RNA-Protein Interaction

AN ANALYSIS WITH RNA OLIGONUCLEOTIDES OF THE RECOGNITION BY \( \alpha \)-SARCIN OF A RIBOSOMAL DOMAIN CRITICAL FOR FUNCTION*

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\( \alpha \)-Sarcin is a cytotoxic protein that inactivates ribosomes by hydrolyzing a single phosphodiester bond on the '3' side of G-4325 in eukaryotic 28 S rRNA. We have examined the requirements for the recognition by \( \alpha \)-sarcin of this domain using a synthetic oligoribonucleotide (35-mer) that reproduces the sequence and, we presume, the secondary structure (a stem, a bulged nucleotide, and a loop) at the site of modification. The wild type structure and a large number of variants were transcribed in vitro from synthetic DNA templates with phage T7 RNA polymerase. Recognition of the substrate is strongly favored by a G at the position that corresponds to 4325. There is an absolute requirement for a helical stem; however, it can be reduced from the 7 base pairs in the natural structure to 3 without loss of specificity. The nature of the base pairs in the stem modifies but does not abolish recognition; whereas, the bulged nucleotide does not contribute to identification. Cleavage is materially affected by altering the nucleotides in the universal sequence surrounding G-4325 and changing the position in the loop of the tetranucleotide GAG(sarcin)A leads to loss of recognition by the toxin. We propose that the \( \alpha \)-sarcin domain RNA participates in elongation factor catalyzed binding of aminoacyl-tRNA and of translocation; that translocation is driven by transitions in the structure of the \( \alpha \)-sarcin domain RNA initiated by the binding of the factors, or the hydrolysis of GTP, or both; and that the toxin inactivates the ribosomes by preventing this transition.

(Received for publication, September 11, 1989)
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RESULTS

We refer to the oligoribonucleotide (the 35-mer in Fig. 1A) that reproduces the α-sarcin domain in 28 S rRNA as wild type. Treatment of the oligomer with α-sarcin in concentrations ranging from 2.94 × 10^{-8} to 2.94 × 10^{-6} M led to the formation of two fragments; one of 21 nucleotides derived from the 5'-side of the substrate and the other of 14 nucleotides from the 3'-end (Fig. 1B and Ref. 16). This is the same specificity as the toxin has with ribosomes (16). The extent of the cleavage of the substrate as a function of α-sarcin concentration was calculated from a densitometric tracing of the radioautograph; the curve (Fig. 1C) closely approximates that observed with ribosomes as substrate. Specific and effective cleavage of the substrate occurred at a concentration of the toxin of 2.94 × 10^{-8} M (Fig. 1B, lane 2); at 2.94 × 10^{-7} M

FIG. 1. The effect of α-sarcin on a synthetic oligoribonucleotide that mimics the toxins domain in 28 S rRNA. The synthetic radioactive α-sarcin domain oligoribonucleotide (35-mer) depicted in A was renatured and then incubated at a concentration of 1.58 μM for 20 min at 20 °C without α-sarcin (lane 1) or with increasing concentrations of the toxin: 2.94 × 10^{-8} M (lane 2); 5.88 × 10^{-8} M (lane 3); 2.94 × 10^{-7} M (lane 4); 2.94 × 10^{-6} M (lane 5). The products of the digestion were separated by electrophoresis in 20% polyacrylamide-urea gels and radioautographs were made (5). The radioactivity in each of the bands (of 35, 21, and 14 nucleotides) was determined for lanes 2–5, and the percentage of the original substrate that was specifically cleaved was plotted in C as a function of the concentration of α-sarcin. The lowest concentration of the toxin (2.94 × 10^{-8} M) was chosen for the determination, since the reaction velocity is closest to linear at low concentrations (14). The amount of the oligonucleotide fragments produced by α-sarcin cleavage was determined from the x-ray film after radioautography with a LKB 2202 Ultrascan Laser Densitometer. The experiment with the wild type 35-mer served as a standard for comparison. The extent of digestion by α-sarcin varied somewhat from experiment to experiment. The estimate of sensitivity is an approximation since the reaction rate in these experiments is pseudo first-order with respect to α-sarcin concentration (22). The symbols are: O—O, wild type 35-mer from A; ●—●, 34-mer lacking the wild type bulged nucleotide at position 6 (cf. Fig. 3). In A, the S and the arrow designate the site of cleavage by α-sarcin. The numbers on the left indicate the number of nucleotides in the RNA. N.B. The shadow bands that form a ladder are oligoribonucleotides formed most probably by degradation perhaps due to radiation damage.

EXPERIMENTAL PROCEDURES

General—The preparation of the RNA oligonucleotides using synthetic DNA templates, phage T7 RNA polymerase, 5'-(α-32P)ATP, and unlabeled GTP, UTP, and CTP, and the purification by polyacrylamide gel electrophoresis of the radioactive oligoribonucleotides were described before (16, 21).

Assay of the Effect of α-Sarcin on Synthetic Oligoribonucleotides—The RNA oligonucleotides (in concentrations ranging from 1.08 to 2.26 μM) were heated at 90 °C for 2 min in 10 mM Tris-HCl, pH 7.6, 50 mM NaCl, and 10 mM MgCl2 and allowed to renature at 0 °C. The concentrations of the substrate oligoribonucleotides varied with the number of labeled adenosines since the amount of radioactivity was kept constant. After renaturation appropriate amounts of α-sarcin, water, buffer (50 mM Tris-HCl, pH 7.6; 250 mM KCl), and EDTA (in an amount sufficient to chelate the magnesium ions in the renaturation buffer) were added to give a final concentration of 15 mM Tris-HCl, pH 7.6, 15 mM NaCl, 50 mM KCl. The concentrations of the substrate RNAs and of α-sarcin (specified in the figure legends) were chosen so as to approximate those that attend on cleavage of 28 S rRNA in intact ribosomes (22). Incubation was for 20 min at 20 °C. The reaction was stopped by addition of an equal volume of 178 mM Tris-HCl, pH 8.3, 178 mM boric acid, 5 mM EDTA, containing 0.05% bromophenol blue and saturated with urea; an aliquot was analyzed by electrophoresis for 3 h at 1.5 kV in 20% polyacrylamide gels containing 7 M urea (16); the oligonucleotides were visualized by autoradiography.

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noacyl-tRNA to ribosomes and EF-2 catalyzed GTP hydrolysis and translocation. These are the partial reactions most adversely affected by α-sarcin (3) and by ricin (18), respectively, and cleavage at the α-sarcin site in Escherichia coli 23 S rRNA interferes solely with the binding of EF-Tu and EF-G (19). Moreover, EF-Tu and EF-G footprint in the α-sarcin/ricin domain (20), EF-Tu protects only four nucleotides in prokaryotic 23 S rRNA against chemical modification and these correspond in eukaryotic 28 S rRNA to A-4324 (ricin), G-4325 (α-sarcin), and G-4319 and A-4324 which latter are also in the universal sequence (20). EF-G also protects only four nucleotides and three are the same as the ones protected by EF-Tu; they are the bases that correspond to G-4319, A-4324, and G-4325 (20).

We have undertaken to determine how α-sarcin recognizes a single phosphodiester bond in a particular domain in rRNA. This is part of an effort, unfortunately but necessarily oblique, to understand how ribosomal proteins which like α-sarcin are small and basic recognize specific sites in rRNA. An RNA oligonucleotide, a 35-mer, was prepared using a synthetic DNA template and phage T7 RNA polymerase (21); the oligoribonucleotide has the sequence and should have the secondary structure (a stem, a bulged nucleotide, and a loop) of the α-sarcin region of 28 S rRNA. This synthetic oligoribonucleotide is cleaved by α-sarcin at a position that corresponds to G-4325, precisely where the toxin hydrolizes the nucleic acid in intact ribosomes (16).

We have now systematically altered nucleotides in the α-sarcin domain RNA. The aim is a more precise definition of the sequence of nucleotides and of the higher order structure that prescribes the binding of the protein to the RNA and allows the catalysis of hydrolysis. The determinants of recognition that we have examined are: 1) nucleotide specificity at the site of covalent modification; 2) the necessity for a bulged nucleotide in the helical stem; 3) the requirement for the stem itself; 4) the influence of context, i.e. the importance of the nucleotides in the universal sequence in the loop in which the α-sarcin guanosine is located; and, finally, 5) the effect of the position in the loop of the tetranucleotide GAG(α-sarcin)A.

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In order to understand how ribosomal proteins which like α-sarcin are nucleoprotein in intact ribosomes (16); the oligonucleotides were visualized by autoradiography.
about 85% of the substrate is cleaved (Fig. 1B, lane 4, and Fig. 1C). At higher concentrations of α-sarcin, 2.94 \times 10^{-6} \text{ M} or greater, cleavages occur at appreciably all the purines and it is no longer possible to distinguish between specific and nonspecific hydrolysis (Fig. 1B, lane 5). The extent of the nonspecific cleavage cannot be accurately evaluated from these gels since only the adenosines in the substrate are radioactive. We do not know what it is that distinguishes the specific from the nonspecific action of α-sarcin.

Position 21 in the wild type substrate (corresponding to G-4325 in 28 S rRNA) was systematically varied. An oligoribonucleotide with a transition of the G to an A was a less suitable substrate for α-sarcin; cleavage was decreased to 35% of the control (Fig. 2B). Transversions of the wild type G to U (Fig. 2C) or to C (Fig. 2D) reduces hydrolysis to approximately 17 and to 1%, respectively. (The comparison, by densitometry of the radioautograph, was of the results in the experiments with the lowest concentrations of α-sarcin (5.88 \times 10^{-8} \text{ M}), i.e. of lanes 2 in A and B and of lanes 2 in C and D (Fig. 2). Thus, there is strong but by no means absolute dependence on preservation of the G at the site of covalent modification; the preference is G > A > U >> C. What is notable is that α-sarcin which has been presumed to be a purine-specific nuclease (22) has activity with pyrimidines. The results prejudice one to consider structure rather than sequence as the more important determinant of specificity.

Implicit in the interpretation of these and subsequent experiments is the assumption that the decrease in sensitivity to the hydrolytic effect of α-sarcin is predominantly if not exclusively on \( K_{\text{on}} \) rather than \( K_{\text{cat}} \). We are testing the assumption.

The Requirement for a Bulged Nucleotide in the Stem of the α-Sarcin Domain—The α-sarcin domain RNA has a canonical protein binding structure: a stem, a loop, and a bulged nucleotide. The last occurs in a number of ribosomal protein binding sites, albeit the bulged nucleotide is usually an A rather than the U that is found here (23). We suspected that the bulged U at position 6 in the substrate (position 4310 in 28 S rRNA) is not necessary for α-sarcin action since it does not occur at the comparable site in \( E.\ coli \) 23 S rRNA (24), although the bacteria's ribosomes are sensitive to the toxin (6, 25). Still we tested the possibility directly by synthesizing a variant oligoribonucleotide (a 34-mer) that lacked the bulged U in the stem. Just as we suspected the variant is as sensitive to the toxin as the wild type substrate (Figs. 3 and 1C). Thus, a bulged nucleotide is not required for recognition by α-sarcin.

Witherell and Uhlenbeck (26) found that a bulged adenosine in the bacteriophage Qβ genomic RNA was not necessary either for association with the coat protein.

The Requirement for a Helical Stem for Recognition by α-Sarcin—To test the importance of the stem for α-sarcin action we constructed a linear molecule (35-mer) that retained the 17 nucleotides in the loop, including the universal purine-rich sequence of 14 bases, but with 5′ (11 nucleotides) and 3′ (7 nucleotides) ends altered so that they would not pair. The
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linear molecule migrated slower during electrophoresis in 20% polyacrylamide-urea gels than the wild type oligomer (Fig. 4, compare lanes M and I), which is consistent with the former lacking the organized secondary structure of the latter. α-Sarcin did not cleave the linear substrate specifically; there was no formation of the 21 and 14 nucleotide fragments that mark specific hydrolysis by the toxin (Fig. 4). However, there was nonspecific digestion as is apparent from the progressive disappearance of the radioactive oligoribonucleotide with increasing concentrations of α-sarcin (Fig. 4); indeed, the linear RNA may be more sensitive to this nonspecific effect of the toxin than the one with higher order structure.

Having established that the stem is required, we inquired as to the number of base pairs in the helix that are needed. Successive additional base pairs and the bulged uridine were deleted from the wild type oligoribonucleotide (Fig. 5A). Omission of nucleotides 4, 6, and 34, leaving 6 pairs (Fig. 5B); 4–6 and 34, 35, leaving 5 pairs (Fig. 5C); 4–7 and 33–35, leaving 4 pairs (Fig. 5D); and 4–8 and 32–35, leaving 3 pairs (Fig. 5E), yielded variant oligoribonucleotides all of which are recognized specifically by α-sarcin. It is possible that the helix is necessary only to tether the ends of the loop and in nature may be longer and hence more stable than is required for recognition by α-sarcin; we presume it is neither longer nor more stable than is required for its contribution to the function of this ribosomal domain in protein synthesis.

There is evidence that the nature of the base pairs in a RNA helix, i.e. GC as opposed to AU, can affect the structure of an included loop (27). For this reason we have begun to systematically change those in our wild type substrate (Fig. 6A). The variants we have assessed so far are: 1) CG to AU at positions 11 and 29 at the base of the loop (Fig. 6B); 2) GC to UA at positions 10 and 30 (Fig. 6C); 3) UA to GC at positions 9 and 31 (Fig. 6D); and 4) CG to GU at positions 11 and 29 (Fig. 6E), this last is a change to a noncanonical pair. All are recognized specifically by α-sarcin; however, the change in the second base pair (Fig. 6C) or the substitution of a GU for a GC pair at the base of the loop (Fig. 6E) decreases sensitivity to the toxin. The alterations may affect recognition by modifying the conformation of the loop.

The Requirement for Ribonucleotides for Recognition by α-Sarcin—Synthetic oligodeoxynucleotides corresponding to E.

![Fig. 4. Effect of α-sarcin on a linear oligoribonucleotide (35-mer) having the α-sarcin site sequence.](http://www.jbc.org/Downloaded_from http://www.jbc.org)

![Fig. 5. The number of base pairs in the helical stem required for the recognition by α-sarcin of the toxin domain RNA.](http://www.jbc.org/Downloaded_from http://www.jbc.org)

![Fig. 6. The role of Watson-Crick and non-Watson-Crick base pairs in the helical stem in recognition by α-sarcin of the toxin domain DNA.](http://www.jbc.org/Downloaded_from http://www.jbc.org)
coli tRNA\(^{Phe}\) or tRNA\(^{Lys}\) are recognized by their cognate synthetases (28). This led us to synthesize a DNA oligomer (32-mer) that corresponds to the sequence of the wild type \(\alpha\)-sarcin RNA; in this instance the three 5' guanosines were omitted and deoxythymidine replaced ribouridine. This synthetic oligodeoxynucleotide was not cleaved specifically by \(\alpha\)-sarcin (data not shown). However, there was nonspecific digestion and, as we had observed before (22), the hydrolysis of the DNA required magnesium; nonspecific cleavage of RNA by \(\alpha\)-sarcin on the other hand is inhibited by magnesium (22).

The Effect of the Context of the \(\alpha\)-Sarcin Site Guanosine on Recognition—The purpose was to evaluate the contribution to recognition of the nucleotides in the single-stranded region surrounding the \(\alpha\)-sarcin site guanosine (G-21 in the synthetic oligonucleotide). It was anticipated that alterations in the 5’ adjacent adenosine would not have an appreciable effect since depurination of A-4324 by pretreatment with ricin did not affect subsequent cleavage by \(\alpha\)-sarcin at G-4325 in the same ribosomes (29). Nonetheless, a series of variants were constructed with alterations of the ricin site A to G, U, or C (Fig. 7). As we had expected all were recognized by \(\alpha\)-sarcin with specificity and normal efficiency (Fig. 7).

The context was changed in another way: the tetranucleotide GAG(sarcin)A was left intact for reasons that will be apparent shortly and the remainder of the universal portion of the loop sequence was engineered so it was entirely uridines (Fig. 8A) or in a second variant uridines and guanosines (Fig. 8B). Neither oligonucleotide is a competent substrate for \(\alpha\)-sarcin (Fig. 8). Thus, this part of the context is an essential feature of the recognition of the substrate by \(\alpha\)-sarcin.

The Contribution of the Position of the Loop Tetranucleotide GAGA to Recognition—E. coli ribosomes are not sensitive to ricin (30); the ribosomes are not inactivated and A-2660 in the \(\alpha\)-sarcin/ricin domain is not depurinated (15). However, naked E. coli rRNA is a substrate for the N-glycosidase activity of the toxin (14). The two sites of covalent modification, one each in 16 and 23 S rRNAs, have stems with 7 base pairs and loops that have the sequences GAGA. There are sites with this structure, however, that are not modified by ricin. A comparison of the two subsets, modified and unmodified by ricin, indicated that the tetranucleotide GA(ricin)G(\(\alpha\)-sarcin)A in the loop had to have a particular position with respect to the stem. In 28 S rRNA the ricin-modified adenosine in the loop has an equal number of nucleotides on either side of it and, hence, in two dimensions is centered over the stem.

In agreement with a prediction that comes from this observation, if the tetranucleotide GAGA is moved either four (Fig. 9B) or two (Fig. 9C) positions closer to the 5’ end or two positions (Fig. 9D) closer to the 3’ end the specific response to \(\alpha\)-sarcin is entirely lost; only the nonspecific hydrolytic effect on purines is seen. The results with the variant in which the GAGA is moved two positions closer to the 5’ end (Fig. 9D) are instructive. Recognition of the original guanosine would have yielded fragments of 19 and 16 nucleotides from the 5’ and 3’ sides of the substrate. Oligonucleotides of this length were not found (Fig. 9C). In this variant there is still a guanosine at position 21 as in the wild type substrate (cf. Fig. 9A); nonetheless, there was no cleavage there; i.e. fragments of 21 and 14 nucleotides were not obtained. Thus, recognition is not merely of a guanosine at the correct position
in the loop to the recognition by α-sarcin of the toxin domain in the sequence. The wild type and the mutant oligoribonucleotides in which the tetranucleotide GAGA in the loop was shifted toward the 5' or 3' end (the structures are given) were incubated for 20 min at 20 °C at a concentration of 1.58 μM without (lanes 1) or with α-sarcin: 5.88 × 10⁻⁸ M (lanes 2); 2.94 × 10⁻⁷ M (lanes 3); 5.88 × 10⁻⁶ M (lanes 4); 2.94 × 10⁻⁵ M (lanes 5). The tetranucleotide GAGA is underlined to facilitate appreciation of the change in the structure.

The conclusion from these experiments is that specific recognition of rRNA by α-sarcin requires, in the first instance, a stem and a loop but that a bulged nucleotide is not necessary. There is a strong preference for a guanosine at the site of covalent modification in the loop and the context surrounding this nucleotide, i.e. the 14-base universal sequence, affects binding of α-sarcin and catalysis. The exception is the immediate 5' adenosine which has no influence on identification of the RNA. The stem is absolutely essential; however, only 3 of the 7 base pairs found in 28 S rRNA are needed; the identity of the pairs modifies but does not abolish recognition. It is possible that the helical stem contributes to recognition only indirectly by tethering the ends of the loop and conferring on the latter a specific conformation. Finally, the position of the tetranucleotide GAG(sarcin)A in the loop conditions recognition perhaps directly, perhaps indirectly by altering the context.

Although, the problem has not been addressed directly in these experiments the results have bearing on the important question of the function of the α-sarcin domain in 28 S rRNA. There are earlier observations that are consistent with the proposition that the structure of this region is complex and capable of reversible alterations (13, 16, 31). We cite now others: oligodeoxynucleotides complimentary to the universal sequence in the loop of the α-sarcin domain will not bind to either E. coli (32) or rat ribosomes (3) suspended in buffer, suggesting, but by no means proving, that the structure is not simply single-stranded. Occlusion by ribosomal proteins could account for the failure of the cDNA to bind to the site; however, this seems less likely since α-sarcin and ricin have access to the domain. It is most important in this regard that if ribosomes are catalyzing protein synthesis they will bind the complimentary oligodeoxynucleotide (manifest as sensitivity to ribonuclease H) suggesting a reversible change in the structure of the domain. A similar observation has been made with E. coli ribosomes. Further support for the proposal comes from experiments with inhibitors of protein synthesis (cycloheximide, GMP-PNP, and sparsomycin) in which α-sarcin is used as a probe of structure. These indicate that the α-sarcin domain RNA alternates between open and closed states during translation since ribosomes are sensitive to α-sarcin only when peptidyl-tRNA is in the A-site prior to translocation. The interpretation we favor is that during translocation the loop goes from a more complex structure, possibly one involving a tertiary interaction, to a simple single-stranded one. The process, if it occurs, could provide the motive force for transitions in the structure of ribosomal 60 S subunits or in the relationship of the large to the small subunit and might underlie the movement required for the translocation of peptidyl-tRNA from the A- to the P-site and for the movement of mRNA one codon after each of the reiterative rounds in translation. In this paradigm it is the elongation factors EF-1 and EF-2 which, directly or indirectly (i.e. indirectly through the binding or the hydrolysis of GTP), initiate the reversible transition or switch in rRNA structure that propels translocation. Cleavage at G-4325 in 28 S rRNA by α-sarcin or depurination at A-4324 by ricin might abolish the capacity to reversibly switch structures and account in this way for the catastrophic effect of the toxins on ribosome function.

Acknowledgments—We are grateful to Arlene Timosciek for help in the preparation of the manuscript and to Paul Gardner for the synthesis of oligodeoxynucleotides.

REFERENCES

1. Olson, B. H., and Goerner, G. L. (1965) Appl. Microbiol. 13, 314–318.
2. Olson, B. H., Jennings, J. C., Roga, V., Jusek, A. J., and Schurman, D. M. (1965) Appl. Microbiol. 13, 322–326.
3. Fernandez Puentes, C., and Vazquez, D. (1977) FEBS Lett. 78, 143–146.
4. Conde, F. P., Fernandez Puentes, C., Montero, M. T. V., and Vazquez, D. (1978) FEBS Microbiol. Lett. 4, 349–355.
5. Holcomb, A. N., and Cardillo, E. (1978) Biochem. J. 170, 57–61.
6. Schindler, D. G., and Davies, J. E. (1977) Nucleic Acids Res. 4, 1097–1110.
7. Chan, Y. L., Endo, Y., and Wool, I. G. (1983) J. Biol. Chem. 258, 12768–12770.
8. Wool, I. G. (1986) in Structure, Function, and Genetics of Ribosomal RNAs, ed. M. L. Barany (Cold Spring Harbor, NY: Cold Spring Harbor Laboratory).
9. Endo and I. G. Wool, unpublished data.
10. Y. Endo, unpublished data.
11. C. Merryman and W. E. Hill, personal communication.
The Substrate Specificity of α-Sarcin

somes, (Hardesty, B., and Kramer, G. eds) pp. 391-411, Springer-Verlag, New York Inc., New York

9. Raue, H. A., Klootwijk, J., and Musters, W. (1988) Prog. Biophys. Mol. Biol. 51, 77-129

10. Wool, I. G. (1984) Trends Biochem. Sci 9, 14-17

11. Cahn, F., Schachter, E. M., and Rich, A. (1970) Biochim. Biophys. Acta 209, 512-520

12. White, T. C., Rodenko, G., and Boret, P. (1986) Nucleic Acids Res. 14, 9471-9489

13. Endo, Y., and Tsurugi, K. (1987) J. Biol. Chem. 262, 8128-8130

14. Endo, Y., and Tsurugi, K. (1988) J. Biol. Chem. 263, 8726-8739

15. Endo, Y., Mitsui, K., Motizuki, M., and Tsurugi, K. (1987) J. Biol. Chem. 262, 5908-5912

16. Endo, Y., Chan, Y.-L., Lin, A., Tsurugi, K., and Wool, I. G. (1989) J. Biol. Chem. 263, 7917-7920

17. Stern, S., Powers, T., Changchien, L. M., and Noller, H. F. (1989) Science 244, 753-750

18. Montanaro, L., Sperti, S., Mattioli, A., Testoni, G., and Stirpe, F. (1975) Biochem. J. 146, 127-131

19. Haasner, T.-P., Atmadja, J., and Nierhaus, K. H. (1987) Biochimie 69, 911-923

20. Moazed, D., Robertson, J. M., and Noller, H. F. (1988) Nature 334, 362-364

21. Milligan, J. F., Groebe, D. R., Witherell, G. W., and Uhlenbeck, O. C. (1987) Nucleic Acids Res. 15, 8783-8798

22. Endo, Y., Huber, P. W., and Wool, I. G. (1983) J. Biol. Chem. 258, 2662-2667

23. Peatlie, D. A., Douthwaite, S., Garrett, R. A., and Noller, H. F. (1981) Proc. Natl. Acad. Sci. U. S. A. 78, 7331-7335

24. Noller, H. F. (1984) Annu. Rev. Biochem. 53, 119-162

25. Endo, Y., and Wool, I. G. (1982) J. Biol. Chem. 257, 9054-9060

26. Witherell, G. W., and Uhlenbeck, O. C. (1989) Biochemistry 28, 71-70

27. Seong, B. L., and RajBhandary, U. L. (1987) Proc. Natl. Acad. Sci. U. S. A. 84, 334-338

28. Khan, A. S., and Doe, B. A. (1988) Science 241, 74-79

29. Terao, K., Uchiumi, T., Endo, Y., and Ogata, K. (1988) Eur. J. Biochem. 174, 459-463

30. Gale, E. F., Cundliffe, E., Reynolds, P. E., Richmond, M. H., and Waring, M. J. (1981) in Antibiotic Inhibitors of Ribosome Function, 2nd Ed., pp. 402-547, Wiley-Interscience, New York

31. Walker, T. A., Endo, Y., Wheat, W. H., Wool, I. G., and Pace, N. R. (1983) J. Biol. Chem. 258, 333-338

32. White, G. A., Wood, T., and Hill, W. E. (1988) Nucleic Acids Res. 16, 10817-10831

33. Chan, Y. L., Olvera, J., and Wool, I. G. (1983) Nucleic Acids Res. 11, 7819-7831

34. Gutell, R. R., and Fox, G. E. (1988) Nucleic Acids Res. 16, r75-r269
RNA-protein interaction. An analysis with RNA oligonucleotides of the recognition by alpha-sarcin of a ribosomal domain critical for function. Y Endo, A Glück, Y L Chan, K Tsurugi and I G Wool

*J. Biol. Chem. 1990, 265:2216-2222.*

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