A potential casein kinase II (CK II) recognition site is located within the conserved carboxyl (COOH) terminus of the ribosomal P (phospho) proteins P0, P1, and P2. To determine whether the COOH termini of the P proteins are physiological substrates for CK II, we studied the phosphorylation of the P proteins in vitro and in intact cells. The results show that the addition of exogenous purified CK II and ATP to intact ribosomes in vitro resulted in the relatively selective phosphorylation of all three P proteins. A synthetic peptide corresponding to the COOH-terminal 22 amino acids of P2 (C-22) was also phosphorylated by CK II with a K_m of 13.4 μM. An endogenous ribosome-associated CK II-like enzyme also phosphorylated the P proteins relatively selectively in the presence of 10 mM Mg^{2+} and ATP. The endogenous kinase was inhibited by heparin, utilized either ATP or GTP as a phosphate donor, and phosphorylated casein.

The eukaryotic P (phospho) proteins PO, P1, and P2 are physiological substrates for CK II, and the (3-22 peptide inhibited the phosphorylation of the P proteins by the endogenous kinase, providing further evidence for its CK II-like properties and for localization of the CK II phosphorylation site to the COOH termini of the P proteins. Tryptic phosphopeptide maps of P1 and P2 phosphorylated by exogenous CK II and the endogenous ribosome-bound kinase were virtually identical. These phosphopeptides comigrated with the tryptic digest of C-22 and with the tryptic phosphopeptides derived from P1 and P2 isolated from intact cells metabolically labeled with [32P]orthophosphate in vivo. These studies demonstrate that exogenous CK II and a ribosome-bound, CK II-like enzyme phosphorylate the ribosomal P proteins in vitro and localize the target site for phosphorylation to the COOH terminus. The incorporation of phosphate into the same target site in intact cells indicates that the P proteins are substrates of CK II.

The eukaryotic P (phospho) proteins P0, P1, and P2 are acidic ribosomal proteins which are located within the 60 S ribosomal subunit (1, 2). Cross-linking studies and their known homology to the Escherichia coli ribosomal protein L7/L12 (3), suggest that one P1 homodimer and one P2 homodimer are attached to P0 via their NH2-terminal ends, while the COOH termini protrude from the ribosomal stalk (4, 5). The COOH-terminal 17 amino acids of P0, P1, and P2 are almost identical except for conservative amino acid substitutions and are highly conserved between species (5-7). Selective elution of P1 and P2 (8, 9) as well as antibody inhibition studies (10-12) suggest that the P proteins play an essential role(s) in protein synthesis. The P proteins are also of interest because approximately 15% of patients with systemic lupus erythematosus (SLE) develop autoantibodies against these ribosomal proteins (13, 14). The levels of anti-P autoantibodies fluctuate in patients with certain neuropsychiatric manifestations of the disease (15). The dominant epitope recognized by SLE autoantibodies has been localized to the common COOH terminus of the P proteins (16), and all sera tested bound to a synthetic peptide comprising the COOH-terminal 22 amino acids (16). The COOH terminus of the P proteins contains a cluster of acidic amino acid residues surrounding potential phosphorylation sites at Ser-102 and Ser-105. These serines are within the sequence Glu-Glu-Ser-Glu-Glu-(Asp)-Glu, which represents a possible recognition site for casein kinase II (CK II) (17, 18). Since phosphorylation could potentially influence the antigenicity (19) and regulate the function (9, 20) of the P proteins, we determined whether the P proteins are substrates of CK II and whether they were phosphorylated by CK II at their COOH termini in vitro and in vivo.

**MATERIALS AND METHODS**

**Preparation of Ribosomes and Isolation of P1 and P2**—Ribosomes were isolated from Hela and Ehrlich ascites cells, rabbit reticulocytes and Artemia salina cysts as described (13, 21). In all cases, ribosomes were washed with buffer containing 50 mM Tris, pH 7.6, 500 mM KCl, 5 mM MgCl2, (Takara), and 250 mM sucrose (22). Total ribosomal protein concentrations were measured by the method of Bradford (23). P1 and P2 were selectively extracted from the ribosomes with TmKmM, and 50% ethanol (9) and were precipitated by the addition of 5 volumes of acetone.

**Synthetic Peptides—Synthesis of the peptides comprising the carboxyl-terminal 22 amino acids of human P2 (C-22 peptide) and Artemia P2 (eL12) was by solid phase methods (16). The peptides were purified by reverse phase HPLC on a Bondapak C18 column.**

**Protein Kinase Assays**—CK II reactions were carried out in a final volume of 30 μl with the indicated concentrations of [γ-32P]-ATP (30-3000 Ci/mmol) or 50 μM [γ-32P]-GTP in TmN10M, buffer, pH 7.6 where N is NaCl) (25). Purified bovine CK II (specific activity 13815

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**To whom reprint requests should be addressed:** The Hospital for Special Surgery, 535 E. 70th St., New York, NY 10021. Tel.: 212-606-1087; Fax: 212-606-1994.

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*The abbreviations used are: SLE, systemic lupus erythematosus; CK II, casein kinase II; SDS, sodium dodecyl sulfate; PAGE, polyacrylamide gel electrophoresis.
approximately 3 μmol/min/mg protein), a kind gift from Dr. D. Litchfield (Howard Hughes Medical Institute, Seattle, WA), was added at a dilution of 1:1000 for reactions with exogenous CK II. Following addition of the substrate (75 μg of ribosomal protein, 15 μg of hydrolyzed and partially dephosphorylated casein (Sigma), and/or varying concentrations of synthetic peptides), samples were incubated at 30°C for 30 min, unless indicated otherwise. Phosphorylation by the catalytic (C-) subunit of the cAMP-dependent protein kinase A (Sigma) was performed under the same conditions as those for CK II.

Antiserum—Human IgG anti-P autoantibodies were detected in the sera of patients with SLE by immunoblotting (26) and enzyme-linked immunosorbent assay using the C-22 synthetic peptide or recombinant P2 fusion protein antigens as described (27).

One- and Two-dimensional SDS-Polyacrylamide Gel Electrophoresis (PAGE) and Immunoblotting—One-dimensional SDS-PAGE was performed on 15% gels essentially as described by Laemmli (28). For two-dimensional gel electrophoresis (29) the first dimension gels contained the ampholytes pH 5-8 and 3-10 (Biozyme, Bio-Rad) in a 4:1 ratio (15). Following SDS-PAGE and elektrophoretic transfer to nitrocellulose paper, immunoblots were sequentially probed with primary (human SLE anti-P sera) and alkaline phosphatase-conjugated secondary antibodies and developed with nitroblue tetrazolium/5-bromo-4-chloro-3-indoly phosphate. Dried gels and blots were exposed to Du Pont Cronex x-ray film with intensifying screens at -70°C. Where appropriate, scanning laser densitometry of autoradiographs was performed.

Immunoprecipitation—IgG from SLE serum containing anti-P antibodies was adsorbed to protein A-Sepharose beads in phosphate-buffered saline (10 mM phosphate buffer, pH 7.4, 150 mM NaCl). The IgG-coated beads were washed and then incubated with ribosomes at room temperature for 2 h. The beads were washed with buffer containing 50 mM Tris, pH 7.5, 150 mM NaCl, 2 mM EDTA, 250 mM sucrose, 2.5% Triton X-100, and 1% SDS and eluted with Laemmli sample buffer.

Cell Culture and [32P]Orthophosphate Labeling—Ehrlich ascites cells (kindly provided by Dr. Francis M. Sirotnak, Memorial Sloan-Kettering Cancer Center, New York, NY) were maintained in Dulbecco's minimal essential/F2 medium supplemented with 10% fetal calf serum and 1% L-glutamine. Prior to labeling in vitro, cells were suspended at 2 x 10^6/ml in phosphate-free medium for 30 min. The medium was removed and the cells were resuspended in 1 ml of the same medium containing 2 μCi of [32P]orthophosphate. After 2 h, the labeling medium was replaced with 1 ml of Tris-KOH-MgCl2 buffer, pH 7.8, and the cells were disrupted by vortexing. The nuclei were pelleted by centrifugation at 8000 x g for 5 min, and the supernatant was used as cytoplasmic extract.

Tryptic Digestion and Two-dimensional Peptide Mapping—P1 and P2 phosphorylated in vitro were purified by preparative SDS-PAGE and electroelution (36). The phosphorylated C-22 peptide was purified by reverse phase HPLC as described above. The P proteins phosphorylated in vitro were isolated by immunoprecipitation from the cytoplasmic extract of Ehrlich ascites cells, SDS-PAGE, and electroelution (36). P1/P2 and the C-22 peptide were digested with trypsin by standard procedures (31). The peptides were first separated by high voltage electrophoresis on cellulose thin layer plates at 800 V for 45 min in 1% NH4HCO3 buffer, pH 8.9 (32), followed by ascending thin layer chromatography in the second dimension using pyridine/n-butyl alcohol/acetic acid/H2O = 60:7:15:60. Tryptic phosphopeptides were located by autoradiography.

RESULTS

Phosphorylation of the P Proteins by Exogenous CK II—Stripped intact ribosomes obtained from Artemia were incubated with exogenous CK II in the presence of [γ-32P]ATP. The ribosomal proteins were then resolved by SDS-PAGE and blotted onto nitrocellulose paper. The autoradiograph of the blot showed incorporation of radiolabel into several bands (Fig. 1A, lane 1). The 36-, 17.5-, and 16-kDa molecular mass phosphorylated proteins were assumed to be the Artemia P proteins P0, P1, and P2. When the same piece of nitrocellulose was probed with an anti-P serum, positive bands with molecular masses identical to the putative P proteins on the autoradiograph were observed (Fig. 1A, lane 2). Similarly, immunoblotting following separation of proteins by two-dimensional gel electrophoresis revealed co-localization of the signals obtained by autoradiography and immunoblotting (Fig. 1, B1 and B2). To determine whether SLE anti-P serum could immunoprecipitate the phosphorylated proteins, Artemia ribosomes were phosphorylated with exogenous CK II and incubated with either anti-P or normal serum. As shown in Fig. 1C, three labeled proteins of 36, 17.5, and 16 kDa were precipitated by the anti-P serum (lane 1) but not by the normal serum (lane 2). Taken together, these results indicate that the three ribosomal P proteins are in vitro substrates of exogenous CK II.

To determine whether the P proteins were specifically phosphorylated by CK II, stripped ribosomes were incubated with either CK II or with the catalytic subunit of protein kinase A. CK II phosphorylated the three P proteins, but only minimally phosphorylated other ribosomal proteins (Fig. 2, lane 1). Protein kinase A, on the other hand, phosphorylated several other ribosomal proteins, but did not incorporate phosphate into the P proteins (Fig. 2, lane 2). This was verified by the failure of SLE anti-P serum to immunoprecipitate the phosphoproteins labeled by protein kinase A. These observations indicate that the ribosomal P proteins are not nonspecifically phosphorylated by multiple kinases.

Ribosomal Protein Kinase Activity—High salt-washed, stripped, Artemia ribosomes were incubated with or without exogenous CK II in the presence of [γ-32P]ATP. Although weak labeling of the P proteins was observed in stripped ribosomes incubated without exogenous CK II (Fig. 3A, lane 2), the addition of exogenous CK II resulted in a significant increase in phosphorylation of P0, P1, and P2 (Fig. 3A, lane 1). When unstripped Ehrlich ascites ribosomes were incubated with [γ-32P]ATP alone (Fig. 3A, lane 3) or with [γ-32P]ATP together with exogenous CK II (Fig. 3A, lane 4), no difference in the level or pattern of phosphorylation of the P proteins was observed, suggesting that the ribosome-bound kinase maximally phosphorylated the P proteins in vivo. In order to determine whether the ribosome-associated kinase had the same properties as those described for purified CK II (33, 34), different substrates (casein, CK II peptide, and calf thymus histone 2A), different ribonucleotides (ATP and GTP) and a selective inhibitor (heparin) of CK II were examined. When unstripped ribosomes were incubated with dephosphorylated casein or histone 2A, extensive phosphorylation of casein was observed (Fig. 3A, lane 5) but no phosphorylation of histone 2A (not shown). Heparin almost completely inhibited the phosphorylation of the P proteins (Fig. 3B, lane 1). GTP substituted for ATP as the phosphate donor and phosphorylation was also inhibited by heparin (not shown).

To determine whether the synthetic CK II-specific peptide Arg-Arg-Arg-Glu-Glu-Thr-Glu-Glu-Glu (24) could compete with the P proteins for phosphorylation by the endogenous CK II-like enzyme, ribosomes were incubated with [γ-32P]ATP in the presence or absence of the peptide competitor. Fig. 4 shows that 1 mM of the peptide almost completely (78%) inhibited phosphorylation of the P proteins by the associated kinase (lanes 1 and 5). The inhibition was concentration-dependent, with 0.1 mM producing only 18% inhibition (lane 4). Inhibition of substrate phosphorylation by the CK II peptide, utilization of both ATP and GTP as phosphate donors and inhibition by heparin indicate that the endogenous kinase responsible for phosphorylation of the P proteins has the same properties as purified CK II (33, 34). The C-22 peptide also competed for phosphorylation of the ribosomal bound P proteins by the endogenous CK II-like enzyme. 1 mM of C-22 completely inhibited phosphorylation (lane 3).
Ribosomal P Proteins are Phosphorylated by Casein Kinase II

FIG. 1. CK II phosphorylates the ribosomal P proteins in vitro. 75 µg of stripped Artemia ribosomes were incubated with exogenous CK II and 100 µM [γ-32P]ATP. The ribosomal proteins were resolved by one- (A) and two-dimensional (B) electrophoresis and then transferred to nitrocellulose paper. Following autoradiography, the same nitrocellulose strips were sequentially probed with

FIG. 2. Purified CK II, but not protein kinase A, phosphorylates the P proteins in vitro. Ribosomes isolated from Hela cells were centrifuged through a discontinuous sucrose gradient (stripped). They were then incubated with 100 µM [γ-32P]ATP and either CK II (lane 1) or protein kinase A (lane 2). Phosphorylated proteins were detected by SDS-PAGE and autoradiography.

FIG. 3. Ribosomes possess a CK II-like activity. A, stripped ribosomes isolated from Artemia were incubated with exogenous CK II (lane 1) or without exogenous CK II (lane 2). Unstripped ribosomes isolated from Ehrlich ascites cells were incubated with exogenous CK II (lane 3), without exogenous CK II (lane 4), or without exogenous CK II but with casein as a substrate (lane 5) in the presence of [γ-32P]ATP. The ribosomal proteins were resolved by SDS-PAGE and subjected to autoradiography. B, effect of heparin on CK II-like activity. Hela ribosomes were incubated with 1.5 µg/ml heparin (lane 1) or with [γ-32P]ATP alone (lane 2).

and 0.1 mM inhibited phosphorylation by 16% (lane 2).

Phosphorylation of the C-22 Peptide—Since the P proteins were phosphorylated by CK II and the predicted CK II site (see Fig. 5) was located within the COOH-terminal 22 amino acids shared by all three P proteins, we attempted to phosphorylate C-22 with CK II in vitro. C-22 and a peptide SLE anti-P antibodies (diluted 1/500) and alkaline phosphatase conjugated anti-human IgG (diluted 1/1000). The labeled ribosomes were also immunoprecipitated with anti-P antibodies, resolved on a 15% SDS polyacrylamide gel, and subjected to autoradiography (C). A, lane 1, autoradiograph; lane 2, immunoblot. B1, autoradiograph; B2, immunoblot. C, immunoprecipitation with anti-P serum (lane 1) and with normal serum (lane 2). The locations of P0, P1, and P2 are indicated, and the positions of molecular weight markers are indicated to the right of C.
Ribosomal P Proteins are Phosphorylated by Casein Kinase II

Fig. 4. Substrate specificity of the ribosome-bound kinase. Ehrlich ascites ribosomes were incubated with 100 μM [γ-32P]ATP alone (lane 1), with 0.1 mM or 1 mM C-22 peptide (lanes 2 and 3), with 0.1 mM or 1 mM CK II-specific peptide (lanes 4 and 5), or with casein (lane 6).

Fig. 5. Location of potential phosphorylation sites in human P2. The amino acid sequences of the NH2- (N) and COOH-terminal tryptic peptides are those predicted from the cDNA sequence of human P2 (6). The possible phosphate acceptor sites are shown by arrows. No evidence for phosphorylation of the basic (pI = 9.3) NH2-terminal peptide was observed on high voltage electrophoresis of in vivo or in vitro 32P-labeled P1/P2.

containing a CK II phosphorylation site on the myc-encoded protein (Myc) (35) (kindly provided by Dr. B. Lüscher, Fred Hutchinson Cancer Research Centre, Seattle, WA) were incubated with exogenous CK II as described under "Materials and Methods." Michaelis-Menten kinetics showed that the $K_m$ value for the C-22 synthetic peptide was 13.4 μM. This value is lower than that for the CK II specific peptide (24), but similar to that reported for a myc peptide (35). About 0.6 mol of phosphate were incorporated by CK II into each mole of peptide comprising the COOH terminus of Artemia P2, which contains 1 serine residue at position 98. In contrast, about 1.2 mol of phosphate were incorporated per mole of human C-22 peptide, indicating that CK II phosphorylated both Ser-102 and Ser-105.

Localization of the Site of Phosphorylation of P1 and P2—To identify the location of the CK II phosphorylation site on the P proteins, unstripped ribosomes were phosphorylated by the endogenous CK II-like enzyme in vitro. P1 and P2 were ethanol-extracted from the ribosomes as described under "Materials and Methods" and the eluted phosphoproteins analyzed by SDS-PAGE and autoradiography. P1 and P2 accounted for greater than 95% of radiolabeled proteins extracted from the ribosomes (not shown). P1 and P2 were digested by trypsin and the resulting phosphopeptides separated on thin layer plates by high voltage electrophoresis in the first dimension and chromatography in the second. As shown in Fig. 6A, two peptide(s) of very similar mobility were phosphorylated by the endogenous kinase. Similarly, when ribosome-bound P1 and P2 were phosphorylated by exogenous CK II, extracted, and subjected to tryptic peptide mapping, a phosphopeptide of almost identical $x/y$ coordinates to the upper peptide described above was observed (Fig. 6B). The upper peptide also comigrated with the phosphorylated and trypsin-digested C-22 synthetic peptide (Fig. 6C). These results indicate that CK II and the ribosome-bound kinase phosphorylate a serine residue(s) located at the COOH terminus of the P proteins. Since two-dimensional PAGE immunoblots revealed two additional spots for P1 and P2 following incubation with CK II in vitro (not shown), we infer that both Ser-102 and Ser-105 were phosphorylated in vitro. To test whether the same site(s) was phosphorylated in vivo, P1 and P2 were isolated from Ehrlich ascites cells that had been metabolically labeled with [32P]orthophosphate. The peptide map of P1 and P2 isolated from Ehrlich ascites cells labeled in vivo (Fig. 6D) was virtually identical to that observed for P1 and P2 phosphorylated in vitro. A mixing experiment confirmed that the P1/P2 tryptic phosphopeptides obtained...
from Ehrlich ascites cells labeled in vivo (Fig. 7A) or in vitro (Fig. 7B) comigrated (Fig. 7C). Since the same peptides were phosphorylated by exogenous CK II and the ribosome-bound kinase in vitro and P proteins labeled in vivo, it seems highly likely that CK II or a very similar kinase phosphorylates the P proteins in vivo.

To exclude the possibility that phosphorylation of other sites on P1/P2 (Fig. 5) occurred, P1/P2 labeled in vitro and in vivo were subjected to tryptic digestion and one-dimensional high voltage electrophoresis as described above, except that samples were applied to the center of the plate. Electrophoresis was performed for 15 min, and the phosphopeptides were located by autoradiography. No phosphopeptides other than those described above were detected.

**DISCUSSION**

The P proteins are three of approximately seven eukaryotic ribosomal proteins known to be phosphorylated in vitro (36). In this report, we have shown that all three P proteins are in vitro substrates of exogenously added CK II, that the target site for CK II phosphorylation is located within the highly conserved COOH terminus, and that the same site is phosphorylated in vivo. The P proteins were not promiscuous targets of protein kinases, since protein kinase A phosphorylated many ribosomal proteins but did not phosphorylate the P proteins in vitro. Conversely, the P proteins were the major ribosomal substrates of CK II in vitro. We also observed that ribosomes contained an endogenous kinase activity which phosphorylated the P proteins. The endogenous kinase showed a pattern of ribosomal protein phosphorylation very similar to exogenously added CK II. Phosphorylation of the P proteins by the ribosome-bound kinase was inhibited by heparin and utilized both ATP and GTP as phosphate donors, properties characteristic of CK II (33, 34). Furthermore, the CK II-specific peptide (24) competitively inhibited the endogenous kinase, strongly suggesting that it is CK II or a CK II-like enzyme. Since the activity of the kinase responsible for phosphorylating the P proteins was considerably decreased following stripping of accessory proteins off the ribosomes, it seems likely that the kinase is a cytoplasmic protein that transiently binds to the ribosome, rather than an intrinsic ribosomal protein. CK II is known to be located in the cytoplasm as well as the nucleus (37) and to phosphorylate several initiation factors involved in protein synthesis as discussed below.

The ability of the C-22 peptide to inhibit phosphorylation of the ribosomal P proteins in vitro, the low $K_m$ of phosphorylation of C-22 by CK II, as well as the failure to identify additional phosphopeptides by tryptic mapping indicates that the P proteins are phosphorylated at their COOH termini. This finding concurs with that of Amors et al. (38) who observed that Ser-98 of the Artemia P2 was exclusively phosphorylated in vivo. Stoichiometric studies on the C-22 synthetic peptide as well as two-dimensional SDS-PAGE immunoblots of P1/P2 suggested that both Ser-102 and Ser-105 of the mammalian sequence were phosphorylated by CK II in vitro. Tryptic peptide mapping of P1, P2, and the C-22 peptide phosphorylated in vitro, as well as P1 and P2 labeled in vivo, confirmed that the COOH termini of these proteins were the targets for phosphorylation by CK II and the ribosome-bound kinase. These experiments also suggest that CK II, or a closely related enzyme, phosphorylates P1 and P2 at their COOH termini in vivo. The detection of two closely spaced, acidic phosphopeptides is most likely explained by incomplete tryptic digestion or a post-translational modification that differs between P1 and P2.

The high degree of conservation of the COOH termini among species and their surface exposure on the ribosomal stalk suggest that they are important for the function of the P proteins (5). The functions of the P proteins have been investigated by selectively eluting P1 and P2 from the ribosome (9) and by observing the effect of anti-P antibodies on ribosomal function (10–12). Ribosomal cores stripped of P1 and P2 do not bind EF-1 (39) or EF-2 and lose EF-2-dependent GTPase activity (8, 9). Similarly, human and rabbit polycyonal anti-P antibodies inhibit EF-2-dependent GTPase activity (9, 20) and protein synthesis in general (11). Using mouse monoclonal anti-P antibodies, Uchiumi et al. (12) showed that a monoclonal antibody which recognized the COOH terminus on all three P proteins inhibited both EF-2-mediated GTP hydrolysis and poly(U)-directed polyphenylalanine synthesis, whereas antibodies which were specific for either P1 or P2 did not. Since the inhibition of GTP hydrolysis and polyphenylalanine synthesis could be reversed by preincubating the monoclonal antibody with a synthetic peptide corresponding to the COOH terminus, the authors (12) concluded that the functional site of the P proteins was located in the COOH terminus. Several studies have suggested that phosphorylation of P1 and P2 influences binding of these proteins to the ribosome and/or ribosomal function (9, 20). MacConnell and Kaplan (9) reported that EF-2-mediated GTP hydrolysis and polyphenylalanine synthesis were dependent upon the state of phosphorylation of P1 and P2. They were, however, unable to restore these functions by rephosphorylating P1 and P2 with protein kinase A (9). Since we were not able to phosphorylate the P proteins on intact ribosomes with protein kinase A and these proteins do not have protein kinase A sites predicted by the primary amino acid sequences (6, 7, 40), failure to restore function with protein kinase A does not seem surprising. Reconstitution studies with P1 and P2 phosphorylated by CK II should be of considerable interest.

Since the activity of CK II responds to stimulation of cells with epidermal growth factor, insulin-like growth factor, and insulin (41, 42), it has been suggested that the enzyme plays a role in cellular signal transduction. CK II is known to phosphorylate several initiation factors (eIF-2, eIF-3, and eIF-4) of protein synthesis (43), but it is unclear whether phosphorylation of these factors by CK II or other kinases has any influence on protein synthesis (37, 43). In the nucleus, CK II phosphorylates DNA-topoisomerase II, RNA-polymerase II as well as Myc (35) and the myb-encoded oncoprotein (Myb) (44). Transcription by RNA-polymerase II was initiated when the inactive, unphosphorylated form was phosphorylated by CK II at the highly conserved, acidic COOH terminus (45). Phosphorylation of Myb by CK II also appears to be functionally important, since the binding of Myb to DNA was reduced when Myb was phosphorylated by CK II and the CK II target site was deleted in cell lines whose transformation was ascribed to Myb activity (44). Whether growth factor induced stimulation of CK II results in the phosphorylation of the ribosomal P proteins and thereby affects protein synthesis is the subject of ongoing studies.

Anti-P autoantibodies are highly specific for the disease SLE (13). Epitope mapping by partial proteolysis revealed that the major epitope recognized by anti-P autoantibodies is located within the COOH-terminal 22 amino acids (16). The existence of a CK II phosphorylation site in the immunodominant epitope of the P proteins raises important questions regarding the antigenicity and immunogenicity of the unmodified and post-translationally modified forms of the P proteins. The role of phosphate groups in anti-P binding activity.
is currently under investigation.

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