinetic analyses of GlcN6P activation of the *glmS* ribozyme.

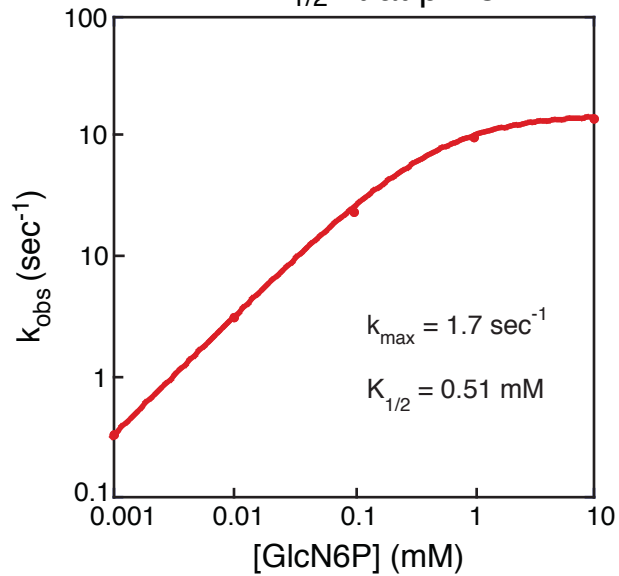
$K_{1/2}$ fit at pH 6



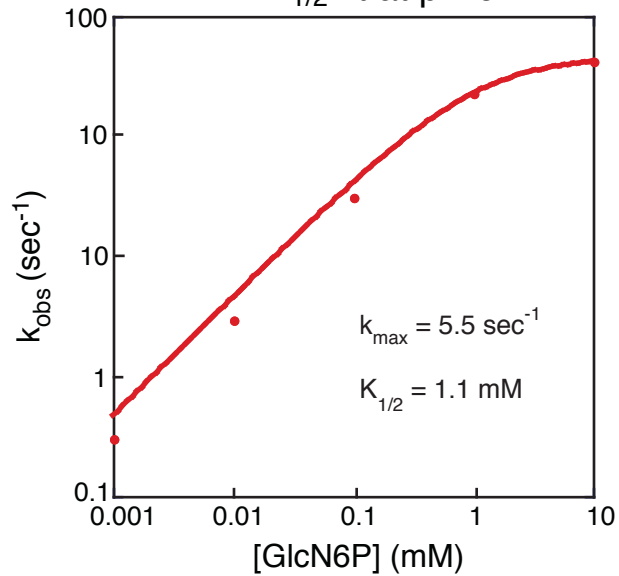
$K_{1/2}$ fit at pH 7



$K_{1/2}$ fit at pH 8



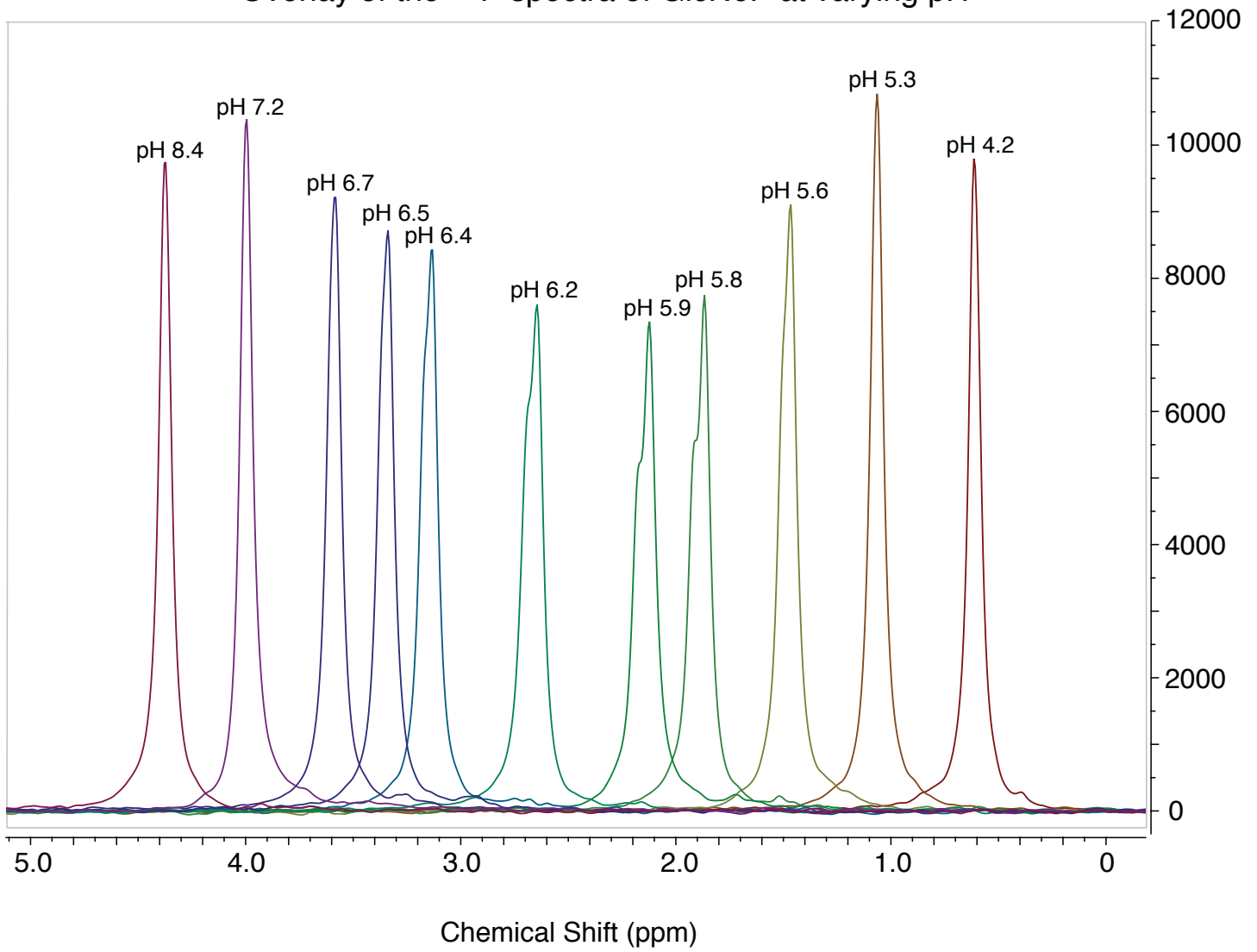
$K_{1/2}$ fit at pH 9



S2. Titration of GlcN6P monitored by ^{31}P NMR

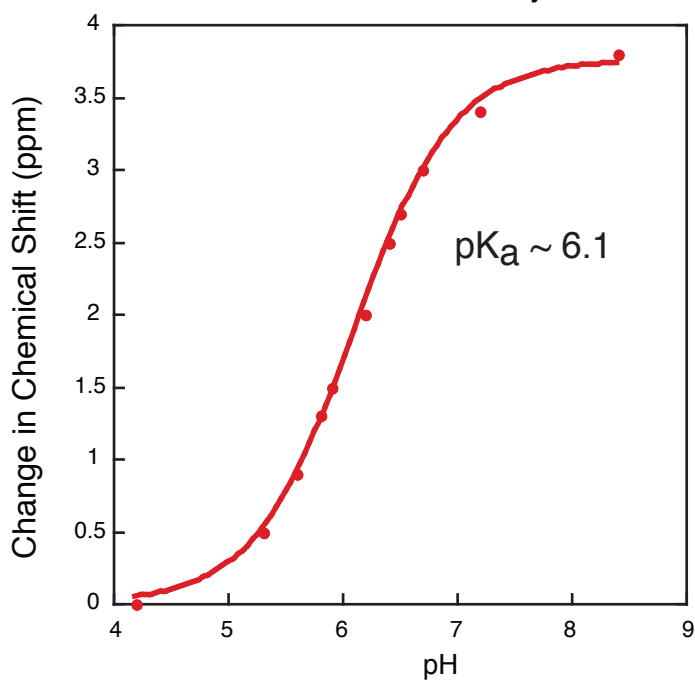
A

Overlay of the ^{31}P spectra of GlcN6P at varying pH



B

Titration of GlcN6P monitored by ^{31}P NMR



S3. Kinetic analyses of MaN6P activation of the *glmS* ribozyme.

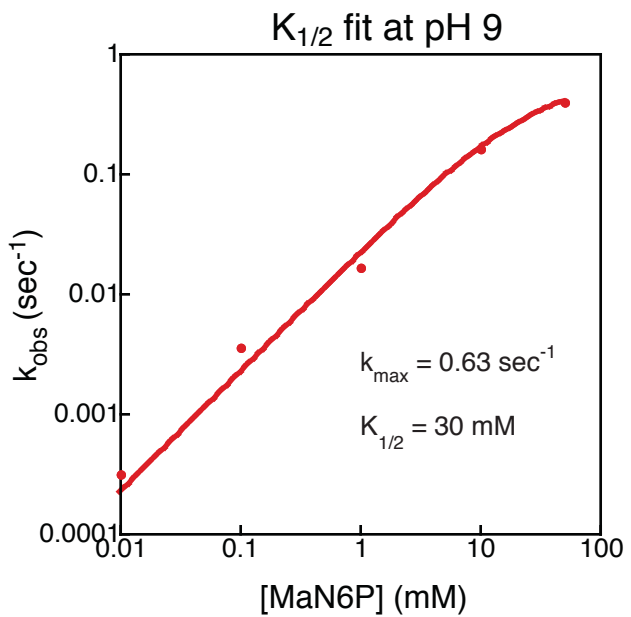
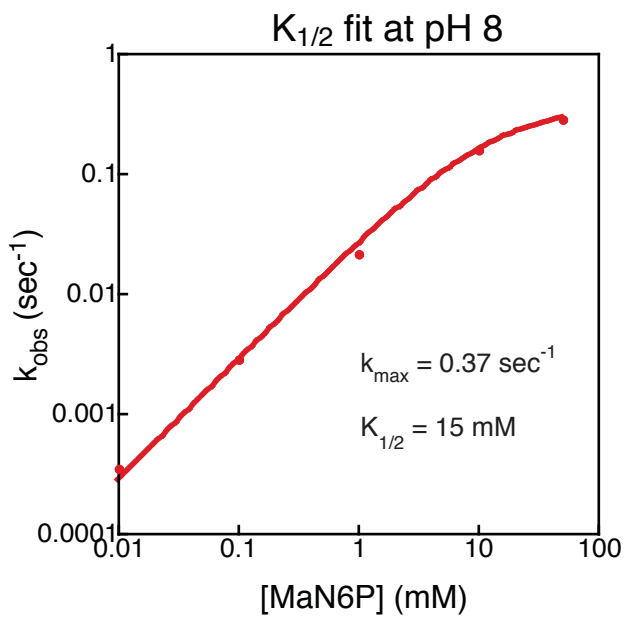
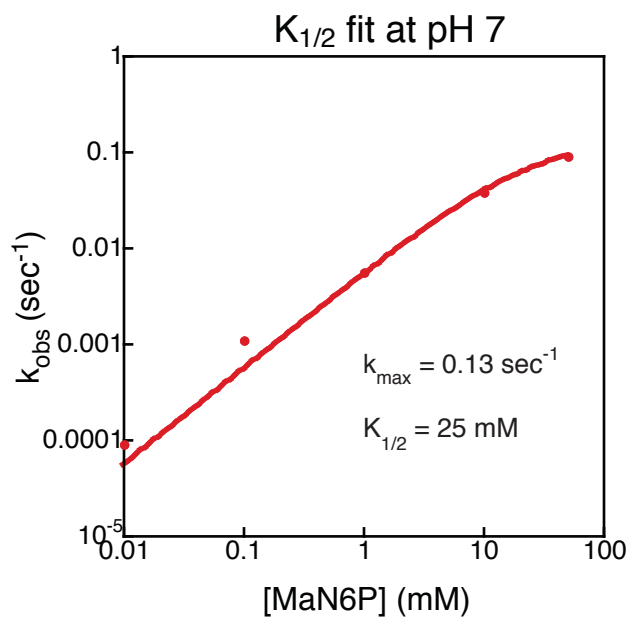
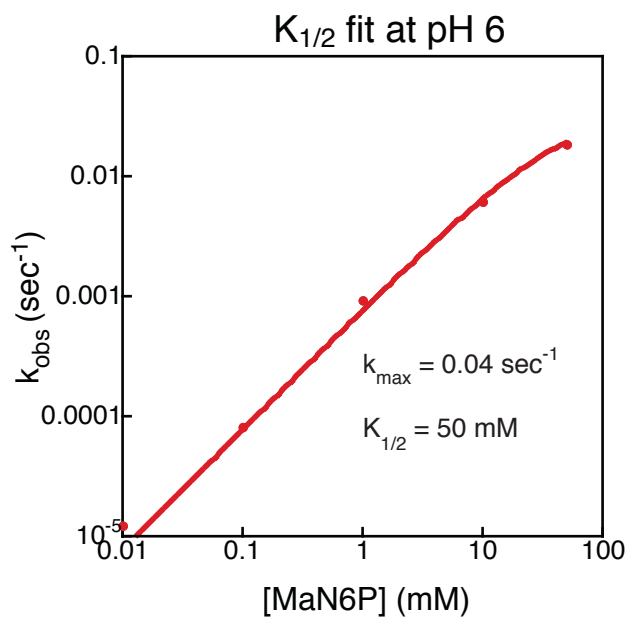


Table S1. Summary of crystallographic statistics.

	Pre-cleavage	Product	G33A	Glc6P bound	MaN6P bound
Data collection					
Space group	P2 ₁	P2 ₁	P2 ₁	P2 ₁	P2 ₁
Cell dimensions					
a, b, c (Å)	47.4, 228.5, 103.9	49.6, 229.8, 101.7	48.4, 232.7, 106.6	47.9, 234.9, 104.5	47.4, 232.4, 104.7
α , β , γ (°)	90.0, 90.7, 90.0	90.0, 90.1, 90.0	90.0, 92.2, 90.0	90.0, 90.6, 90.0	90.0, 90.6, 90.0
Resolution	50-3.10 (3.21-3.10)	50-2.90 (3.00-2.90)	50-3.00 (3.11-3.00)	50-2.85 (2.95-2.85)	50-3.00 (3.11-3.00)
R _{sym} (%) ^a	21.9 (>100)	13.5 (>100)	12.0 (>100)	6.2 (83.2)	14.0 (77.9)
$\langle I \rangle / \langle \sigma I \rangle$ ^a	5.8 (1.3)	10.7 (1.3)	9.8 (1.3)	18.5 (1.1)	8.5 (1.6)
Completeness (%) ^a	99.4 (98.4)	99.6 (97.6)	97.6 (94.6)	98.5 (85.5)	92.9 (90.6)
Redundancy ^a	4.0 (3.8)	4.0 (3.9)	4.1 (4.0)	4.0 (3.2)	3.9 (3.9)
Wavelength (Å)	1.1	1.1	1.1	1.1	1.1
Refinement					
Resolution (Å)	43.8-3.10	49.7-2.90	43.8-3.10	50.0-2.85	40.2-3.0
No. reflections ^b	37270 (1964)	47487 (2538)	44048 (2360)	49868 (2665)	39278 (2127)
R _{work} /R _{free} (%)	28.1/32.7	26.6/30.7	25.7/31.2	24.9/30.9	24.8/29.1
R.M.S. deviations					
Bond lengths (Å)	0.006	0.006	0.006	0.01	0.007
Bond angles (°)	1.261	1.328	1.239	1.502	1.359

^aValues in parentheses are for the highest resolution shell. ^bValues in parentheses are for the cross validation test set.

Table S2. Summary of *glmS* structures to date.

PDB ID	Source	Description	Ref.
2GCS	<i>Thermoanaerobacter tengcongensis</i>	2'-amino-A ₋₁ , pre-cleavage	9
2GCV	<i>Thermoanaerobacter tengcongensis</i>	Post-cleavage	9
2HO6	<i>Thermoanaerobacter tengcongensis</i>	Post-cleavage, all RNA crystal soaked with GlcN6P	9
2HO7	<i>Thermoanaerobacter tengcongensis</i>	Pre-cleavage, all RNA crystal soaked with Glc6P	9
2HOW	<i>Thermoanaerobacter tengcongensis</i>	2'-amino-A ₋₁ , post-cleavage	9
2H0X	<i>Thermoanaerobacter tengcongensis</i>	2'-amino-A ₋₁ , pre-cleavage	9
2H0Z	<i>Thermoanaerobacter tengcongensis</i>	2'-dA ₋₁ , pre-cleavage, Glc6P bound	9
2NZ4	<i>Bacillus anthracis</i>	2'-oMeA ₋₁ , pre-cleavage, GlcN6P bound	3
2Z74	<i>Thermoanaerobacter tengcongensis</i>	2'-dA ₋₁ , pre-cleavage, GlcN6P bound	4
2Z75	<i>Thermoanaerobacter tengcongensis</i>	2'-dA ₋₁ , pre-cleavage, Glc6P bound	4
3B4A	<i>Thermoanaerobacter tengcongensis</i>	G40A, pre-cleavage, GlcN6P bound	10
3B4B	<i>Thermoanaerobacter tengcongensis</i>	G40A, pre-cleavage, 3'-d-, 2'-5'-linked-A ₋₁ , GlcN6P bound	10
3B4C	<i>Thermoanaerobacter tengcongensis</i>	Pre-cleavage, 3'-d-, 2'-5'-linked-A ₋₁ , GlcN6P bound	10
	<i>Bacillus anthracis</i>	2'-oMeA ₋₁ , Pre-cleavage	*
	<i>Bacillus anthracis</i>	Post-cleavage	*
	<i>Bacillus anthracis</i>	G33A, pre-cleavage, GlcN6P bound	*
	<i>Bacillus anthracis</i>	2'-oMeA ₋₁ , pre-cleavage, Glc6P bound	*
	<i>Bacillus anthracis</i>	2'-oMeA ₋₁ , pre-cleavage, MaN6P bound	*

* Presented in this work.