Butler, K. W., Johnson, K. G., & Smith, I. C. P. (1978) *Arch. Biochem. Biophys. 191*, 289–297.
Chang, T. Y., Telakowsky, C., Vanden Heuvel, W., Alberts, A. W., & Vagelos, P. R. (1977) *Proc. Natl. Acad. Sci. U.S.A. 74*, 832–836.
Clark, A. F., & Bloch, K. (1959) *J. Biol. Chem. 234*, 2578–2582.
Dahl, C. E., Dahl, J. S., & Bloch, K. (1980) *Biochemistry* (preceding paper in this issue).
Folch, J., Lees, M., & Sloane-Stanley, G. H. (1957) *J. Biol. Chem. 226*, 497–509.
Huang, S. H. (1977) *Lipids 12*, 348–355.
Lala, A. K., Lin, H. K., & Bloch, K. (1978) *Bioorg. Chem. 7*, 437–445.
Lowry, O. H., Rosenbrough, N. J., Farr, A. B., & Randall, R. J. (1951) *J. Biol. Chem. 193*, 265–275.
Nes, W. R., & McKean, H. L. (1977) in *Biochemistry of Steroids and Other Isopentenoids*, pp 411–533, University Park Press, Baltimore MD.
Odriozola, J. M., Waitzkin, E., Smith, T. L., & Bloch, K. (1978) *Proc. Natl. Acad. Sci. U.S.A. 75*, 4107–4109.
Oldfield, E., & Chapman, D. (1972) *FEBS Lett. 23*, 285–297.
Razin, S., & Rottem S. (1976) in *Biochemical Analysis of Membranes* (Maddy, A. H., Ed.) pp 3–26, Chapman and Hall, London.
Rodwell, A. W. (1971) *J. Gen. Microbiol. 68*, 167–172.
Rodwell, A. W., Peterson, J. E., & Rodwell, E. S. (1970) *Pathog. Mycoplasmas, Ciba Found. Symp.*, 123–144.
Rottem, S., Yashouv, J., Ne'eman, Z., & Razin, S. (1973a) *Biochim. Biophys. Acta 323*, 495–508.
Rottem, S., Cirillo, V. P., DeKruyff, B., Shinitzky, M., & Razin, S. (1973b) *Biochim. Biophys. Acta 323*, 509–519.
Shinitzky, M., & Inbar, M. (1974) *J. Mol. Biol. 85*, 603–615.
Shinitzky, M., & Inbar, M. (1976) *Biochim. Biophys. Acta 433*, 133–149.
Sinensky, M. (1978) *Proc. Natl. Acad. Sci. U.S.A. 75*, 1247–1249.
Yeagle, P. L., Martin, R. B., Lala, A. K., & Bloch, K. (1977) *Proc. Natl. Acad. Sci. U.S.A. 74*, 4924–4926.

Phosphorylation of Nuclear Poly(adenylic acid) Polymerase by Protein Kinase: Mechanism of Enhanced Poly(adenylic acid) Synthesis†

Kathleen M. Rose* and Samson T. Jacob

ABSTRACT: Previous studies have shown that nuclear poly(A) polymerase (EC 2.7.7.19) is a phosphoprotein and that phosphorylation of the enzyme in vitro enhances its activity [Rose, K. M., & Jacob, S. T. (1979) *J. Biol. Chem. 254*, 10256–10261]. The present investigation was undertaken to elucidate the mechanism by which phosphorylation of poly(A) polymerase by exogenous protein kinase in vitro alters its catalytic activity. Phosphorylated poly(A) polymerase has an elevated primer requirement and synthesizes a greater number of poly(A) chains relative to control enzyme. Phosphorylation does not significantly alter the product size. Reactions catalyzed by the activated enzyme display rapid, linear kinetics whereas poly(A) synthesis by the nonphosphorylated enzyme exhibits a lag phase at early time points of the incubation. Over prolonged periods of incubation, the net synthesis of poly(A) remains unaffected by phosphorylation of the enzyme. These results indicate that phosphorylation of poly(A) polymerase in vitro does not alter the extent but augments the rate of poly(A) synthesis as a result of increased affinity of enzyme for its polynucleotide primer. These data suggest that the rate of polyadenylation of mRNA in vivo may be regulated by posttranslational modification of poly(A) polymerase.

The nuclear DNA of eucaryotes is complexed with an array of proteins, both basic (histones) and acidic (nonhistones) in nature. Recent observations suggest a role for histones in packaging the DNA into nucleosome structures [see Kornberg (1977) and Felsenfeld (1978)]. In contrast, the function of most of the nonhistone proteins remains obscure. In spite of this fact, much evidence has been accumulated correlating the phosphorylation of nonhistone proteins with elevated rates of RNA synthesis and increased gene expression in vivo [see Kleinsmith (1974)]. In vitro investigations have indicated that DNA-dependent RNA polymerases I (Hirsch & Martelo, 1976) and II (Kranias et al., 1977) can be phosphorylated with resultant increases in RNA synthesis, suggesting that phosphorylation of these two nonhistone chromatin proteins in vivo may regulate the rate of RNA synthesis.

Over the past several years we have been involved in characterizing the enzyme poly(A) polymerase, EC 2.7.7.19 [see Jacob & Rose (1978)]. In the nucleus, this enzyme is tightly bound to the chromatin matrix (Rose et al., 1977a,b), where it most probably catalyzes the initial addition of poly(A) to the mRNA precursors. Although the exact function of poly(A) has not been completely elucidated, poly(A) addition can apparently modulate gene expression by affecting the processing of the nuclear precursors (Derman & Darnell, 1974), the transport of the mRNA into the cytoplasm (Darnell et al., 1971), and/or the stability of the mRNA (Marbaix et al., 1975; Levy et al., 1975). Recently, we have ascertained that nuclear poly(A) polymerase is a phosphoprotein and that the degree of phosphorylation is under biological control, with the enzyme from a rapidly growing hepatoma more highly phosphorylated than its counterpart from normal liver (Rose & Jacob, 1979). These results have suggested that in vivo phosphorylation of the nonhistone chromatin protein, poly(A) polymerase, may augment gene expression via increased polyadenylation. In the present investigation we have explored the mechanism by which phosphorylation of poly(A) polym-

† From the Department of Pharmacology and Specialized Cancer Research Center, The Milton S. Hershey Medical Center, The Pennsylvania State University, Hershey, Pennsylvania 17033. *Received June 28, 1979.* Supported by U.S. Public Health Service Grant No. CA-25078 from the National Cancer Institute.

0006-2960/80/0419-1472$01.00/0

erase affects poly(A) synthesis in vitro.

Experimental Section

Purification of Poly(A) Polymerase and Protein Kinase. Poly(A) polymerase was solubilized from rat liver nuclei as described by Rose et al. (1976a). The enzyme was purified by successive fractionation on DEAE-Sephadex, phosphocellulose, hydroxylapatite, and QAE-Sephadex columns as detailed by us previously (Rose & Jacob, 1976a).

Protein kinase was obtained during the purification of nuclear poly(A) polymerase from Morris hepatoma 3924A. The kinase, which could be separated from poly(A) polymerase by either phosphocellulose or hydroxylapatite chromatography, was further purified by affinity chromatography on casein–Sepharose (K. M. Rose, unpublished experiments). The specific activity of the kinase preparations ranged from 13 000 to 17 000 pmol of phosphate transferred to casein per mg of protein in 15 min by using [γ-^{32}P]ATP as phosphate donor. This protein kinase, which is cyclic nucleotide independent, can utilize either $MgCl_2$ or $MnCl_2$ as divalent ion.

Assay for Poly(A) Polymerase Activity. Poly(A) synthesis was measured essentially as described earlier (Rose & Jacob, 1976a, 1979). Briefly, reaction mixtures contained 52 mM Tris-HCl (pH 8), 0.4 mM $MnCl_2$, 0.225 mM [^{3}H]ATP (4000 cpm/nmol), 35 mM KCl, 0.5 mM dithiothreitol, 200 μg/mL bovine serum albumin, 1 mM $MgCl_2$, 375 μg/mL poly(A), and 0.5–2 μg of poly(A) polymerase in a total volume of 120 μL. Samples were incubated for 30 min at 37 °C and processed on Whatman DE81 filters as described previously (Rose et al., 1976b).

Determination of Product Size and 3'-OH Termini. Poly(A) synthesizing reactions were carried out as indicated above, increasing the specific acticity of [^{3}H]ATP to 6000 cpm/nmol. Triplicate reactions were pooled, precipitated with trichloroacetic acid, hydrolyzed with 0.3 N KOH, and subjected to thin-layer chromatography in two dimensions on poly(ethylenimine)–cellulose as described previously (Rose & Jacob, 1976b). When A-A-A was used as primer, 100 μg of poly(A) was added as carrier prior to acid precipitation.

Results

Effect of Phosphorylation on the Kinetics of Poly(A) Synthesis. Protein kinase, obtained from Morris hepatoma 3924A, has been shown to transfer a phosphate group from the γ position of ATP to serine or threonine residues of purified nuclear poly(A) polymerase (Rose & Jacob, 1979). The phosphorylated enzyme was activated and this activation was linearly dependent on the amount of protein kinase added to the reaction (Rose & Jacob, 1979). Poly(A) synthesis was carried out under a number of conditions in order to confirm the dependence of enzyme phosphorylation on the presence of protein kinase. As indicated in Table I, experiment 1, poly(A) polymerase activity increased from 66 to 375 units upon addition of protein kinase in the assay. When the protein kinase was added but the $MgCl_2$ was omitted, the poly(A) polymerase activity was ~64% (241 units) of that observed in the presence of both $MgCl_2$ and protein kinase. This partial activation of enzyme is consistent with the ability of the protein kinase to utilize $MnCl_2$ as well as $MgCl_2$; $MnCl_2$ is present in the assay for poly(A) synthesis, but at a suboptimal concentration for the kinase. Since both the protein kinase and poly(A) polymerase have an absolute requirement for ATP, it was impossible to measure the dependence of the activation of poly(A) polymerase by protein kinase on ATP by simply deleting ATP from the poly(A) polymerase assay. Therefore, the effect of preincubation with protein kinase in the presence and absence of ATP was ascertained. As shown in Table I, experiment 2, preincubation in the absence of protein kinase, followed by incubation for poly(A) polymerase activity, in the presence of protein kinase, yielded 400 units of activity. This value was comparable to that obtained in the complete system in experiment 1. Addition of protein kinase during the preincubation resulted in a further stimulation of the rate of poly(A) synthesis (675 units). This stimulation exhibited an absolute dependence on ATP, as only 433 units was observed when the nucleotide was omitted during the preincubation.

Table I: Phosphorylation of Nuclear Poly(A) Polymerase by Protein Kinase[a]

reaction conditions		act. (units/mg of protein)
preincubn	incubn	
expt 1		
no preincubn	minus protein kinase	66
no preincubn	minus $MgCl_2$	241
no preincubn	complete	375
expt 2		
minus protein kinase plus ATP	complete	400
plus protein kinase minus ATP	complete	433
plus protein kinase plus ATP	complete	675

[a] The effect of protein kinase on poly(A) polymerase activity was measured under two sets of conditions. In experiment 1, the effect of protein kinase and $MgCl_2$, added directly to the poly(A) polymerase assay, was determined. "Complete" refers to enzyme activity in the presence of protein kinase [1 μg/μg of poly(A) polymerase] and the cofactors at the concentration described under Experimental Section. Reactions were incubated for 30 min at 37 °C and processed as described under Experimental Section. In experiment 2, poly(A) synthesis was measured after preincubation of poly(A) polymerase with protein kinase; the preincubation mixture contained poly(A) polymerase, 1 mM $MnCl_2$, 12 μM ATP (as indicated), 1 μg of protein kinase per μg of poly(A) polymerase (as indicated), and 0.2 mM dithiothreitol. After 15 min at 30 °C, cofactors and protein kinase were adjusted to the final concentrations of the "complete" system in experiment 1. One unit represents 1 nmol of AMP incorporated into poly(A) in 30 min.

The results presented previously (Rose & Jacob, 1979) and in Table I demonstrate that activation of poly(A) polymerase is mediated through a protein kinase dependent enzyme phosphorylation. In those experiments, poly(A) synthesis was measured at a fixed time (30 min). Poly(A) polymerase assays were carried out for varying times in the presence or absence of exogenous protein kinase by using reaction conditions appropriate for both protein phosphorylation and poly(A) synthesis in order to ascertain whether the stimulation of poly(A) polymerase activity by protein kinase persisted throughout the entire time course of the poly(A) synthesizing reaction. As shown in Figure 1, the synthesis of poly(A) by purified liver nuclear poly(A) polymerase was not linear with time and a lag was observed in the initial phase of the reaction. Such nonlinear kinetics are common for purified poly(A) polymerases [see Jacob & Rose (1978)]. In this case, due to the small quantities of enzyme used, the lag phase was pronounced and poly(A) synthesis was not detected prior to 10 min. However, in the presence of protein kinase, which can phosphorylate poly(A) polymerase under these reaction conditions (Rose & Jacob, 1979), the kinetics of poly(A) synthesis were markedly different. Specifically, the time lag was completely eliminated and poly(A) synthesis was linear with time up to 20 min. Notably, at these early time points, there was a dramatic stimulation of poly(A) polymerase by the protein kinase. By 60 min, however, roughly equivalent amounts of

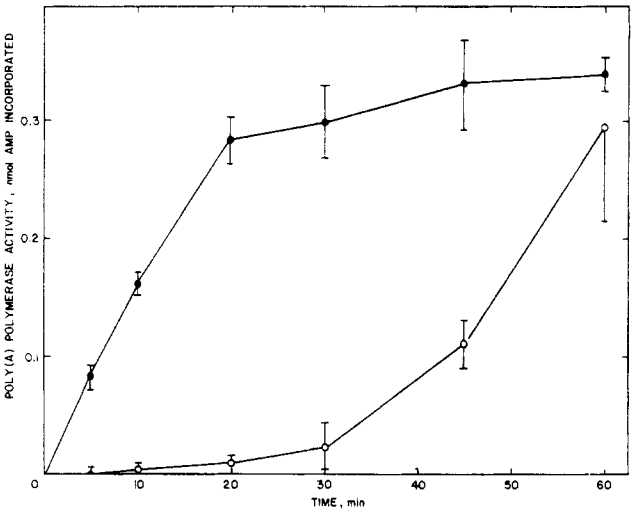

FIGURE 1: Effect of phosphorylation of poly(A) polymerase. Time course of poly(A) synthesis. Poly(A) synthesis was measured as described under Experimental Section by using purified liver nuclear poly(A) polymerase (1.7 μg) in the presence (●) and absence (○) of excess protein kinase. Reactions, terminated at the times indicated, were performed in quadruplicate, and results are expressed as the means ± SEM.

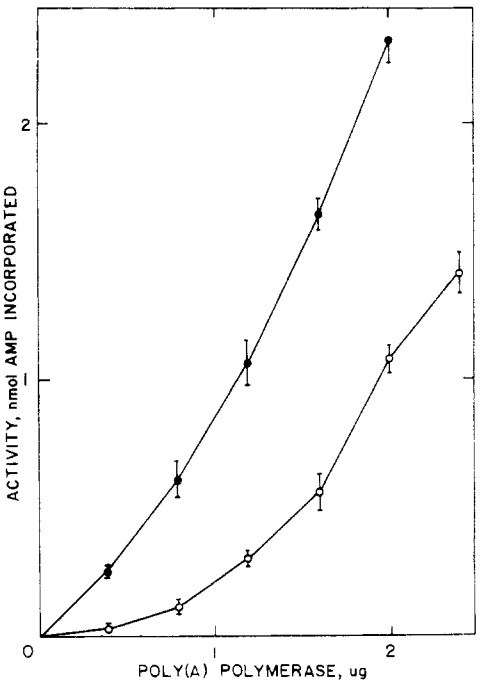

FIGURE 2: Effect of phosphorylation of poly(A) polymerase. Velocity vs. enzyme concentration. Poly(A) polymerase was assayed as described under Experimental Section by using enzyme as indicated in the presence (●) and absence (○) of protein kinase (1 μg). Assays terminated after 30 min at 37 °C were performed in quadruplicate, and results are expressed as the means ± SEM.

poly(A) were synthesized in the presence and absence of the phosphorylating enzyme, indicating that the kinase had little effect on the net quantity of poly(A) synthesized over a prolonged period of time. When poly(A) polymerase was preincubated with protein kinase prior to the poly(A) synthesizing reaction, the rate of poly(A) synthesis was even more accelerated (approximately twice as fast in the first 5–10 min) and reached plateau levels before 20 min (data not shown).

Reactions catalyzed by poly(A) polymerase are generally nonlinear with respect to both time and enzyme concentration (Jacob & Rose 1978). Since phosphorylation of poly(A) polymerase resulted in linear reaction kinetics, it was of interest to determine whether the reaction catalyzed by phosphorylated poly(A) polymerase was also linear with regard to enzyme concentration. For this purpose, enzyme assays were performed with different concentrations of poly(A) polymerase and a fixed amount (excess) of protein kinase. Reactions were terminated after 30 min, at which time the effect of phosphorylation was significant. The results of such an experiment are shown in Figure 2. As expected, poly(A) synthesis in the absence of protein kinase was not linear with increasing amounts of poly(A) polymerase. However, in the presence of the phosphorylating enzyme, product formation was nearly linear with poly(A) polymerase concentration.

Phosphorylation of Poly(A) Polymerase: Effect on Substrate Saturation Curves. The mechanism by which phosphorylation alters poly(A) polymerase activity was further investigated by examining the effect of protein kinase on the saturation curves produced by the two substrates ATP and polynucleotide primer. Figure 3 shows the double-reciprocal plot of velocity vs. ATP concentrations in the presence and absence of protein kinase. It can be seen that both the maximal velocity and the apparent K_m^{ATP} for the poly(A) polymerase reaction were increased in the presence of the phosphorylating enzyme. The apparent V_m for phosphorylated poly(A) polymerase was 5.3 nmol of AMP incorporated per 30 min, a value 2.5 times that of the nonphosphorylated enzyme. The effect of protein kinase on the apparent K_m^{ATP} was less dramatic than on the V_m with the values being 64 μM for the reactions in the presence of the protein kinase and 46 μM for those conducted in the absence of kinase. Since ATP is

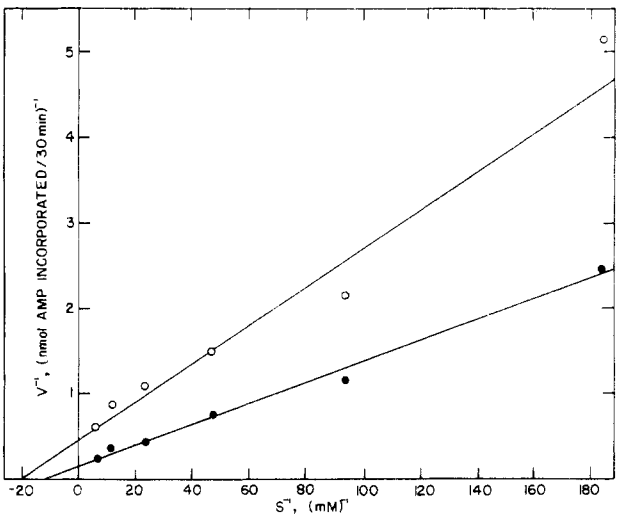

FIGURE 3: Effect of phosphorylation of poly(A) polymerase. Double-reciprocal plot of velocity vs. ATP concentration. Poly(A) polymerase activity was measured under standard assay conditions with the [^{3}H]ATP concentration ranging from 5.2 to 170 μM and corresponding specific radioactivities from 233 000 to 7000 cpm/nmol. Reactions, containing 2 μg of poly(A) polymerase, were conducted in the presence (●) and absence (○) of protein kinase (0.6 μg). Values are the means of quadruplicate determinations. Lines were fitted by linear regression analysis.

also a substrate for protein kinase (with an apparent K_m of 20 μM; K. M. Rose, unpublished experiments), the difference in the K_m^{ATP} values for the poly(A) polymerase reactions cannot be considered significant.

In addition to increasing the rate of poly(A) synthesis, phosphorylation of poly(A) polymerase resulted in an alteration of the saturation curves generated by increasing concentrations of the polynucleotide primer. As shown in Figure 4, in the absence of protein kinase, poly(A) synthesis increased proportionally to the poly(A) concentration, reaching a plateau

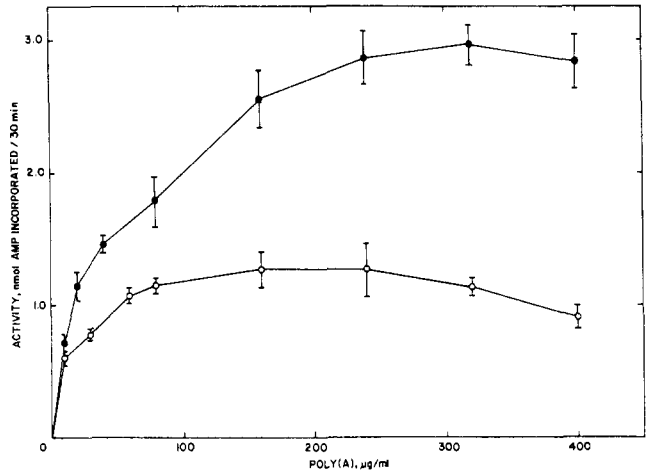

FIGURE 4: Effect of protein kinase on poly(A) synthesis at varying poly(A) concentrations. Poly(A) polymerase activity was measured in the presence (●) and absence (○) of protein kinase as described in the legend to Figure 3 except that ATP was constant (225 μM) and the poly(A) concentration was varied as indicated.

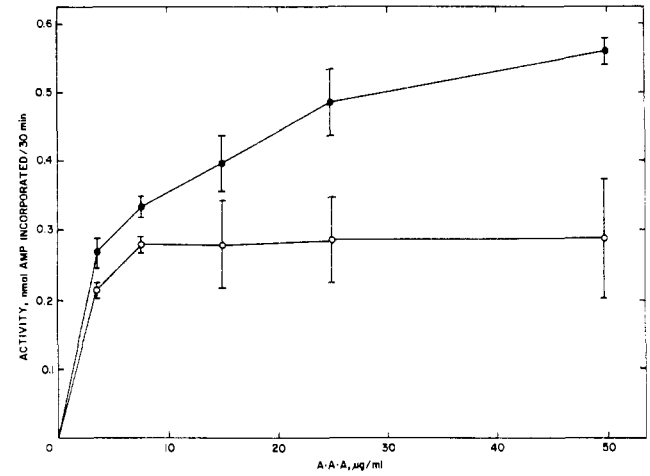

FIGURE 5: Effect of protein kinase on poly(A) synthesis at varying A-A-A concentrations. Poly(A) polymerase activity was measured in the presence (●) and absence (○) of protein kinase as described in the legend to Figure 4 except that A-A-A was substituted for poly(A) at the concentration indicated.

at a polynucleotide level of ~175 μg/mL. In contrast, when the phosphorylating enzyme was present, the plateau region of poly(A) synthesis was attained only at a primer concentration of 325 μg/mL. The increased primer requirement of phosphorylated poly(A) polymerase was confirmed by examining another primer, the trinucleotide A-A-A. As indicated in Figure 5, the poly(A) synthesizing reaction in the absence of kinase was saturated at an A-A-A concentration of less than 10 μg/mL. As was the case for the poly(A)-primed reaction, phosphorylation of poly(A) polymerase resulted in an elevated requirement for this polynucleotide. In fact, poly(A) synthesis by phosphorylated enzyme was not saturated with primer even when the trinucleotide concentration was raised to 50 μg/mL. In addition to increasing the primer requirements for poly(A) synthesis, protein kinase stimulated poly(A) synthesis at all concentrations of the polynucleotide, with a greater degree of stimulation occurring at higher primer concentrations (Table II). For example, when the poly(A) concentration was 10 μg/mL, addition of protein kinase to the poly(A) polymerase reaction resulted in a 1.1-fold stimulation of enzyme activity relative to control. However, when the poly(A) concentration was 400 μg/mL, phosphorylation of poly(A) polymerase caused a 3-fold stimulation of the enzyme activity.

Table II: Effect of Poly(A) Concentration on the Extent of Stimulation of Poly(A) Polymerase by Protein Kinase[a]

poly(A) (μg/mL)	poly(A) polymerase act. [(plus protein kinase)/ (minus protein kinase)]
10	1.11
20	1.50
30	1.68
40	1.80
160	1.92
240	2.21
320	2.62
400	3.14

[a] Some of the data presented in Figure 4 were expressed as the ratio of reactions performed in the presence of protein kinase to those in the absence of phosphorylating enzyme.

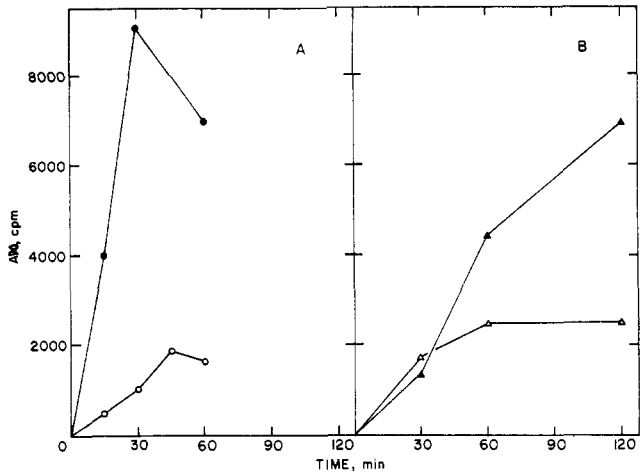

FIGURE 6: Effect of protein kinase on the number of poly(A) chains synthesized by poly(A) polymerase. Poly(A) synthesis was measured by using purified liver poly(A) polymerase and protein kinase [0.5 μg/μg of poly(A) polymerase] as described in the legend to Figure 1 with either 375 μg/mL poly(A) (panel A) or 25 μg/mL A-A-A (panel B) as primer. At the time indicated, reactions were terminated by addition of cold trichloroacetic acid, products were subjected to base hydrolysis, and AMP and Ado were separated by thin-layer chromatography on poly(ethylenimine)–cellulose in two dimensions as described under Experimental Section. The number of 3′-OH termini are represented by cpm recovered in Ado and are expressed per reaction. Open and closed symbols are reactions conducted in the absence and presence of protein kinase, respectively.

Determination of Average Chain Length and 3′-OH Termini. The increased incorporation of AMP into poly(A) by phosphorylated poly(A) polymerase relative to nonphosphorylated enzyme could have resulted either from synthesis of more poly(A) chains of the same length or from production of the same number of poly(A) molecules, but of a larger size. The number of 3′-OH termini and the average chain length of the products were determined for poly(A) polymerase reactions conducted in the presence and absence of the protein kinase in order to differentiate between these two possibilities. As shown in Figure 6, the number of 3′-OH termini (represented by counts per minute in adenosine) was significantly greater in the presence of the phosphorylating enzyme than in its absence. When poly(A) was used as primer (panel A), phosphorylation of poly(A) polymerase resulted in a 5–10-fold increase in [^{3}H]Ado relative to control with the maximum stimulation occurring at 30 min. Incorporation of label at the 3′-OH termini by phosphorylated poly(A) polymerase was also greater when A-A-A was used to prime the reaction (panel B). The time course of the response of the A-A-A primed reaction to protein kinase was somewhat slower than that for the poly(A)-primed reaction, presumably due

Table III: Effect of Phosphorylation of Poly(A) Polymerase on the Average Size of Newly Synthesized Product[a]

time (min)	relative product size [(plus protein kinase)/(minus protein kinase)]
15	0.8
30	0.8
45	0.7
60	1.0

[a] Poly(A) synthesis and separation of AMP and A on poly(ethylenimine)-cellulose were as described in the legend to Figure 6 by using poly(A) as primer. The average chain length of the newly synthesized product was computed from the ratio (cpm of AMP + cpm of Ado)/cpm of Ado. Results are expressed as the average chain length in the presence of protein kinase divided by the chain length in its absence.

to delayed reaction kinetics of the trinucleotide-primed reaction (Rose & Jacob, 1976a). The increased number of poly(A) chains synthesized by phosphorylated enzyme with either primer completely accounted for the stimulation of poly(A) synthesis, since the average chain length (Table III) of the product was not increased by phosphorylation of the enzyme.

Discussion

The present study demonstrates that phosphorylation of poly(A) polymerase markedly increases the rate of poly(A) synthesis. Relative to control enzyme, phosphorylated poly(A) polymerase has an increased primer requirement and synthesizes a greater number of poly(A) chains. These findings suggest that the rate of enzyme–primer complex formation is accelerated by phosphorylation of poly(A) polymerase. The disappearance of the time lag in the poly(A) synthesizing reaction upon phosphorylation of the enzyme indicates that formation of the protein–primer complex is the rate-limiting step in poly(A) synthesis in vitro. Coupled with the observation that neither the net quantity of poly(A) nor its size is increased by phosphorylation of poly(A) polymerase, these data imply that phosphorylation increases the rate, *but not the extent*, of polyadenylation.

The phosphorylation of acidic proteins in the nucleus has long been implicated in regulation of gene expression. It has been observed that proteins bound to the mRNA precursors in the nucleus are also phosphoproteins (Gallinaro-Matringe et al., 1975; Blanchard et al., 1978). As a result of phosphorylation, these proteins appear to have a greater affinity for the RNA to which they bind. Until recently, the biological significance of the mRNA binding proteins was obscure. Our demonstration (Rose et al., 1979) that the poly(A)-specific mRNA binding protein is antigenically related to or, perhaps, identical with poly(A) polymerase itself provided the first evidence for an enzymatic function of one of these binding proteins and has enabled us to interpret the investigations on the poly(A)-specific binding protein in light of its possible role in polyadenylation. Our recent observation that poly(A) polymerase is a phosphoprotein (Rose & Jacob, 1979) and the present demonstration that phosphorylation apparently increases the affinity of enzyme for polynucleotide are concordant with the studies on the effects of phosphorylation on mRNA binding proteins. Thus, it is reasonable to expect that phosphorylation of nuclear poly(A) polymerase in vivo will cause the enzyme to bind more efficiently to the mRNA precursors. Indeed, some evidence to support this hypothesis exists in the case of the rat hepatoma 3924A. Nuclear poly(A) polymerase of this rapidly growing tumor is known to be phosphorylated to a greater extent in vivo than is normal rat liver enzyme (Rose & Jacob, 1979). At the same time, almost 40% of the total nuclear poly(A) polymerase of hepatoma is found tightly complexed with nuclear polynucleotides. This high degree of polynucleotide binding is in contrast with the case of rat liver, where less than 1% of the enzyme is in a similar state (Rose, 1977). These observations, together with the present investigation, indicate that the extent of post-translational modification of poly(A) polymerase is under biological control and that the modified enzyme exhibits an enhanced affinity for its primer molecule. The resultant rapid rate of polyadenylation by phosphorylated poly(A) polymerase would then lead to more efficient mRNA processing and/or a decreased turnover of mRNA, which could serve to augment gene expression posttranscriptionally.

Acknowledgments

The authors thank Ilona Baran for expert technical assistance.

References

Blanchard, J. M., Brunel, C., & Jeanteur, P. (1978) *Eur. J. Biochem. 86*, 301–310.

Darnell, J. E., Philipson, L., Wall, R., & Adesnik, M. (1971) *Science 174*, 507–510.

Derman, E., & Darnell, J. E. (1974) *Cell 3*, 255–264.

Felsenfeld, G. (1978) *Nature (London) 271*, 115–122.

Gallinaro-Matringe, H., Stevenin, J., & Jacob, M. (1975) *Biochemistry 14*, 2547–2554.

Hirsch, J., & Martelo, O. J. (1976) *J. Biol. Chem. 251*, 5408–5413.

Jacob, S. T., & Rose, K. M. (1978) *Methods Cancer Res. 14*, 191–241.

Kleinsmith, L. J. (1974) in *Acidic Proteins of the Nucleus* (Cameron, I. I., & Jeter, J. R., Jr., Eds.) pp 103–135, Academic Press, New York.

Kornberg, R. D. (1977) *Annu. Rev. Biochem. 46*, 931–954.

Kranias, E. G., Schweppe, J. S., & Jungmann, R. A. (1977) *J. Biol. Chem. 252*, 6750–6758.

Levy, G., Smukler, M., Frank, J., Karpetsky, T., Jewett, P., Hieter, P., LeGendre, S., & Dorr, R. (1975) *Nature (London) 256*, 340–341.

Marbaix, G., Huez, G., Burny, A., Cleuter, Y., Hubert, E., Leclercq, M., Chantrenne, H., Soreq, H., Nudel, U., & Littauer, U. (1975) *Proc. Natl. Acad. Sci. U.S.A. 72*, 3065–3067.

Rose, K. M. (1977) Ph.D. Thesis, The Pennsylvania State University.

Rose, K. M., & Jacob, S. T. (1976a) *Eur. J. Biochem. 67*, 11–21.

Rose, K. M., & Jacob, S. T. (1976b) *Biochemistry 15*, 5046–5052.

Rose, K. M., & Jacob, S. T. (1979) *J. Biol. Chem. 254*, 10256–10261.

Rose, K. M., Ruch, P. A., Morris, H. P., & Jacob, S. T. (1976a) *Biochim. Biophys. Acta 432*, 60–72.

Rose, K. M., Lin, Y.-Ch., & Jacob, S. T. (1976b) *FEBS Lett. 67*, 193–197.

Rose, K. M., Bell, L. E., & Jacob, S. T. (1977a) *Nature (London) 267*, 178–180.

Rose, K. M., Roe, F. J., & Jacob, S. T. (1977b) *Biochim. Biophys. Acta 478*, 180–191.

Rose, K. M., Jacob, S. T., & Kumar, A. (1979) *Nature (London) 279*, 260–262.