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Interaction of polynucleotides with natural and model membranes

V.G.Budker, A.A.Godovikov, L.P.Naumova and I.A.Slepneva

Novosibirsk Institute of Organic Chemistry, Siberian Division of Academy of Sciences, Novosibirsk 90, USSR

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ABSTRACT

Polynucleotides adsorb on natural and model phospholipid membranes in the presence of Mg^{2+}-cations. Adsorption of nucleic acids on membranes results in a considerable change of their secondary structure. The presence of model phosphatidylcholine membranes greatly stimulates the rate of the synthesis of RNA by E.coli RNA-polymerase on DNA template.

INTRODUCTION

Association of nucleic acid with membranes is a necessary prerequisite for many biological processes. It is known that replication of DNA in prokaryotic cells proceeds within a membrane complex of DNA[1-3]. Biochemical evidence suggests that association of chromosomes with the nuclear membrane is necessary also for the replication of DNA in eukaryotes[4,5]. Interaction of polynucleotides with membranes may be of importance for transformation and viral infection.

The efforts of the workers who study complexes of nucleoproteins with membranes are applied majorly to functions of membrane proteins. However, there is a considerable bulk of information pointing to a strong effect of lipids upon the state of nucleic acids. For example, equeous-methanol solutions of lipids considerably change the stability of nucleic acid double helices[6-8]. At appropriate concentration they decrease the thermal stability of both free DNA and its complex with histones. Particularly efficient to this end is sphingomyelin. These data strongly suggest that the structure of nucleic acids may be considerably changed within membrane

complexes due to direct interaction with its lipid part.

The present paper reports some new information on the different types of interaction of nucleic acids with membrane phospholipids and on the state of nucleic acids within phospholipid membrane complexes. Preliminary accounts of some of the data reported here have been published earlier[9-11].

MATERIALS AND METHODS

Polyuridilic acid and tRNA (E.coli) were purchased from Special Bureau for Design and Technology of Biologically Active Compounds (Novosibirsk,USSR), [^{14}C]phenylalanine (200 mCi/mmole) - from Chemapol (ČSSR), sodium [^{3}H] borohydride (1000 mCi/mmole) - from Izotop (USSR), [^{3}H] glucose (150 mCi/mmole) - from Amersham (England). Rat liver DNA was a generous gift of Dr.G.M.Dimshits, [^{14}C] DNA from E.coli (6 mCi/mmole) - of Dr. T.E.Vahrusheva. For membrane binding experiments, both DNA preparations were sonicated to fragments of average MW about 500 000. [^{14}C] RNA (8 mCi/mmole) was a transcript of T7 DNA kindly given by Dr. G.L. Dianov. RNA polymerase (E.coli) was a gift of Dr. E.F. Zaichikov, T7 DNA - of Dr. T.G.Maksimova. Heptaadenylate and pentaadenylate have been obtained from polyA as described in ref.[12]. [^{3}H] Heptaadenylate (1.2 mCi/mmole) and [^{3}H] pentaadenylate (2 mCi/mmole) have been obtained by oxidation of the corresponding oligonucleotides by $NaIO_4$ followed by reduction with [^{3}H]$NaBH_4$[13]. Phenylalanyl-tRNA was obtained according to ref.[14]. Mitochondria and mitoplasts of rat liver were isolated as described in ref.[15]; concentration of mitochondrial protein was determined by biuret reaction[16]. Phosphatidylcholine from hen eggs was obtained and purified on aluminium oxide according to ref.[17]; the homogeneity of the lipid was checked by thin layer chromatography on silica gel. Total mitochondrial lipid was obtained as described in ref.[18]. Liposomes were obtained according to ref.[19]; a chloroform solution of lipid was dried under reducted pressure at 45° as a film on the inside of the flask and then hydrated with 2 ml of Tris-HCl (pH 7.4). The mixture was kept under argon and after it was thoroughly mixed in a Vortex mixer, it was sonicated for 4 min at 0° em-

ploying a ultrasound generator USDN (USSR); prior to use, the preparation was centrifuged at 15 000 rpm, and the precipitate discarded. Liposomes loaded with [^{14}C] glucose were obtained by the same method but glucose was added to a concentration 100 000 cpm/ml prior to sonication. Excess glucose was removed by gel filtration on Sepharose 6B.

Binding of radioactive oligo- and polynucleotides with liposomes was performed in 0.002M Tris-HCl pH 7.5 at a concentration of liposomes 0.1 mg lipid per ml. The concentrations of nucleotides varied between 0.1 and 0.01 A_{260} units per ml. The mixture was kept for 10 min at 25° and centrifuged for 3h at 90 000 x g. The radioactivity and the content of lipid in the precipitate were determined.

The effect of polynucleotides upon the turbidity of liposome suspension was studied using a recording spectrophotometer Spekol (GDR). Polynucleotide solution was added to liposome suspension placed into spectrophotometric cell under continuous stirring. The turbidity was estimated by measuring the absorbance at 520 nm in 2-4 min after addition of a portion of polynucleotide, in which time change of the absorbance reaches a plateau.

Retention of [^{3}H] glucose by liposomes after various treatments was determined by measuring the radioactivity of the void volume on gel-filtration on Sepharose 6B.

Binding of polynucleotides with mitochondria and mitoplasts was performed in the following medium: 70 mM sucrose, 220 mM D-mannitol, 0.5 mM EDTA, 20 mM sodium succinate, 2.5 mM potassium phosphate, 2 mM HEPES, 25 mM $MgCl_2$, 0.5 mg/ml bovine serum albumine, pH 7.4.

The complex polyU:$(A)_5$ was obtained in 0.01M Tris-HCl pH 7.5 - 0.01M $MgCl_2$ (1 μmole of polyU plus 1.5 μmole [^{3}H] $(A)_5$). In experiments with non-radioactive $(A)_5$, its total amount was 6 μmole. In all the cases the mixtures were supplemented with NaCl to a concentration 0.5M, and the complex polyU:$(A)_5$ puryfied from non-bound $(A)_5$ by gel-filtration on Sephadex G-25. Radioactivity of the complex was measured in dioxane scintillating liquor.

Hydrolysis of native and denatured DNA with S_1-nuclease in the presence of liposomes. Fish sperm DNA was kindly given by Dr. A.A.Sokolenko, S_1-nuclease (EC 3.1.4.21) - by Dr. N.M. Pustoshilova. DNA was denatured for 5 min at 100° and rapidly cooled. Hydrolysis of DNA in the presence of liposomes was performed at 37° in 0.01M acetate buffer pH 4.6 containing 10 mM $MgCl_2$, 1 mM $Zn(CH_3COO)_2$, 1.3 mM native or denatured DNA, 280 activity units of S_1-nuclease and 0 to 70 mM lipid. At time intervals, 0.5 ml aliquots were taken, and treated with 0.5 ml 1M $HClO_4$. Precipitates were removed by centrifugation, and absorbance at 260 nm was measured of the supernatant diluted with methanol (2 ml per 0.5 ml).

Modification of DNA with dimethylsulfate in the presence of liposomes. Modification was performed with: (i) native DNA; (ii) denatured DNA; (iii) DNA in the presence of liposomes. Reaction mixture contained 1 mg/ml DNA,60 mg/ml lipid,0.01M medinal - HCl (pH 7.5), 0.01M $MgCl_2$. 3 μmol [^{3}H] dimethylsulfate (155 mCi/mmol) was added in the first two cases in methanol solution, in the third case liposomes were loaded by the reagent. Reaction mixture was incubated at 30° during 3 hours. After the reactions, DNA was precipitated with ethanol, the precipitate washed with ethanol and ether, and hydrolyzed with 100 μl of 8M $HClO_4$ for 1h at 100°. The solution was neutralized with 8M KOH in the presence of phenolphtaleine and kept overnight at 40°. The centrifuged solutions were analyzed by chromatography on Dowex 50W x 8 (4 ml column) in 0.3M ammonium formate pH 6.35. The absorbance was monitored by a flow microcolorimeter. Identification of the bases was performed by comparison with a chromatogram of authentic samples. The fractions (1 ml) were counted in dioxane scintillation liquor.

Synthesis of RNA on DNA template by means of E.coli RNA-polymerase in the presence of model membranes. The reaction was performed at 37° in 0.025M Tris-HCl pH 7.9 - 10 mM $MgCl_2$- 0.5 mM dithiotreitol - 50 mM NaCl with [^{14}C] UTP (1-2 μCi/ml) and 0.1 mM nucleoside-5'-triphosphates on T7 DNA template (250-500 μg/ml). The concentration of RNA-polymerase was 20 μg/ml, of lipid - 0 to 50 mg/ml (as liposomes). To assay newly synthesized RNA, 50 μl aliquots were applied to Whatman

3MM strips (1x2 cm), the strips washed with 5% trichloroacetic acid (five times) at 0°, with ethanol, ethanol:ether 1:1, ether and counted in toluene scintillation liquor.

Hydrolysis with pancreatic RNase in the presence of membranes was performed in 10 mM Tris-HCl pH 7.5 - 10 mM $MgCl_2$ with rRNA (1 mg/ml). The concentration of pancreatic RNase was 0.2 μg/ml, of lipid - 0 to 40 mg/ml.

Hydrolysis of DNA with pancreatic DNase in the presence of model membranes was performed in 10 mM Tris-HCl pH 7.5 - 10 mM $MgCl_2$ - 10^{-5}M $CaCl_2$ with 13-14 A_{260} units/ml DNA, 1 μg/ml DNase and 0 to 20 mg/ml lipid present as liposomes. Analysis was performed in the same way as with ribonuclease and with S_1-nuclease.

RESULTS

Adsorption of polynucleotides on phosphatidylcholine membranes. Addition of various oligo- and polynucleotides to suspensions of phosphatidylcholine in the presence of 0.01M $MgCl_2$ affords stable complexes of nucleic acids with the vesicles which may be isolated by ultracentrifugation. Such complexes are not formed in the absence of magnesium ions (Table 1).

Table 1. Binding of radioactive compounds by phosphatidylcholine liposomes.

Compound	Binding (mmoles per mole of lipid)		
	Buffer	Buffer + $+10^{-2}$M $MgCl_2$	Buffer + $+10^{-2}$M $MgCl_2$ + +0.5M NaCl
[^{14}C] DNA	0.14	13.2	0.5
[^{14}C] RNA	2.20	28.0	3.5
[^{14}C]-phe-tRNA	1.30	23.8	8.1
[^{3}H] $(pA)_7$	0.28	2.0	0.35
[^{14}C] AMP	0.07	0.1	0.12

Data presented in this Table demonstrate that complexes are formed by both DNA and RNA, even tRNA which has a rigid tertiary structure. As for heptaadenylate, it also forms a complex, but the extent of binding is much smaller. We failed to observe any reliable binding of AMP to membranes. As revealed by equilibrium dialysis, the equilibrium constant for AMP is equal to ca. 0.1M. Monovalent cations inhibit the association of polynucleotides with liposomes (Table 1). Fig.1 shows the dependence of the binding of [^{14}C] DNA with phosphatidylcholine liposomes on the concentration of sodium chloride. It is seen that the extent of binding depends on the ratio of mono- and bivalent cations.

Addition of excess non-radioactive polynucleotide to a preformed complex of liposomes with a radioactive polynucleotide results in a strong decrease of bound radioactivity. Polynucleotides may be displaced from the complexes also by an acidic polysaccharide heparin. Therefore it seems to be most probable that bivalent cations cross-link the phosphate groups of polynucleotides with the phosphate groups of liposome surface. However, more complicated interaction might also be in-

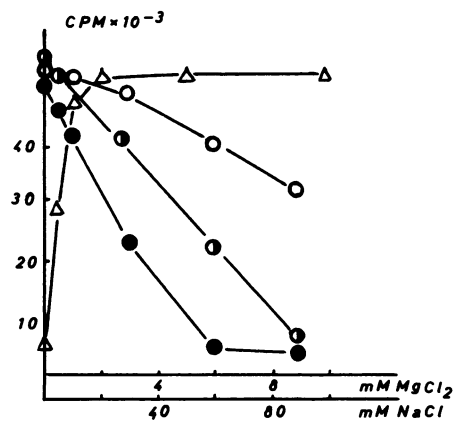

Fig.1. Dependence of the binding of [^{14}C] DNA with liposomes on the concentration of $MgCl_2$ (Δ—Δ) and on the concentration of NaCl at 5 mM (●—●), 10 mM (◑—◑), and 20 mM (○—○) $MgCl_2$.

volved.

State of membrane in the interaction with polynucleotides.

We studied the physical state of liposomes which form complexes with polynucleotides. It appeared that absorption of DNA on liposomes which contained radioactive glucose did not result in leakage of glucose. Subsequent dissociation of the complex caused by addition of EDTA also does not liberate glucose from the internal space of liposomes (Table 2).

Therefore, formation of the complex of liposomes with polynucleotides does not result in disruption or rearrangement of the membranes. However, under appropriate conditions, binding of polynucleotides results in the formation of large aggregates as revealed by increase of turbidity. Fig.2 shows the dependence of the turbidity of liposome suspension on the concentration of some polynucleotides.
Heptaadenylate, tRNA and polyuridylic acid of molecular weight smaller than 20 000 do not change the turbidity.However,polyuridylic acid of molecular weight greater than 300 000 does increase the turbidity. Aggregation of liposomes is induced

Table 2. Retention of [^{3}H] glucose within liposomes subjected to various treatments

Conditions	Radioactivity within liposomes (cpm/5 mg lipid)
Buffer	7880
Buffer + 0.1 mg DNA/ml	8300
Buffer + 10^{-2}M $MgCl_2$, and +1.5×10^{-2}M EDTA in 5 min	6020
Buffer + 0.1 mg DNA/ml + + 10^{-2}M $MgCl_2$ + 1.5×10^{-2}M EDTA in 5 min	7280
10 min at 65°	790

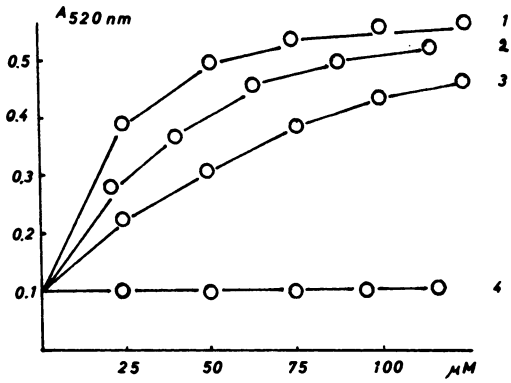

Fig.2. Dependence of the turbidity at 520 nm of phosphatidylcholine liposome suspension on the concentration of different polynucleotides. 1 - DNA; 2 - rRNA; 3 - polyC; 4 - tRNA.

also by heparin. These data are in accord with those of Kim and Nishid[20] who observed formation of insoluble complexes between phosphatidylcholine vesicles and dextrane sulfate in the presence of Ca^{2+}-cations. Aggregation of liposomes in the presence of polynucleotides is reversible. Addition of EDTA results in a rapid change of turbidity to its starting value.

Effect of the length of polynucleotide upon liposomes aggregation may be explained if one assumes that a few liposomes interact with a single polynucleotide molecule. The diameter of liposomes in our experiments is some 300-400 Å, whereas the length of sonicated double-stranded DNA, e.g., is equal to some 1500 Å. If this assumption is correct, aggregation is due to formation of a gigantic network.

Binding of polynucleotides with liposomes obtained from mitochondrial lipids, and with mitochondria.

Bivalent cation-dependent binding of polynucleotides is a general property of phospholipid membranes. Liposomes obtained from unfractionated mitochondrial lipids form a complex with DNA in the presence of Mg^{2+} -ions, and this complex dissociates at a high concentration of NaCl. A similar effect is observed with intact mitochondria (Table 3). It is

Table 3. Binding of radioactive polynucleotides with mitochondria

Additives	Pmoles nucleotide/mg mitochondrial proteins	
	[^{14}C] RNA	[^{14}C] DNA
—	270	700
Sodium azide, 10^{-3}M	260	730
Dinitrophenol, 10^{-5}M	260	870
DNA, 5×10^{-6}M	96	0
Heparin, 0.04 mg/ml	80	112
NaCl, 0.25M	22	90

seen that the binding of polynucleotides by mitochondria does not depend upon their energetic state. Neither oxidative phosphorylation decoupler, dinitrophenol, nor cytochrom C oxidase inhibitor, sodium azide, change the extent of binding of DNA with mitochondria. Heparin, on the other hand, does inhibit the binding. It is noteworthy that the polynucleotides bound by mitochondria are adsorbed on their surface and thus must be available to attack of nucleases. The remarkable similarity of the interaction of polynucleotides with model membranes and with mitochondria suggests that the interaction of natural membranes with nucleic acids generally involves bivalent cation-mediated association with the lipid component.

Change of the state of polynucleotides in the interaction with membranes.

Interaction of the complex polyU:pentaadenylate with phosphatidylcholine membranes. Adsorption of polynucleotides on membranes in the presence of bivalent cations places the polynucleotides on the interface between polar and non-polar phases. Obviously, this must strongly affect the secondary structure of the polynucleotide. There is evidence suggesting that various lipids in aqueous-methanolic solutions strongly decrease the melting temperature of DNA[6-8].

We obtained a complex of polyU with [^{3}H] pentaadenylate (2:1). The melting temperature of this complex is 36°. All the experiments with this complex were made at 4°. Addition of a four-fold excess of non-radioactive pentaA results in a 36% decrease of radioactivity bound by polyU (from 39 000 cpm to 25 000 cpm). Addition of liposomes to the polyU: [^{3}H] pentaA complex in the presence of Mg^{2+} results in its adsorption on the membranes. Subsequent addition of NaCl results in liberation of polyU:[^{3}H] pentaA from liposomes. Such treatment does not decrease the radioactivity of [^{3}H] pentaA bound by polyU (36 850 cpm, 95%). However, if adsorption of the labelled complex on liposomes is performed in the presence of a four-fold excess of non-radioactive pentaA, subsequent desorption results in a decrease of polyU-bound radioactivity to 9 130 cpm (23%) corresponding to complete isotopic dilution. Therefore, binding by liposomes strongly catalyzes the exchange of non-labelled pentaA with [^{3}H] pentaA within the complex polyU:[^{3}H]pentaA.

Modification of DNA with [^{14}C] dimethylsulfate. Dimethylsulfate is widely employed to study the structure of nucleic acids and nucleoproteins. Methylation of single-stranded DNA results in me^1A and me^7G as major products, whereas methylation of double-stranded DNA gives majorly me^7G[21]. The rates of methylation of guanosine in these two states do not differ considerably.

Table 4 shows the results of methylation of T7 DNA under

Table 4. Methylation of native and denatured T7 DNA by dimethyl sulfate and effect of liposomes on methylation of native DNA

DNA	me^1A/me^7G ratio
Native	0.025
Denatured	0.463
Native + liposomes	0.213

various conditions.

It is seen that adenine remains intact in double-stranded DNA. Heat denaturation of DNA results in a dramatic increase of me^1A. We have studied the methylation of DNA adsorbed on phosphatidylcholine membranes. It is noteworthy that dimethylsulfate is present under the conditions mainly within the lipid bilayer, as suggested by gel-filtration of liposomes obtained in the presence of the reagent, as well as by the ninefold decrease of the rate of hydrolysis of dimethylsulfate in the presence of liposomes. Table 4 demonstrates that adsorption of DNA on liposomes results in a considerable increase of the relative reactivity of the position 1 of adenine residues. Hence, binding with liposomes leads to disruption of the hydrogen bonds of the A:T pair.

Hydrolysis of DNA with S_1-nuclease. An important proof of the change of the state of polynucleotides caused by their adsorption on menbranes are the results of hydrolysis of DNA by S_1-nuclease. This enzyme is known to hydrolyze only single--stranded DNA[22]. Fig. 3A shows kinetics of the hydrolysis of double-stranded fish sperm DNA.
It is seen that addition of liposomes results in a considerable increase of the rate of hydrolysis, DNA is adsorbed on the membranes under the conditions. Fig. 3B shows the dependence of the rate of hydrolysis of DNA on the concentration of lipid. At a molar ratio DNA/lipid = 1:50, the rate of hydrolysis increases some 25 times and becomes equal to that of hydrolysis of heat-denatured DNA in the presence of the same concentration of lipid.

Hydrolysis of membrane-adsorbed RNA by pancreatic RNase.

Fig. 4 (curve 1) shows the dependence of the rate of hydrolysis of RNA by pancreatic RNase on the concentration of lipid. It is seen that liposomes inhibit this enzymatic reaction. At a ratio RNA/lipid = 1:20, RNA is protected from hydrolysis. Presumably, this ratio provides complete adsorption of polynucleotide on membrane surface. Inhibition is observed both with double-stranded polynucleotide polyI:

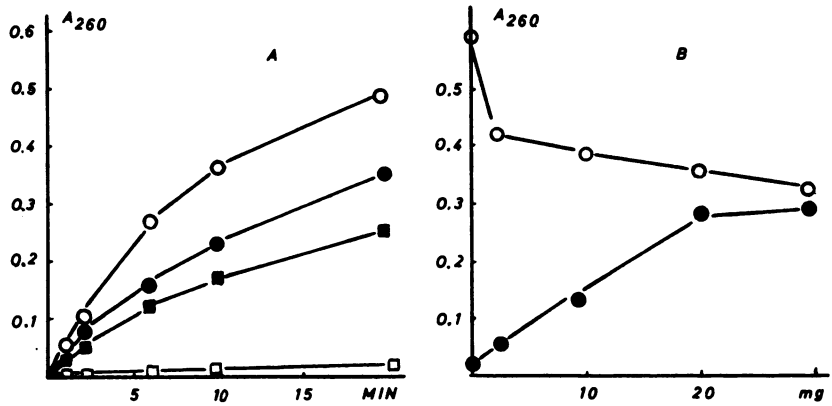

Fig.3. Action of S1-nuclease on DNA. A.Kinetics of hydrolysis of native and denatured fish sperm DNA in the absence of liposomes (□ -- □ and o -- o, respectively), and in the presence of liposomes at a concentration 20 mg lipid per 5 A_{260} units DNA (■ -- ■ and ● -- ● , respectively). B. Dependence of the initial rate of hydrolysis of denatured (o -- o) and native (● -- ●) DNA on the concentration of lipid. Abscissa - absorbance of the nucleotide material soluble in 0.5M $HClO_4$ after incubation of DNA with S1-nuclease during 10 min at 37°. Ordinate - concentration of liposomes, mg lipid per 5 A_{260} units DNA.

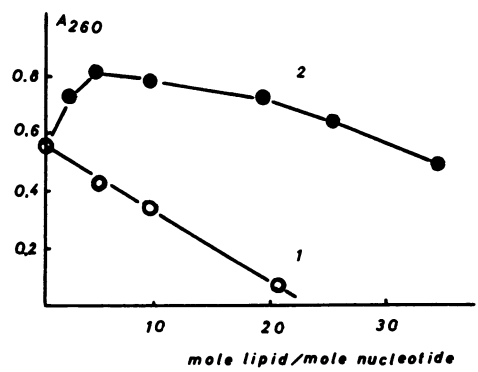

Fig.4. Dependence of the rate of hydrolysis: of rRNA (E.coli) by pancreatic RNase (o—o), and of fish sperm DNA by pancreatic DNase (●—●) on the concentration of liposomes.

polyC, and with singlestranded polyC. Ribonuclease is not bound by membranes under these conditions, as suggested by the following data. The enzyme was mixed with multilayer liposomes (under the polynucleotide-binding conditions), and liposomes were precipitated by 20 min centrifugation at 10 000 g. All RNase remained in the supernatant.

Hydrolysis of membrane-adsorbed DNA by pancreatic DNase.

Quite a different effect is the case with the susceptibility of membrane-bound DNA to pancreatic DNase. The same polynucleotide/lipid ratia as those at which profound inhibition of ribonuclease activity occurs give an increase of the rate of DNase action by a factor of 1.5-2 (Fig. 4, curve 2). Further increase of the concentration of lipid results in slight inhibition. It is noteworthy that DNase is not adsorbed by liposomes.

Effect of lipid membranes on the activity of RNA-polymerase.

Model phosphatidylcholine membranes considerably increase the rate of the synthesis of RNA by RNA-polymerase of E.coli on DNA template (Fig.5). A similar phenomenon has been ob-

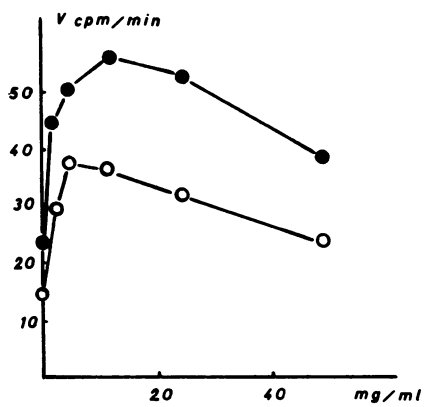

Fig. 5. Dependence of the rate of the synthesis of RNA by E.coli RNA-polymerase on the concentration of liposomes at concentrations of template DNA 5 A_{260} units/ml (o--o) and 10 A_{260} units/ml (● — ●).

served by Lezius and Muller-Lorsen[23] with one of the RNA-polymerases of mouse cells. These workers belived that the increase was due to membrane origin of their enzyme.

In order to find out whether RNA-polymerase of E.coli interacts with membrane, the enzyme was modified by [^{14}C]-iodoacetate to an extent of some 1 mole of carboxymethyl residues per mole of enzyme. A mixture of the radioactive protein with liposomes was centrifuged to sediment liposomes, and radioactivity of the precipitate was measured. Less than 5% of the radioactivity was found in the precipitate. Therefore it seems more likely that the effect of membranes upon RNA-polymerase activity is due to a change of the template properties of DNA. The following evidence is in favour of this explanation: after two-fold increase of the concentration of DNA in the reaction mixture, maximum increase of the rate of RNA-polymerase reaction occurs only when lipid concentration is also increased twice (Fig. 5).

In order to find out which particular stage is activated by membranes, rifampicin was added to the reaction mixture two minutes before the addition of substrates. This treat-

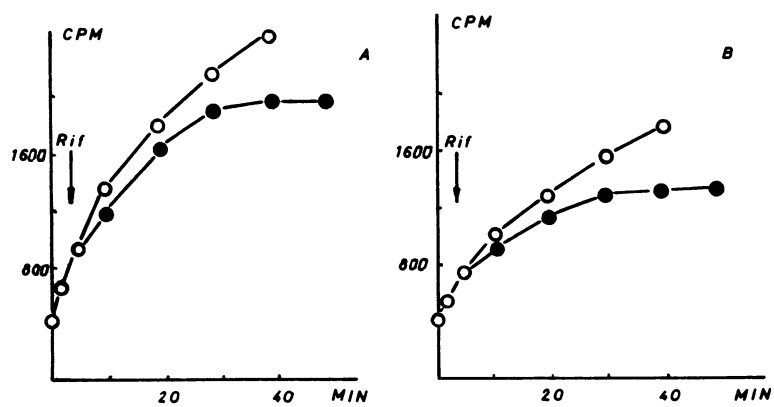

Fig. 6. Kinetics of the synthesis of RNA by E.coli RNA - polymerase in the presence of liposomes 5 mg/ml (A) and in the absence of liposomes (B). The reaction mixture contained 60 μg/ml rifampicin (● -- ●). The reaction mixture did not contain rifampicin (o — o).

ment precludes re-initiation so that the surviving RNA-polymerase activity is only due to elongation. It was found in other experiments that rifampicin inhibits initiation in the presence of liposomes. Fig. 6 shows that the rate of RNA synthesis is some two times greater in the presence of phosphatidylcholine liposomes. Therefore, adsorption of DNA on lipid membranes at least results in an increase of the rate of the elongation stage of RNA synthesis by RNA-polymerase of E.coli. An explanation attractive to us is that the effect is due to the change of the structure of DNA somewhat similar to denaturation. Remarkably, activation of RNA-polymerase reaction is not the case with single-stranded templates like polyuridylic acid (data not shown).

DISCUSSION

There is numerous biochemical and electron-microscopic evidence on complexes of nucleic acids with membranes. However, the attention is paid predominantly to the interaction of nucleic acids with membrane proteins.

The data presented in this paper clearly demonstrate that polynucleotides may also form stable complexes with the lipid component of membranes. The stability of these complexes depends on the ratio of mono- and bivalent cations. The ratio which is typical of living cells (for E.coli, e.g., see ref[25]) is such that all polynucleotides which are not protected by proteins must be adsorbed by the lipid component of membranes. The procedures used for the preparation of electron-microscopic specimens degrade these reversible lipid-nucleic acid complexes, and visualized are normally only the sites of binding of nucleic acids with membrane proteins.

Adsorption of nucleic acids on membranes results in a considerable change of their secondary structure. The simplest term to describe this change is denaturation, as prompted by the above discussed data. However, the real state of nucleic acid within the complex is unclear. Particularly, preliminary data of scanning calorimetry suggest that binding of DNA with liposomes increases its melting temperature some 10° or more

compared with free DNA.

The following model may be proposed. Adsorption of nucleic acid on membrane provides the possibility of interaction of bases with the hydrophobic bilayer. At a certain probability, bases leave double helix and are incorporated into the fatty acid component.

It seems to us likely that adsorption of DNA on the lipid component of biological membranes may facilitate separation of strands of the double helix of DNA during the action of transcription and replication systems. This speculation is supported by the data obtained with RNA-polymerase. Hence, the rate of the synthesis of DNA and RNA may be regulated to some extent by the adsorption of DNA on membranes which depends on the ratio of mono- and bivalent cations.

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REFERENCES

1. Sueoka,N., Quinn,W.,(1968) Cold Spring Harbor Symp.Quant. Biol33,695-706
2. Harmon,J.M., Taber,H.W.(1977) J.Bacteriol.129,789-795
3. Voikov,P.Ya., Shevchenko,N.A.(1977) Molekularnaja Biol. (USSR) 11,766-779
4. Cabradilla,C., Toliver,A.P.(1975) Biochim.Biophys.Acta 402,188-198
5. Infante,A., Firshein,W., Hobart,P., Murray,L.(1976) Biochemistry,15,4810-4817
6. Manzoli,F.A., Muchmore,H.J., Bonora,B., Sabioni,A., Stefoni,S.(1972) Biochim.Biophys.Acta 277,251-255
7. Manzoli,F.A., Muchmore,H.J., Bonora,B., Capitani,S., Bortoli,S.(1974) Biochim.Biophys.Acta 340,1-10
8. Capitani,S., Maraldi,N.M., Santi,P., Martoni,E., Manzoli-Guidotti,H.(1977) Boll.Soc.ital.biol.sper.52,966-972
9. Budker,V.G., Kazachkov,Yu.A., Naumova,L.P.(1978) Biokhimija 43,2105-2107
10. Budker,V.G., Kazachkov,Yu.A., Naumova,L.P.(1978) FEBS Letters 95,143-145
11. Budker,V.G., Naumova,L.P.(1979) Bioorg.Khim.5,135-137
12. Vasilenko,S.K., Serbo,N.A., Venjaminova,A.G., Boldireva, L.G., Budker,V.G., Kobets,N.D.(1976) Biokhimija 41,260-263
13. Randerath,K., Randerath,E.(1969) Anal.Biochem.28,110-113

14.Knorre,D.G., Sirotuk,V.I., Stephanovich,L.E.(1967) Molekul- Biol.(USSR) 1,837-841.
15.Greenavalt,J.M.(1974) Methods in Enzymol.XXXI,310-323
16.Layne,E.(1957) Methods in Enzymol.III,450-451
17.Wells,M.A., Hanahau,D.J.(1969) Biochemistry 8,414-424
18.Dod,B.J., Gay,G.M.(1968) Biochim.Biophys.Acta 150,397-404
19.Kamp,H., Wirtz,K. (1974) Methods in Enzymol.XXXII,140-146
20.Kim,J.,Nishid,T.(1977) J.Biol.Chem.252,1243-1249
21.Mirzabekov,A.D., Melnikova,A.F.(1974) Mol.Biol.Repts.1, 379-384
22.Sutton,W.D.(1971) Biochim.Biophys.Acta 240,522-531
23.Lezius,A., Muller-Lorsen,B.(1972) Hoppe-Seyler's Z.Physiol. Chem.353,1872-1876
24.Damadian,R.(1971) Biophys.J. 11,739-760.