UNFOLDING OF RIBOSOMAL SUBUNITS OF *DICTYOSTELIUM DISCOIDEUM* BY SALTS

Kunihiko ITO

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SUMMARY

The 61-S and 42-S ribosomal subunits of the cellular slime mold, *D.discoideum*, were transformed to slower sedimenting particles under the influence of high concentrations of KCl, NaCl and LiCl. When the ribosomal subunits were treated with various concentrations of KCl under the different conditions, they were converted to a series of particles having the sedimentation coefficients of 50, 46, 38 and 36 S (from the 61-S subunit), and of 38, 29, 25 and 23 S (from the 42-S subunit). The KCl-induced conversion was dependent on the concentration of potassium ion and was partially reversible on addition of magnesium. On treatment of the ribosomal subunit with 0.5 M NaCl or LiCl, the 61-S subunits were converted to 42-S particles and the 42-S subunits to 25-S particles. The KCl-treated ribosomes were more sensitive to the degradation by pancreatic RNase than normal ribosomes. The salt-induced conversion of ribosomes occurred without loss of appreciable ribosomal protein or degradation of ribosomal RNA.

INTRODUCTION

The cellular slime mold, *Dictyostelium discoideum*, has 83-S monomeric ribosomes which are dissociated into 61-S and 42-S ribosomal subunits containing 26-S and 17-S rRNA, respectively.[1,2] Previously we reported[3] that the sedimentation coefficients of ribosomal subunits of this organism were greatly decreased under the influence of 0.5 M KCl, NaCl or LiCl in the presence 0.1 mM Mg^{++}-acetate.

The present paper is concerned with some observations of the ribonucleoprotein particles derived from ribosomal subunits by treatment with KCl, NaCl and LiCl. It will be shown that the ribosomal subunits are converted to particles with lower sedimentation coefficients under the influence of these salts without the dissociation of protein and RNA, and that the KCl-induced conversion is partially reversible on addition of magnesium.

MATERIALS AND METHODS

Culture of organisms and isotope labeling

Amoeba cells of *Dictyostelium discoideum*, strain NC-4 (haploid), were grown in liquid medium containing dead *Escherichia coli* cells as previously described.[4,5]

For labeling RNA or protein of ribosomes, exponentially growing cells were exposed to [^{14}C]uracil (3 μCi/ml of culture), [^{3}H]uridine (30 μCi/ml of culture) or [^{14}C]lysine (3.5 μCi/ml of culture) for 24 h (about four generations). Labeled cells were harvested in a late log-phase of growth, washed four times by centrifugation with cold standard Tris-K^{+}-Mg^{++}

buffer(10mM Tris-HCl(pH 7.6), 25 mM KCl and 0.1 mM Mg^{++}-acetate) to remove dead bacterial cells and stored at -30℃ until use. *E.coli* B cells exponentially grown in minimal Tris-glucose medium[6] were labeled with [^{3}H]uridine (5 μCi/ml of culture) or [^{32}p]orthophosphate (3 μCi/ml of culture) for three generations and then harvested with the above buffer.

Preparation of ribosomal subunits and RNA

For preparation of the 61-S and 42-S ribosomal subunits, the crude extract which had been prepared, as described previously[1,2], with standard Tris-K^+-Mg^{++} buffer from cells labeled with radioactive compounds was centrifuged on a sucrose gradient using an SW25.1 rotor of Spinco L2 ultracentrifuge. After centrifugation, the gradient was fractionated into the appropriate number of tubes and 0.05 ml portion in each tube was taken to determine the radioactive profile. Fractions corresponding to 61-S and 42-S ribosomal subunits were pooled and used as ribosome samples. The same procedure was applied for the preparation of *E.coli* ribosomal subunits.

RNA was extracted by the sodium dodecyl sulfate-phenol method from the pooled fractions of ribouncleoprotein particles isolated by sucrose gradient centrifugation. All operations were done at 2-4℃.

Salt treatment of ribosomes

Ribosomal subunits were incubated with various concentrations of salts in the presence of 0.1 mM Mg^{++}-acetate and 10 mM Tris-HCl (pH 7.6) for 1 h at 0℃. At the end of incubation, the reaction mixture was dialyzed twice against 1000 volumes of standard Tris-K^+-Mg^{++} buffer in the cold for 3 h to remove excess salts and then analyzed by sucrose gradient centrifugation.

Sucrose density-gradient centrifugation

Sucrose density-gradient centrifugation was conducted with an SW 25.1 or SW 50L rotor of a Spinco L2 preparative ultracentrifuge. Appropriate volume of the sample was layered onto the top of a 27 ml (SW 25.1) or 4.7 ml (SW 50L) linear sucrose gradient and centrifuged under the conditions indicated in the legends of the respective Figures. After centrifugation, the tube content was fractionated into 30 to 35 fractions using the apparatus devised by Oumi and Osawa.[7] For the determination of the absorbance profile, two ml of water were added to each fraction and the absorbance of each at 260 mμ was measured.

Measurement of radioactivity

Ribosomal particles or RNA were precipitated in cold 10% trichloroacetic acid-5% acetone. Yeast RNA was added to each fraction as a carrier at the final concentration of about 50 μg/ml before the addition of trichloroacetic acid. The precipitated material was collected on a glass fiber filter (Whatman GF/C) and washed with cold 5% trichloroacetic acid. The filter was dried and then placed into a vial containing toluen based scintillation fluid

for measurement of radioactivity in a Beckman liquid-scintillation spectrometer. The radioactivity in the sample doubly labeled with ^{3}H- and ^{14}C-compounds was counted with an aid of the double setting of ^{3}H and ^{14}C as described previously.[8]

Radioactive compounds

[^{3}H]Uridine (20 Ci/mmole), [^{14}C]uracil (59 mCi/mmole) and L-[^{14}C]lysine (165 mCi/mmole) were obtained from Schwartz BioResearch, Inc.,Orangeburg, N.Y., and [^{32}p]orthophosphate from the Radiochemical Center, Amersham, England.

RESULTS

Sedimentation properties of ribosomal subunits treated with KCl

To examine the KCl-induced modification of the sedimentation properties of ribosomes, 61-S or 42-S ribosomal subunits which had been prepared from cells labeled with [^{14}C] uracil were incubated with various concentrations of KCl in 10mM Tris-HCl(pH 7.6) and 0.1 mM Mg^{++}-acetate for 1 h at 0°C. The incubation mixture was then dialyzed against standard Tris-K^{+}-Mg^{++} buffer and centrifuged on sucrose gradients with [^{3}H]uridine-labeled *E.coli* ribosomes as a marker so that the sedimentation coefficients of KCl-treated ribosomes could be estimated. The sedimentation profiles of 61-S and 42-S ribosomal subunits treated with KCl are shown in Fig.1. When 61-S subunits were treated with 0.15 M KCl (Fig. 1b), they were mostly converted to particles sedimenting at about 46-S.* On treatment with 0.5 M KCl (Fig. 1c), homogeneous particles of approx. 38-S were obtained. When the concentration of KCl was raised to 2 M (Fig. 1d), 61-S subunits were quantitatively converted to particles with the sedimentation coefficient of 36-S. With the 42-S subunit, three discrete particles having the sedimentation coefficients of about 29 S, 25 S and 23 S were produced by treatment of the subunit with 0.15,0.5 and 2 M KCl respectively (Fig. 1f-1h). However, no change in the sedimentation coefficient of ribosomal subunits was observed when the concentration of Mg^{++} was 10 to 50 mM on treatment of ribosomes with 0.5 M KCl.

There are at least three possible explanations for decreasing sedimentation coefficients of ribosomal subunits treated with KCl. One is the breakdown of ribosomes due to partial degradation of rRNA. The second is the release of ribosomal proteins from ribosomes sufficiently to account for the decrease of sedimentation values, as found in *E.coli* ribosomes after treatment with CsCl[10-13] or LiCl.[14,15] The third is the unfolding of ribosomes, as observed with *E.coli* ribosomes by many workers.[16-22] In order to examine these possibilities, the following two experiments were performed.

RNA was extracted from a series of KCl-treated particles derived from [^{14}C]uracil-labeled ribosomal subunits and analyzed by sucrose gradient centrifugation with unlabeled 26-S and 17-S rRNA as a marker. Fig.2 shows the sedimentation profiles of RNA components of the 38-S and 25-S particles obtained respectively from 61-S and 42-S subunits by teatment with 0.5 M KCl. It can be seen that no breakdown of rRNA occurred during the incubation of ribosomes with KCl. The same result was obtained with RNA's from other KCl-treated particles. These results indicate that all KCl-treated particles contain in-

tact rRNA molecules.

Fig. 1.

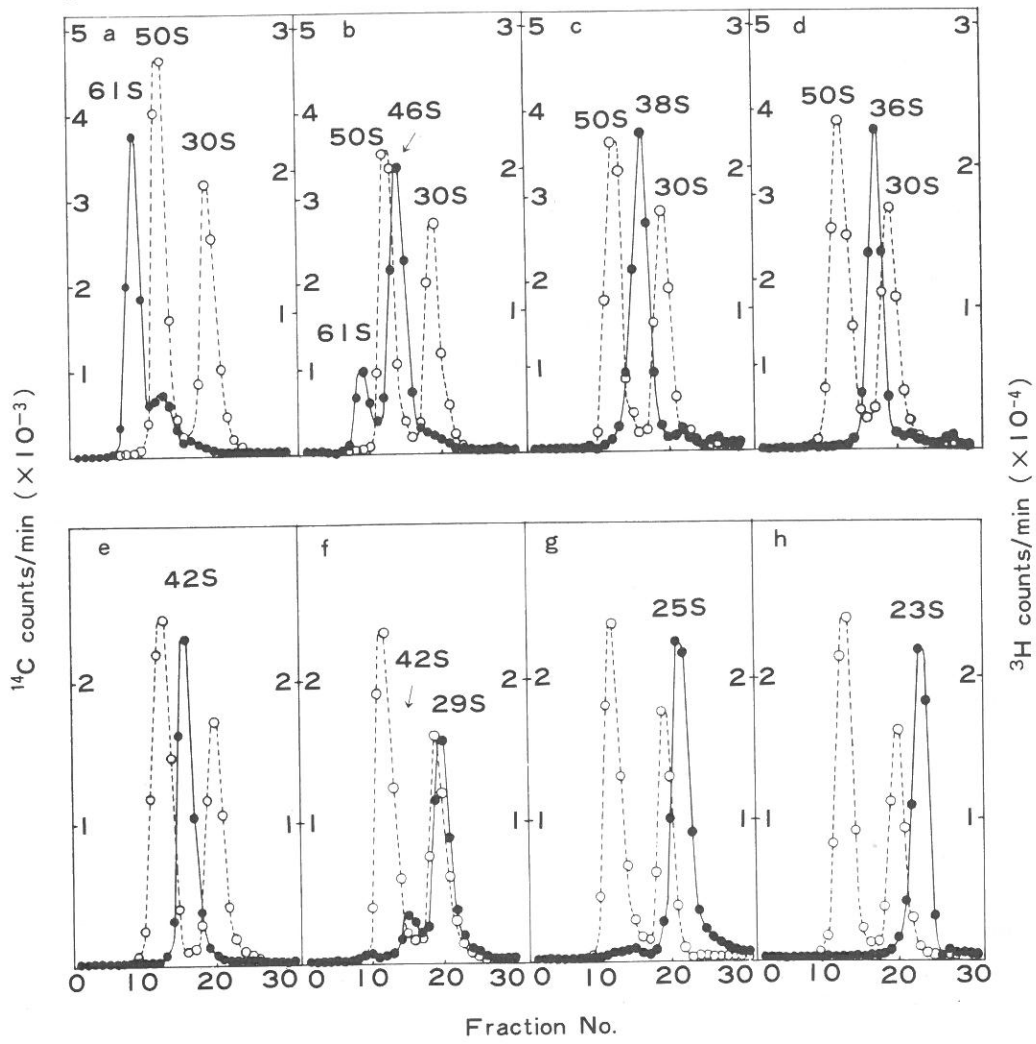

Fig. 1. Sucrose density-gradient centrifugation of 61-S ribosomal subunits (a-d) and 42-S subunits (e-h) after treatment with various concentrations of KCl. [^{14}C]uracil-labeled 61-S or 42-S ribosomal subunits were prepared from exponentially growing cells and incubated with various concentrations of KCl in the presence of 0.1 mM Mg^{++}-acetate and 10 mM Tris-HCl (pH 7.6), as described in Materials and Methods. After incubation for 1 h at 0°, the reaction mixture was dialyzed against Tris-K^{+}-Mg^{++} buffer for 3 h, mixed with [^{3}H]uridine-labeled *E. coli* ribosomes as a marker and centrifuged in a 10-25%(w/v) linear sucrose gradient (27 ml) made with the above buffer using an SW 25.1 rotor. Centrifugation was at 24 500 rev./min for 9.5 h at 4°. The concentrations of KCl in the incubation mixture were : (a) and (e) 0.025 M. (control), (b) and (f) 0.15 M, (c) and (g) 0.5 M, (d) and (h) 2 M. ●—● , ^{14}C; ○···○, ^{3}H.

In another series of experiments, 61-S or 42-S ribosomal subunits which had been prepared from cells doubly labeled with [^{14}C]lysine and [^{3}H]uridine were treated with KCl according to the procedure described in the above and then run on sucrose gradients. As shown in Fig.3, the radioactive peaks of ^{14}C and ^{3}H coincided well with each other. The ^{14}C/^{3}H ratio of a series of KCl-treated particles was nearly equal to that of normal ribosomes (see Table 1). These results suggested that no ribosomal protein was released from the original ribosomes during the KCl-treatment. Thus the above findings demonstrate conclusively that the decrease of the sedimentation coefficient of ribosomal subunits by KCl treatment is mainly due to the unfolding of ribosomes.

Fig. 2.

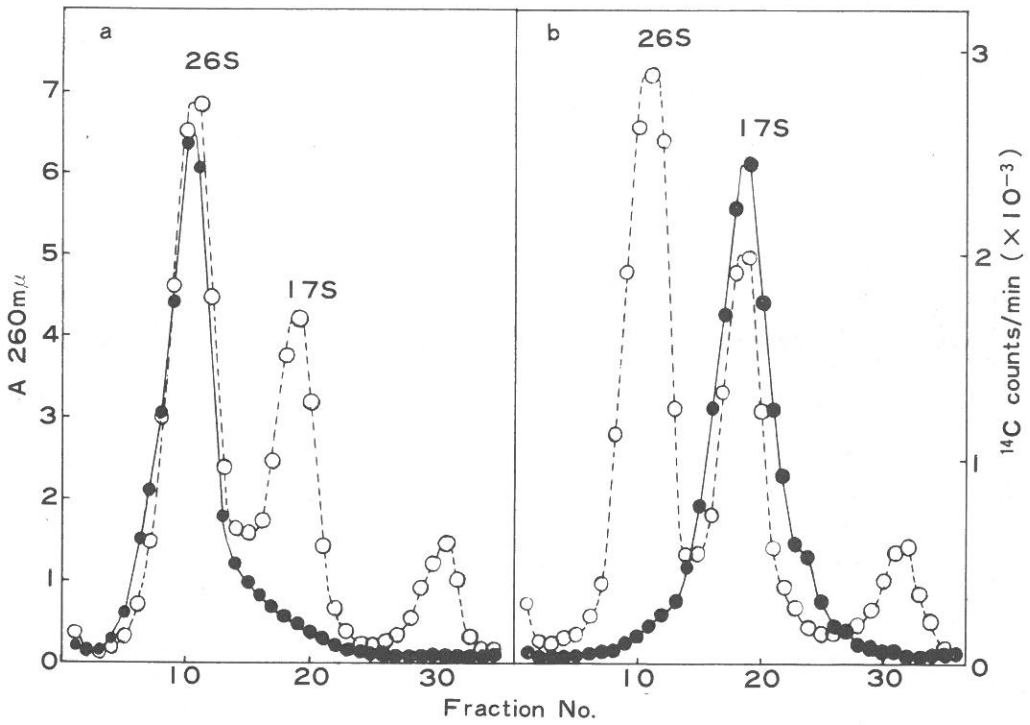

Fig. 2. Sucrose density-gradient centrifugation of RNA prepared from 38-S particles (a) and 25-S particles (b) derived respectively from 61-S and 42-S subunits by KCl treatment. (a) [^{14}C]uracil-labeled RNA from 38-S particles; (b) [^{14}C]uracil-labeled RNA from 25-S particles. RNA sample was mixed with unlabeled 26-S and 17-S rRNA's and then centrifuged on a 5-20% (w/v) linear sucrose gradient (4.7 ml) in 10 mM Tris-HCl (pH 7.2) containing 50 mM NaCl and 10 μg/ml polyvinyl sulfate. Centrifugation was at 39 000 rev./min for 5 h at 6° using an 50L rotor. ●—● , ^{14}C; ○---○ , A260 mμ.

Sensitivity of KCl-treated ribosomes to RNase

The sensitivity of KCl-treated ribosomes to pancreatic RNase(EC 2.7.7.16) was compared with that of normal ribosomes. In this experiment, normal and KCl-treated ribosomal subunits which were labeled with [^{14}C]uracil were incubated with RNase at 23°C. As shown

in Fig.4, the KCl-treated ribosomes were more susceptible to the degradation by RNase than normal ribosomes. The difference in RNase sensitivity among these ribosomal particles is probably ascribed to the conformational change of ribosomes.

Fig. 3.

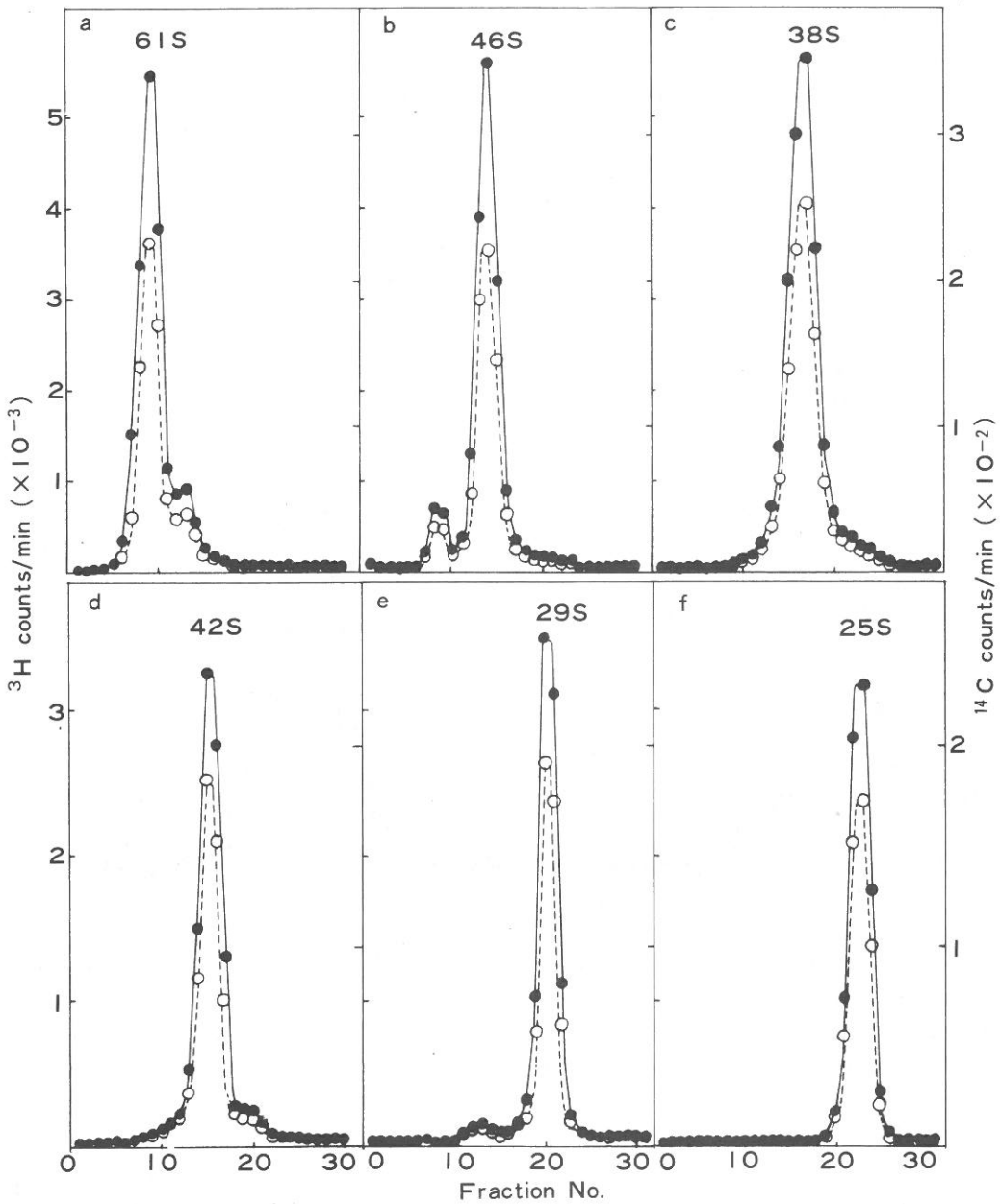

Fig. 3. Sucrose density-gradient centrifugation of normal and KCl-treated ribosomal subunits doubly labeled with [^{14}C]lysine and [^{3}H]uridine. (a) 61-S ribosomal subunits, (b) 46-S particles from 61-S subunits, (c) 38-S particles from 61-S subunits, (d) 42-S ribosomal subunits, (e) 29-S particles from 42-S subunits, (f) 25-S particles from 42-S subunits. The preparation of each particle and the condition for sucrose gradient centrifugation were as for Fig. 1. ●—● , [^{14}C]protein; ○---○ , [^{3}H]RNA.

Table 1. Properties of ribonucleoprotein particles derived from ribosomal subunits of *D. discoideum* after treatment with KCl, NaCl and LiCl.

Kind and concen.(M) of salts		S-value※ of particles obtained	RNA components of particles	Relative value※※ of the ratio of [^{14}C]protein/[^{3}H] RNA in the particles
			61-S subunit series	
KCl	0.025	61	26-S rRNA, 4-5-S RNA[c]	1.00
	0.025	50[a]	26 S	
	0.15	46[b]	26 S	1.04
	0.50	38	26 S	0.97
	2.00	36	26 S	
NaCl	0.50	42	26 S	1.04
LiCl	0.50	42	26 S	1.06
			42-S subunit series	
KCl	0.025	42	17-S rRNA	1.00
	0.025	38[b]	17 S	
	0.15	29	17 S	1.02
	0.50	25	17 S	1.03
	2.00	23	17 S	
NaCl	0.50	25	17 S	0.98
LiCl	0.50	25	17 S	0.96

※ Only s-value of the particle which was predominantly produced is indicated.

※※ Calculated by the ratio of ^{14}C/^{3}H of radioactive peaks shown in Figs. 3 and 7.

a Produced by treatment with 0.025 M KCl in the presence of 0.5 or 1.5 mM Mg^{++} for 80 h at 0°.

b Produced by treatment with 0.025 M KCl in the presence of 0.1 mM Mg^{++} for 80 h at 0°.

c See Fig. 4 in the paper of Iwabuchi et al.[8]

Reversibility of unfolded particles by magnesium

The partial or complete reversion of unfolded *E.coli* ribosomes to the refolding state has been reported by several workers.[16,17,19,20,22] Attempts were then made to examine whether or not unfolded ribosomes produced by KCl treatment can be reversed to folded particles by the addition of magnesium. Unfolded ribosomes were isolated from[^{14}C]uracil-labeled subunits treated with KCl by sucrose gradient centrifugation. The KCl-treated particles so obtained were dialyzed against various concentrations of Mg^{++} in 10 mM Tris-HCl (pH 7.6) and 25 mM KCl for 16 h and the centrifuged on sucrose gradients in the same buffer as used for dialysis with[^{3}H]uridine-labeled 61-S or 42-S subunits as a marker. When the 38-S and 46-S particles derived from 61-S subunits were dialyzed against 1.5 mM (or 5

mM) Mg^{++} (Figs. 5a and 5b), they were reversed respectively to 46-S and 50-S particles, but not to 61-S particles. The 25-S particles from 41-S subunits were reversed to 29-S particles on dialysis against 0.5 mM (or 1.5 mM) Mg^{++} (Fig.5c), but the 29-S particle was unchanged with its sedimentation coefficient even by dialysis against 5 mM Mg^{++}. The above results indicate that the unfolding of ribosomes by KCl is partially reversible on addition of magnesium.

Fig. 4.

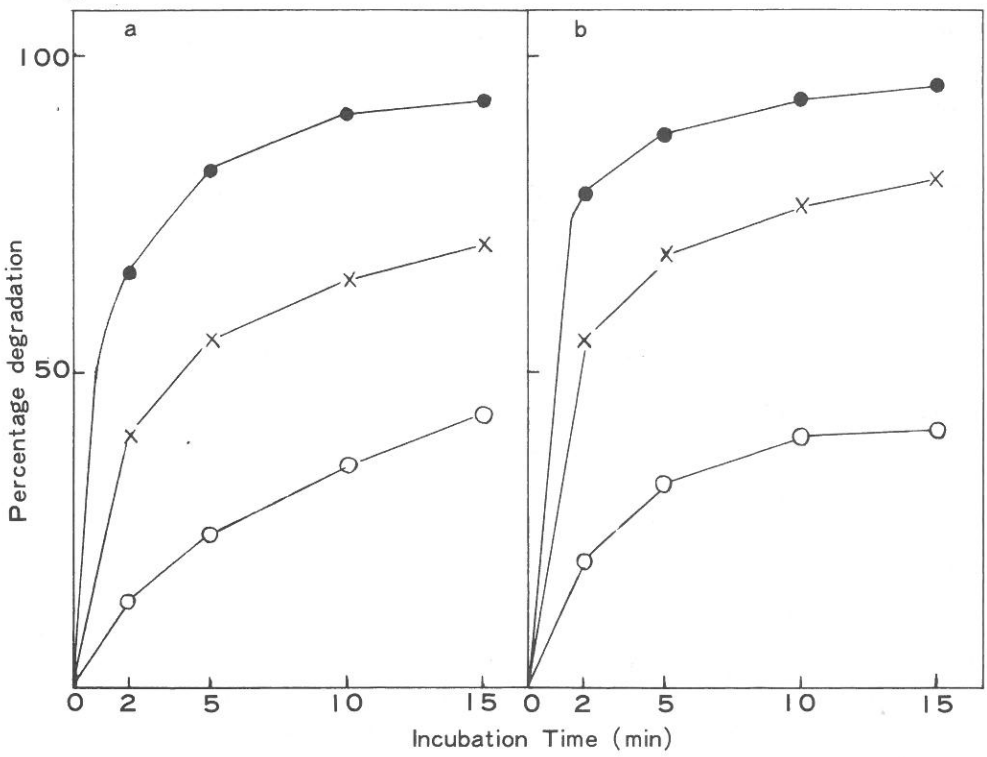

Fig. 4. Effect of RNase on KCl-treated ribosomal subunits. Normal or KCl-treated ribosomal subunits were incubated with pancreatic RNase at 23°. The incubation mixture contained, in 5 ml, 50 μmole Tris-HCl (pH 7.6), 0.5 μmole Mg^{++} -acetate, 125 μmole KCl, 1.1 mg of normal or KCl-treated ribosomes labeled with [^{14}C]uracil, and 1.5 μg of pancreatic RNase (3 × crystallized, from Worthington Biochem.). At indicated times after the addition of the enzyme 0.5 ml samples (with duplicates)were taken and precipitated in cold 10%trichloroacetic acid-5%acetone. Percentage degradation of ribosomal particies was calculated by the amount of ^{14}C-radioactivity of the acid soluble fraction. (a) 61-S subunit series. O—O , Normal ribosomes; ×—× , ribosomes treated with 0.15 M KCl; ●—● , ribosomes treated with 0.5 M KCl.

Unfolding during the storage of ribosomes

In many cases, the 61-S subunits purified by sucrose gradient centrifugation were in

part converted to 50-S particles during recentrifugation on a sucrose gradient in standard Tris-K^+-Mg^{++} buffer(e.g.Fig. la). Then examined whether the sedimentation coefficient decreases during the storage of ribosomes. [^{14}C]Uracil-labeled 61-S or 42-S ribosomal subunits were placed in 10 mM Tris-HCl(pH 7.6)-25 mM KCl containing 1.5, 0.5 or 0.1 mM Mg^{++}-acetate for 80 h at 0°C and then run on sucrose gradients in the same buffer with 61-S or 42-S subunits freshly prepared from cells labeled with [^{3}H]uridine. At 1.5 and 0.5 mM Mg^{++}, 61-S subunits were partially transformed to 50-S particles(Fig.6a and 6b) while at 0.1 mM Mg^{++} they were converted to 46-S paticles (Fig. 6c). The 42-S subunits were converted to 38-S particles when the concentration of Mg^{++} was 0.1 or 0.5 mM(Fig. 6d), whereas at 1.5 mM Mg^{++} the sedimentation coefficient of the subunit was not changed. The sucrose gradient analysis of RNA's prepared from these particles indicated that no fragmentation of rRNA occurred during the storage of ribosomes(data not shown). Thus the unfloding of ribosomal subunits seems to occur during the preservation of ribosomes at low concentrations of magnesium.

Fig. 5.

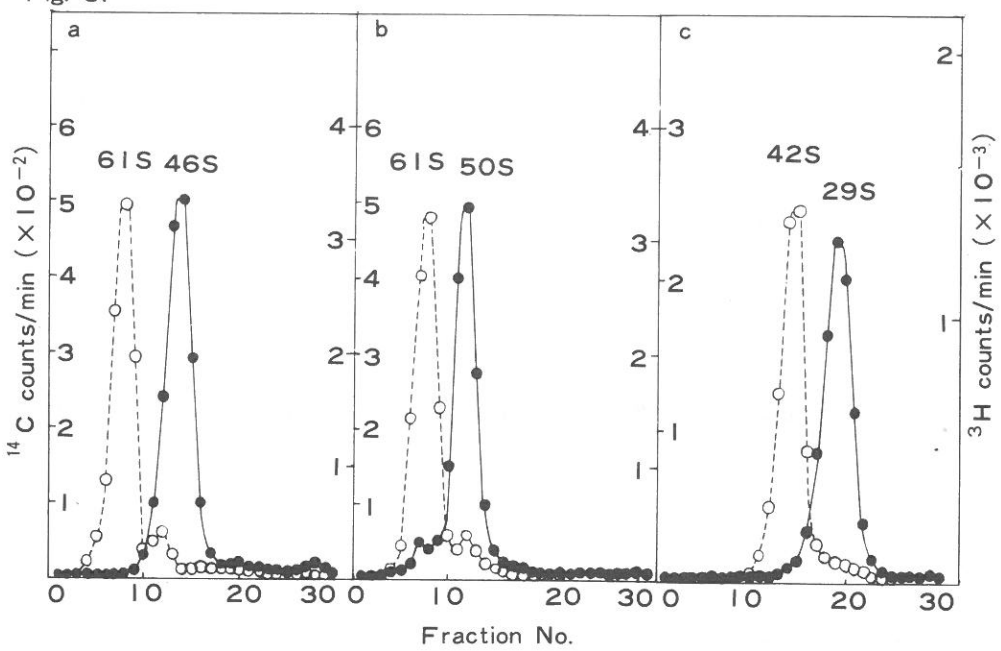

Fig. 5. Sucrose density-gradient centrifugation of ribonucleoprotein particles obtained from KCl-treated ribosomes by dialysis against various concentrations of Mg^{++}. The 38-S and 46-S particles (derived from 61-S subunits) and 25-S particles (from 41-S subunits)were isolated from[^{14}C]uracil-labeled ribosomes treated with KCl by sucrose gradient centrifugation. These particles were dialyzed against 0.5, 1.5 or 5 mM Mg^{++}-acetate in 10 mM Tris-HCl (pH 7.6)-25 mM KCl. After dialysis for 16 h at 2-3°, the diffusate was mixed with [^{3}H]uridine-labeled 61-S or 42-S subunits as a marker and centrifuged on a 10-25%(w/v) sucrose gradient in the above buffer using an SW 25.1 rotor. Centrifugation was at 24 500 rev./min for 9.5 h at 4°. (a) Reconversion of 38-S particles to 46-S particles by dialysis against 1.5 mM (or 5 mM) Mg^{++}, (b) reconversion of 46-S particles to 50-S particles by dialysis against 1.5 mM (or 5 mM) Mg^{++}, (c) reconversion of 25-S particles to 29-S particles by dialysis against 0.5 mM (or 1.5 mM) Mg^{++}. ●—● , ^{14}C; ○----○ , ^{3}H.

Fig. 6.

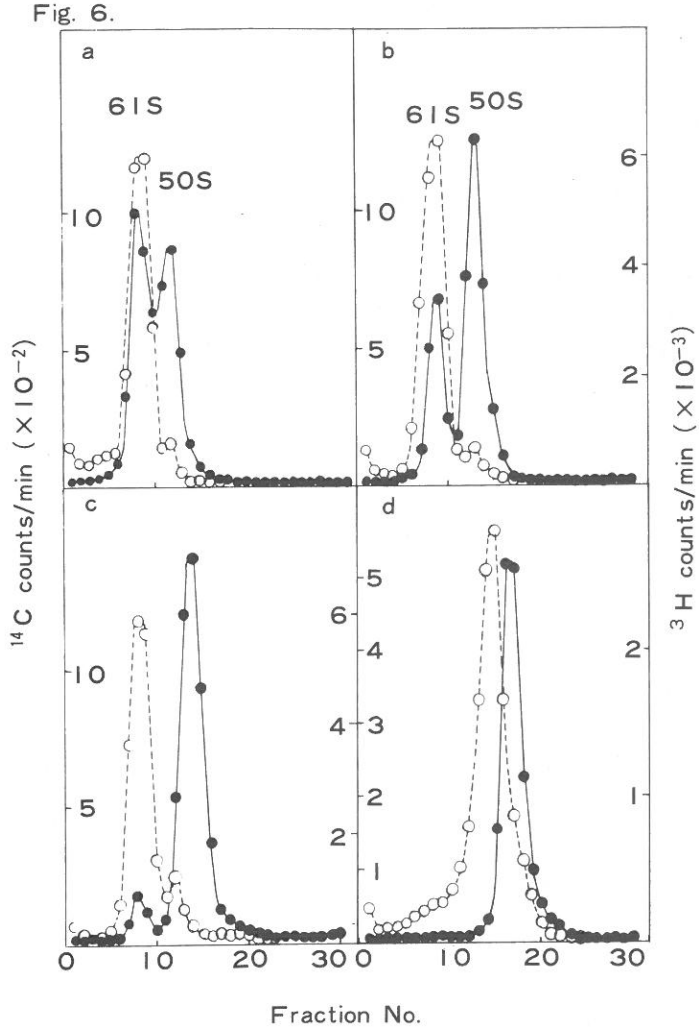

Fig. 6. Changes of the sedimentation coefficients during the storage of ribosomal subunits. [^{14}C] Uracil-labeled ribosomal subunits were stored for 80 h at 0° in 10 mM Tris-HCl (pH 7.6)-25 mM KCl containing Mg^{++}-acetate of 1.5 mM (a), 0.5 mM (b) and 0.1 mM (c and d). Samples were then centrifuged on 10-25% (w/v) sucrose gradients in the same buffer with [^{3}H]uridine-labeled 61-S or 42-S subunits as a marker. Centrifugation was at 24 500 rev./min for 9.5 h at 4° using an SW 25.1 rotor. (a)-(c) 50-S and 46-S particles derived from 61-S subunits, (d) 38-S particles derived from 42-S subunits. ●—● , ^{14}C; ○----○ , ^{3}H.

Unfloding of ribosomes by NaCl and LiCl

To exemine the effect of NaCl or LiCl on the structure of ribosomal subunits, the

mixture of 61-S and 42-S subunits doubly labeled with [^{14}C] lysine and [^{3}H] uridine was incubated with 0.5 M NaCl or LiCl containing 10 mM Tris-HCl (pH 7.6) and 0.1 mM Mg^{++}-acetate. After incubation for 1 h at 0°C, the incubation mixture was dialyzed against standard Tris-K^{+}-Mg^{++} buffer and then centrifuged on a sucrose gradient with [^{32}P]-labeled *E.coli* ribosomes as a refernce. As shown in Fig.7, the mixture of 61-S and 42-S subunits after treatment with NaCl or LiCl was converted to particles sedimenting at approx. 42-S and 25-S. Examination of RNA's extracted from NaCl-treated particles revealed that the 42-S and 25-S particles have 26-S and 17-S rRNA, respectively (Fig.8). The same result was obtained with RNA's from LiCl-treated particles. Therefore, the 61-S subunits would be converted to 42-S particles and the 42-S subunits to 25-S particles. As shown in Fig7 and Table 1, the ratio of ^{14}C/^{3}H in the 42-S or 25-S particles was almost the some to that in the original ribosomal subunits. The above results indicate that the unfolding of ribosomes is induced by NaCl and LiCl.

Fig. 7.

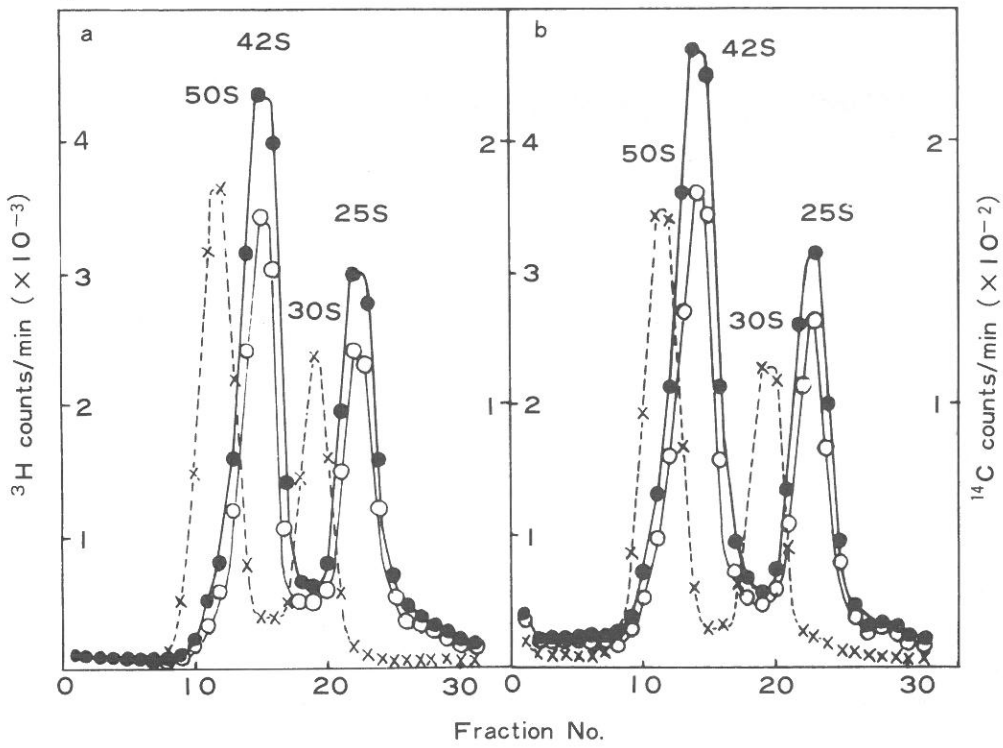

Fig. 7. Sucrose density-gradient centrifugation of ribosomal subunits after treatment with 0.5 M NaCl (a) and 0.5 M LiCl (b). The mixture of 61-S and 42-S subunits of which protein and RNA moities had been labeled respectively with [^{14}C] lysine and [^{3}H] uridine was incubated with 0.5 M NaCl in the presence of 0.1 mM Mg^{++} -acetate and 10 mM Tris-HCl (pH 7.6). After incubation for 1 h at 0°, it was dialyzed against standard Tris-K^{+}-Mg^{++} buffer and then centrifuged on a sucrose gradient with ^{32}P-labeled *E.coli* ribosomes as a reference. The conditions for centrifugation was as for Fig. 1.
●—●, ^{14}C; ○---○, ^{3}H; ×---×, ^{32}P.

Fig. 8.

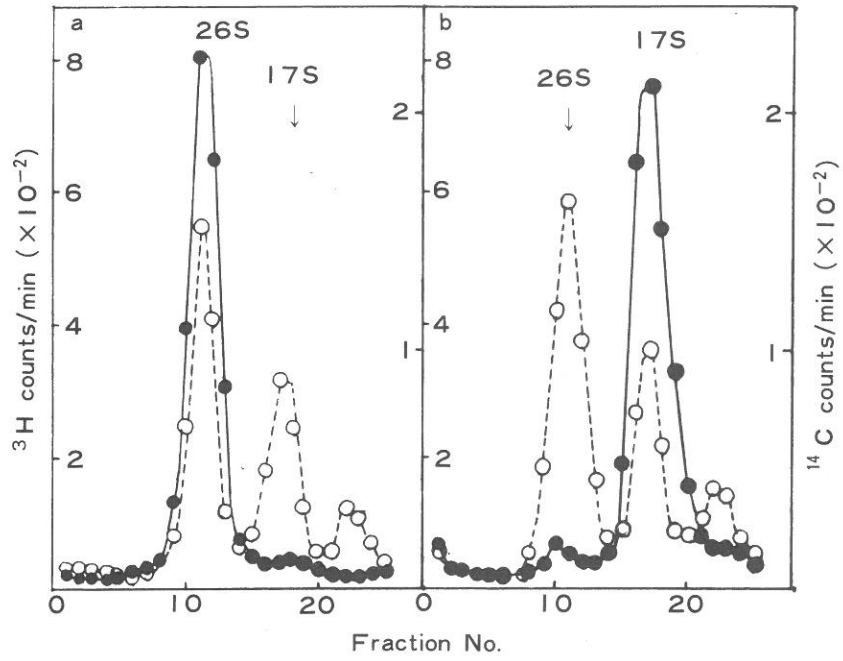

Fig. 8. Sucrose density-gradient centrifugation of RNA prepared from 42-S particles (a) and 25-S particles (b) obtained from the mixture of 61-S and 42-S ribosomal subunits by NaCl treatment. (a) [^{3}H]Uridine-labeled RNA from 42-S particles, (b) [^{3}H]uridine-labeled RNA from 25-S particles. RNA sample was mixed with [^{14}C]uracil-labeled 26-S and 17-S rRNA's and centrifuged on a 5-20% (w/v) sucrose gradient in 10 mM Tris-HCl (pH 7.2) containing 50 mM NaCl and 10 μg/ml polyvinyl sulfate. Centrifugation was at 24 000 rev./min for 14 h at 6° using an SW 25.1 rotor. ●—● , ^{3}H ; O---O , ^{14}C.

DISCUSSION

It has been demonstrated[16-22] that the conformation of *E.coli* ribosomes changes when they were placed in magnesium free buffer or treated with EDTA or with some inorganic salts. The present experiments showed that the 61-S and 42-S ribosomal subunits of *D.discoideum* were converted to particles having lower sedimentatation coefficients under the influence of high concentrations of KCl, NaCl or LiCl. When subunits were treated with various concentrations of KCl., they were converted four discrete particles. The KCl-induced conversion was dependent on the concentration of potassium ion. Since examination of RNA's from salt-treated particles revealed the presence of 26-S or 17-S rRNA of normal size, and since the experiments of double labeling with [^{3}H]uridine and [^{14}C]lysine showed that there was no significant loss of ribosomal protein from these particles, it is reasonable to conclude that the decrease of the sedimentation coefficient of ribosomal sub-

units is due to the conformational change of ribosomes, which has been termed "unfolding".

However, the possibility remains obscure that minor ribosomal proteins which were not detected by double labeling technique were released from ribosomes, and that the removal of small amount of proteins caused marked changes in the sedimentation coefficient.[16, 17, 19, 20, 22]

Several investigators have reported that the unfolding of ribosomes in *E.coli* was completely or partially reversed on addition of magnesium. In this experiments, a partial reversibility of the unfolding process was observed when unfolded ribosomes produced by KCl treatment were dialyzed against high concentrations of magnesium. Thus the KCl-induced conversion of ribosomal subunits of *D.discoideum* may occur as follows:

61-S subunit series: $61S \rightarrow 50S \rightleftarrows 46S \rightleftarrows 38S \rightarrow 36S$

42-S subunit series: $42S \rightarrow 38S \rightarrow 29S \rightleftarrows 25S \rightarrow 23S$

The experiments also showed that NaCl or LiCl induce the unfolding of ribosomes. However,it is not clear why KCl,NaCl and LiCl change the conformation of ribosomes. One possibility is that K^+, Na^+ or Li^+ can displace magnesium in ribosomes, thus resulting in the depletion of magnesium in ribosomes which induce the conformational changes. This assumption may be supported by the experimental results obtained in the previous and present studies;(1) *D.discoideum* ribosomal subunits after treatment with EDTA were transformed to 35-S and 18-S particles,[2] (2) the KCl-induced conversion of ribosomal subunits to slower sedimenting particles did not occur in the presence of high concentrations of magnesium, and (3) the unfolding of ribosomes induced by KCl treatment was reversible, although partially, on the addition of magnesium.

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Footnotes

P. 5.

In this paper, the sedimentation coefficients of ribonucleprotein particles were only approximations estimated from sucrose gradients according to Martin and Ames.[9]

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昭和53年３月29日受理

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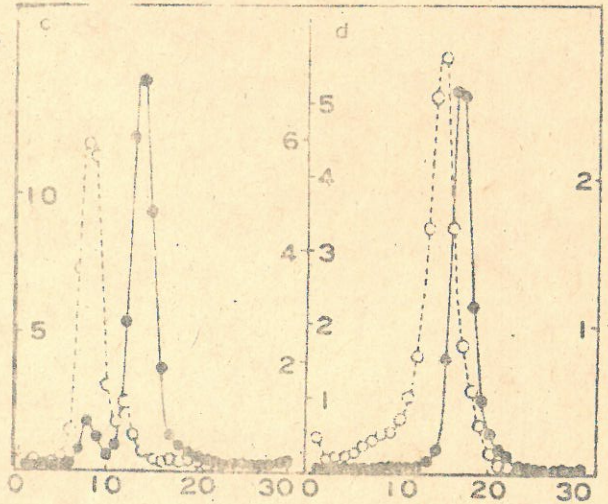
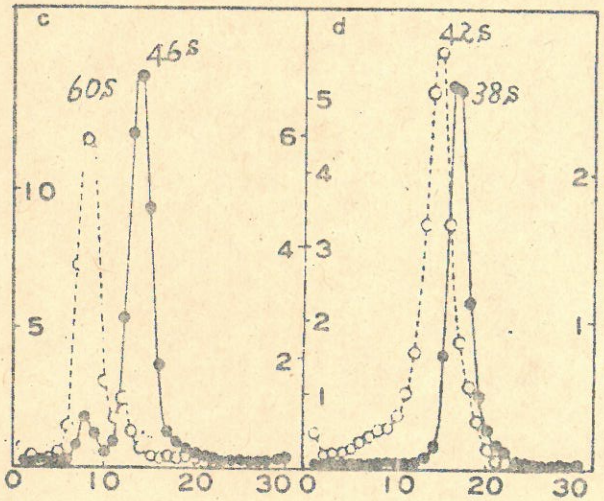
Fig. 6



正

Fig. 6.



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13	下12	coefficients of ribonucleprotein	ribonucleoprotein
20	上19	1951年10月に原則的に	9月
89	上13	Basic P inciples of Nursing	Principles