A single specifically labeled protein ($M_r = 42,000$) is found when *Dictyostelium discoideum* crude membrane (ghost) preparations are photoaffinity-labeled with either $[^{32}P]8N_3$-cAMP or $[^{32}P]8N_3$-AMP. With the cAMP reagent, there is a close correlation between the labeling of this protein and the membrane phosphodiesterase activity. During cell differentiation the developmental time courses of the photoaffinity labeling of the $M_r = 42,000$ protein and the membrane phosphodiesterase are identical. In addition, cyclic nucleotide substrates of the membrane phosphodiesterase also inhibit the photoaffinity labeling of the $M_r = 42,000$ protein. AMP, which is a poor inhibitor of the membrane phosphodiesterase, is a very effective inhibitor of the photoaffinity labeling. These results suggest that the $M_r = 42,000$ protein is labeled by $[^{32}P]8N_3$-AMP produced by the action of membrane phosphodiesterase on $[^{32}P]8N_3$-cAMP. The membrane phosphodiesterase of ghosts does in fact mediate this conversion. When ghosts are photoaffinity-labeled with $[^{32}P]8N_3$-AMP, the $M_r = 42,000$ protein is again specifically labeled. In contrast to the results with the cAMP photoaffinity reagent, labeling with the AMP ligand is inhibited only by the noncyclic adenine nucleotides AMP, ADP, and ATP. The protein containing this AMP binding site is quantitatively extracted from the membrane with 0.6 M KI (but not detergents) and quantitatively binds to DNase I. Tryptic peptide maps of the photoaffinity-labeled protein indicate a relatively discrete localization of the attached radioactivity. Photoaffinity-labeled peptides co-migrate on sodium dodecyl sulfate gels with peptides derived from *D. discoideum* membrane-associated actin. These properties identify the labeled protein as membrane-associated actin. The photoaffinity-labeled AMP binding site may be distinct from the ATP binding site of actin.

Movement of cells which adhere to a solid substratum appears to require actin and associated contractile proteins. In cases where such movement is vectorial in response to a chemoattractant, there must be a mechanism by which the receptor for the chemoattractant regulates this contractile apparatus. *Dictyostelium discoideum* uses cAMP as an "acra-sin" or chemoattractant during the aggregation phase of the developmental transition from a population of single amoeboid cells to a multicellular slug (1, 2). Cell surface receptors for cAMP have been inferred from binding studies with $[^{3}H]cAMP$ (3-8). These receptors appear to be linked to directional cell motility and to guanyl and adenyl cyclase stimulation (9-11). A cell surface cyclic nucleotide phosphodiesterase hydrolyzes the chemoattractant to AMP, thus enhancing the signal/noise ratio for cAMP detection (12, 13). To identify and characterize the cAMP receptor, we have photoaffinity-labeled intact cells with $[^{32}P]8N_3$-cAMP. A single specifically labeled protein of $M_r = 40,000$, which has the nucleotide specificity and developmental time course of the chemoattractant receptor has been found (8).

In the course of photoaffinity-labeling plasma membrane fractions with $8N_3$-cAMP, only a single specifically labeled protein was found. A correlation between the labeling of this protein of $M_r = 42,000$ and the phosphodiesterase activity of the membranes was observed, suggesting that the labeled protein was either the membrane phosphodiesterase itself or a different protein labeled by the $[^{32}P]8N_3$-AMP produced from the photoaffinity reagent by the action of membrane phosphodiesterase. In order to determine which hypothesis was correct, the photoaffinity labeling of the $M_r = 42,000$ protein by both $[^{32}P]8N_3$-cAMP and $[^{32}P]8N_3$-AMP has been characterized and compared. These studies demonstrate that the labeled protein is not membrane phosphodiesterase and that the reagent responsible for the labeling of the membrane protein is in fact $8N_3$-AMP. The binding site labeled by this reagent is specific for noncyclic adenine nucleotides. Tryptic peptide maps of the photoaffinity-labeled protein indicate a relatively discrete localization of the covalently attached radioactivity. The protein containing this labeled AMP binding site has been identified as membrane-associated actin by a number of criteria. The nucleotide binding site labeled by $8N_3$-AMP has a specificity different from that of the well-known ATP binding site of actin.

**EXPERIMENTAL PROCEDURES**

_Materials—*[^{32}P]8N_3$-cAMP and unlabeled $8N_3$-cAMP were purchased from ICN, and unlabeled $8N_3$-AMP was from Sigma. $[^{32}P]8N_3$-AMP was prepared by exposing 1.0 ml of 1 mM $[^{32}P]8N_3$-cAMP to $10^{-10}$ ghosts at pH 6.2 for 30 min. Ghosts were removed by centrifugation. Polygram Cel 300 cellulose thin layer plates were from Brinkmann, and Silica Gel 60 plates were from E. Merck. DNase I Sepharose was purchased from Worthington. Actin purified from rabbit skeletal muscle was a gift from Dr. Mark Willard, Washington University._

_Methods—Amoebae of the *D. discoideum* axenic strain M3 (from W. F. Loomis) were grown on HL-5 medium (2). When vegetative cells reached a density of 3 to 6 $\times$ 10^5 cells/ml, they were harvested_
by centrifugation, washed, and differentiated following resuspension at 10^7 cells/ml in 17 mM sodium/potassium phosphate buffer, pH 6.2, containing 50 μg/ml of streptomycin sulfate (2). The cells were differentiated on a gyratory shaker at room temperature. Ghosts were prepared as described by Sussman and Boaschitz (14) from cells differentiated for 8 h unless otherwise specified. The ghost preparations consist of cells largely depleted of soluble constituents. Electron microscopy reveals a discontinuous plasma membrane containing some intracellular membranes, mitochondria, and disrupted nuclei. Purified plasma membranes were prepared with an aqueous two-phase polymer system and had the reported enrichments of surface markers and depletions of soluble, mitochondrial, and nuclear markers (15, 16).

Photoaffinity labeling was carried out by mixing ghosts, [³²P]8N³-CAMP nucleotide, and any competing nucleotide in 17 mM phosphate buffer, pH 6.2, in 200 to 250 μl total volume in 17-mm diameter wells of a Falcon plastic plate (No. 3008). The preparations were exposed to ultraviolet light from a UVSL-25 mineral light (short wavelength) at a distance of 1 cm for 10 min. After labeling, samples were washed four times by centrifugation at 1,000 × g for 5 min with cold 17 mM Tris-HCl, pH 8.0, containing 0.1 mM phenylmethylsulfonyl fluoride. Samples were solubilized in 3% SDS, 1% mercaptoethanol, 5% glycerol, 1 mM EDTA, 0.1 mM phenylmethylsulfonyl fluoride in 10 mM Tris-HCl, pH 8.0, and electrophoresed on 7.5 to 15% acrylamide gradient slab gels (17). The gels were stained with Coomassie blue and dried. Autoradiography was done on Kodak XR-5 film with a Cronex lightning plus intensifying screen at -70°C (18, 19). A quantitative estimate of the amount of radiolabel incorporated into proteins was obtained by weighing tracings of peaks from densitometer (Joyce-Loebl) scans of autoradiographs.

KI extraction was performed by exposing ghosts at 5 × 10⁷/ml to 0.6 M KI at 0°C for 30 min. Following centrifugation at 45,000 × g for 15 min, the supernatant was collected. Protein in this KI extract was precipitated by making the solution 10% in trichloroacetic acid. The protein pellets were washed three times with acetone and dissolved as described above prior to analysis by electrophoresis and autoradiography.

For determination of binding to DNase I, 100 μl of DNase I-Sepharose was equilibrated with 0.6 M KI. Two hundred microliters of KI extract of labeled ghosts was incubated with the resin for 1 h with periodic mixing. The total excluded volume was made up to 0.5 ml with 0.6 M KI. This supernatant and those from three 0.5-ml washes were pooled. Two hundred microliters of untreated extract was diluted to 2 ml with KI. To both solutions, 0.4 ml of 50% trichloroacetic acid and 25 μl of 1 mg/ml of bovine serum albumin were added. The protein pellets were washed, electrophoresed, and autoradiographed as described above.

Peptide mapping was carried out on a mixture of KI extract from 5 × 10⁷ ghosts and 1 mg of purified rabbit muscle actin. Reduction, carboxymethylation, and trypsin digestion were carried out following the methods of Frazier et al. (20). Electrophoresis was performed as described by Stephens (21). Chromatography was carried out employing either pyridine/butanol/acetic acid/H₂O (10:5:5:12) or chloroform/methanol/ammonium hydroxide (2:2:1) as solvents. Peptides were visualized by ninhydrin staining and autoradiography.

Peptide mapping analysis was also done following limited proteolysis with papain essentially by the methods of Cleveland et al. (22). The proteins from the KI extract of 10⁷ ghosts photoaffinity-labeled with [³²P]8N³-CAMP was electrophoresed in the usual manner. The gel was stained for 15 min and destained for 25 min. The actin bands were cut out and placed in the wells of a second gel consisting of a 10 to 20% acrylamide resolving gel and a 4-cm stacking gel. Various amounts of papain (1 mg to 3 μg) were added to each well. When the dye front was about 0.5 cm above the resolving gel, the power was turned off. After 30 min the run was completed as usual. Staining and autoradiography of the gel were performed as described above.

Phosphodiesterase was measured using a modification of the method of Brooker et al. (23). The reaction mixture consisted of 0.25 × 10⁷ ghosts or cells in 17 mM phosphate (pH 6.2), 1 mg/ml of alkaline phosphatase, 1 μM cAMP, and various concentrations of competing nucleotide in a total volume of 300 μl. The reaction was allowed to proceed for 20 min at room temperature. [³²P]cAMP binding was determined as described by King and Frazier (6, 7).

RESULTS

8-N³-cAMP Labeling of Ghosts—When the photoaffinity reagent [³²P]8N³-cAMP is incubated with the ghost preparation (see "Methods") in the presence of UV illumination, only two membrane-associated proteins become heavily covalently labeled (Fig. 1A). The upper heavily labeled band with an apparent molecular weight of 53,000 appears to be nonspecifically labeled in this membrane preparation since the incorporation of radioactive by this band is not prevented by any of the cyclic and noncyclic nucleotides tested. The lower band, however, with an apparent molecular weight of 42,000, appears to be specifically labeled. cAMP (Fig. 1B), cGMP (Fig. 1C), and AMP (Fig. 1D) were used as described under "Methods."

Fig. 1 (left). Autoradiograph of SDS-gels of photoaffinity-labeled ghosts. Ghosts were labeled using 10⁻⁷ M [³²P]8N³-cAMP plus either: A, no additional nucleotide; B, 10⁻⁵ M cAMP; C, 10⁻⁵ M cGMP; or D, 10⁻⁵ M AMP. Photolysis and autoradiography were carried out as described above.

Fig. 2 (center). Thin layer chromatography of 8-N³-nucleotides. Standards (Lane A), [³²P]8N³-CAMP (Lane B), and the product of 1 ml of 1 μM photoaffinity ligand treated with 5 × 10⁷ ghosts for 30 min at room temperature (Lane C) were run on cellulose thin layer plates in either isopropl alcohol/NH₄OH/H₂O (7:1:3) (I) or butanol/acetic acid/H₂O (5:2:3) (II). Standards are: 1, 8N³-cAMP; 2, cAMP; 3, 8N³-AMP; and 4, AMP.

Fig. 3 (right). Autoradiograph of SDS gels of [³²P]8N³-AMP photoaffinity-labeled ghosts. Ghosts were labeled using 10⁻⁷ M [³²P]8N³-AMP plus either: A, no additional nucleotide; B, 10⁻⁷ M cAMP; C, 10⁻⁷ M cGMP; D, 10⁻⁵ M AMP; E, 10⁻⁵ M ATP; or F, 10⁻⁵ M ADP. Photolysis and autoradiography were carried out as described above."
and AMP (Fig. 1D) were all effective competitors of photoaffinity labeling. Identical results were also obtained using a more purified plasma membrane preparation (data not shown).

The chemotactic receptor and membrane phosphodiesterase, known membrane-associated cAMP binding proteins of D. discoideum, have been well characterized regarding their nucleotide specificities and developmental regulation. These properties were determined for the $M_r = 42,000$ labeled protein. The data (not shown) demonstrate a strong correlation between the activity of membrane phosphodiesterase and the amount of radiolabel incorporated into the $M_r = 42,000$ protein. The fact that AMP readily inhibits photoaffinity labeling of this protein (Fig. 1) at concentrations that do not significantly inhibit membrane phosphodiesterase activity (not shown) suggests that the role of membrane phosphodiesterase in photoaffinity labeling the $M_r = 42,000$ protein is that of converting the 8N3-cAMP to the active labeling species 8N3-AMP. To test directly this hypothesis, [32P]8N3-cAMP was incubated with a preparation of ghosts, and the products of this incubation were analyzed by thin layer chromatography in two solvent systems which resolve AMP and cAMP. These results are shown in Fig. 2. In Solvent System I, the untreated photoaffinity reagent co-migrates with authentic 8N3-cAMP. After treatment of this preparation with the membrane phosphodiesterase of ghosts, the major component migrates with authentic 8N3-AMP. The conversion of the photoaffinity reagent to the AMP form is confirmed by the same distribution of products in Solvent System II. Thus, the ghost preparation can rapidly convert the photoaffinity reagent to the AMP derivative.

8N3-AMP Labeling of Ghosts—When ghosts are photoaffinity-labeled with the [32P]8N3-AMP reagent at concentrations previously used for labeling with 8N3-cAMP, only one labeled band is found upon SDS-gel electrophoresis and autoradiography (Fig. 3A). In contrast to the results obtained with the cAMP reagent, labeling with the AMP compound is not inhibited by cAMP (Fig. 3B) or cGMP (Fig. 3C). AMP (Fig. 3D) is an extremely effective competitor of the photoaffinity labeling. The same results were also observed using purified plasma membranes (data not shown). ATP (Fig. 3E) and ADP (Fig. 3F) are inhibitors of labeling but are less effective than AMP. The detailed nucleotide specificity of the photoaffinity labeling of this protein with the AMP reagent is shown in Fig. 4. The labeling reaction is inhibited most effectively by AMP, ADP, and ATP, while many cyclic nucleotides are only very poor competitors. Thus, only noncyclic adenine nucleotides appear to be effective competitors for the site responsible for binding the photoaffinity reagent. The same protein in the ghost preparation is apparently labeled by either the AMP reagent directly or by the cAMP reagent after its conversion to the AMP form by membrane phosphodiesterase. The molecular weight of this protein is 42,000, identical to that reported for D. discoideum actin (24). The specificity for non-cyclic adenine nucleotides is characteristic of actin as well (25-27). Thus, further studies were undertaken to determine if this labeled protein of the ghost preparations was in fact membrane-associated actin.

**Extraction of the Photoaffinity Labeled Protein**—The membrane-associated actin of D. discoideum can be quantitatively extracted with KI (24). Thus, ghosts were photoaffinity-labeled with the [32P]8N3-AMP reagent in the absence (Fig. 5A) and presence (Fig. 5B) of competing AMP. As in previous experiments, the $M_r = 42,000$ band is heavily labeled and excess AMP substantially inhibits its labeling. These two labeled ghost preparations then were extracted with 0.6 M KI. After extraction, the particulate (Fig. 5, C and D) and soluble (Fig. 5, E and F) fractions of the extract were electrophoresed and autoradiographed in parallel. Clearly, treatment with 0.6 M KI specifically extracts the labeled $M_r = 42,000$ band since the soluble extract contains only a single labeled protein. The
nonspecifically labeled proteins of the ghosts are not extracted with KI (Fig. 5, C and E). The Coomassie staining pattern of the gel (not shown) also indicates an enrichment of a Mr = 42,000 band in the soluble extract. The nonionic detergents, Emulphogene BC720 and Triton X-100, do not extract the Mr = 42,000 protein as completely as 0.6 M KI (not shown). Thus, the extraction properties of the labeled protein are consistent with those of membrane-associated actin in D. discoideum.

Actin purified from rabbit skeletal muscle (28) (predominantly F-actin) was photoaffinity-labeled with [32P]8N3-AMP using the same conditions employed for labeling the ghost preparations. As seen in Fig. 5G, this actin was readily labeled. This demonstrates that 8N3-AMP will bind to an authentic actin preparation.

Binding of the Photoaffinity-Labeled Protein to DNase I-Sepharose—Ghosts were affinity-labeled with the AMP reagent in the absence and presence of competing AMP and extracted with KI as above. The extract prepared from ghosts labeled without competing AMP contained a single band (Fig. 6, Gel Lane A). This KI extract then was adsorbed with a DNase I-Sepharose preparation (29). After adsorption the material which did not bind to DNase I-Sepharose was concentrated and run on an SDS gel in parallel with the untreated half of the KI extract. Gel Lane B in Fig. 6 indicates that the great majority of the photoaffinity-labeled band was removed from the KI extract by DNase I-Sepharose. These results were quantitated by densitometry, and the scans of Gel Lanes A and B are shown in Fig. 6. Peak area measurements indicate that 90% of the photoaffinity-labeled band was adsorbed to DNase I-Sepharose. Since the only conditions known which elute actin from DNase I-Sepharose are treatment with high concentrations of guanidine hydrochloride, elution of the bound labeled protein under these conditions would not serve to distinguish between specific and nonspecific binding to the affinity resin.

Photoaffinity Labeling of Soluble Actin—Soluble extracts of D. discoideum contain large amounts of actin, up to 8% of the soluble protein (30). We have attempted to determine whether this soluble actin has the same photoaffinity labeling properties as the membrane-associated protein. The soluble extract from cells at 5 × 10⁷ cells/ml contains more ATP than was used to inhibit labeling of membrane-associated actin (see Fig. 3E). Thus, it was necessary to dialyze the soluble extract prior to labeling. After dialysis against 17 mM phosphate, pH 6.2, soluble actin could be specifically photoaffinity-labeled, but the Coomassie-stained gel showed that much of the actin and other soluble proteins had been degraded during the dialysis. After dialysis against pyrophosphate with or without sucrose, conditions which stabilize actin (30), the actin did not incorporate photoaffinity label, even though the Coomassie staining pattern confirmed that little or no photolysis had occurred (not shown).

Peptide Mapping of the Photoaffinity-labeled Protein—Since the KI extract of photoaffinity-labeled ghosts contains only one labeled protein (Fig. 5), it is possible to study the distribution of the covalently bound photoaffinity label on the protein without further purification. The nonspecific reactivity of substituted azides may lead to their insertion into a variety of covalent bonds (31). It was thus of interest to determine whether the sites of attachment of the photoaffinity label were relatively heterogeneous or confined to relatively few sites on the labeled protein. Tryptic peptide maps of the carboxymethyl photoaffinity-labeled protein were prepared using thin layer electrophoresis and chromatography in two different solvent systems. Rabbit muscle actin was used as a carrier protein in these experiments. These results are shown in Fig. 7, A and B. The same digest was used in both maps, and both were electrophoresed from right to left at pH 3.5. Chromatography was carried out from the bottom to top of both maps. In Fig. 7A, the system employed was of low pH.
and in Fig. 7B the chromatographic solvent was at a very basic pH. The major ninhydrin positive spots obtained from the S-carboxymethyl rabbit muscle actin are distributed as expected (21, 32). The thin layer peptide maps were autoradiographed to locate the radioactive peptides derived from the photoaffinity-labeled protein. In both mapping systems, relatively few (four) labeled peptides are detected, suggesting a limited diffusion of the photoactivated azido reagent during its reactive lifetime. This observation, along with the low concentrations of reagents at which photoaffinity labeling of this protein occurs, suggests a high affinity of the reagent for the nucleotide binding site on the protein. Since substitution of tryptic peptides by the photoaffinity reagent may alter their mobility in both the electrophoretic and chromatographic dimensions, it is not possible to assign radioactively labeled peptides to their unlabeled counterparts derived from rabbit muscle actin.

To provide a more direct comparison of labeled peptides with those derived from D. discoideum actin, limited proteolysis followed by peptide mapping on SDS-polyacrylamide gels by the method of Cleveland et al. (22) was performed. Fig. 8 shows the Coomassie-stained gel (A) and the autoradiograph (B) of labeled actin exposed to various amounts of papain. The actin in the gel lane farthest to the left was not treated with papain. The actin in the other lanes from left to right was treated with 1, 3, 10, 30, 100, 300, 1,000, and 3,000 ng of papain.

**DISCUSSION**

Only one protein is specifically photoaffinity-labeled by low concentrations of [³²P]8N₃-AMP or [³²P]8N₃-cAMP in D. discoideum ghost or plasma membrane preparations (Figs. 1 and 3). The membrane phosphodiesterase generates the active AMP label when the AMP reagent is used. Several criteria identify the labeled protein as membrane-associated actin. The radioactively labeled protein has the same molecular weight as actin, 42,000 (24). It is selectively extracted from membranes by 0.6 M KI (24), and it quantitatively binds to DNase I (29). Furthermore, it is localized on the cytoplasmic face of the plasma membrane since it is not detected when intact cells are labeled with either 8N₃-AMP or 8N₃-cAMP (8). In addition, the radioactivity co-migrates with peptides derived from D. discoideum membrane actin (Fig. 8).

Photoaffinity labeling of membrane-associated actin has revealed several interesting properties regarding the interaction of adenine nucleotides with this protein. The 8N₃-AMP appears to bind to a relatively high affinity site specific for noncyclic adenine nucleotides since, of the several cyclic and noncyclic nucleotides tested, only AMP, ADP, and ATP inhibited photoaffinity labeling (Fig. 4). Interestingly, AMP appeared to be a more effective inhibitor of the labeling reaction than either ADP or ATP (Fig. 3). This result is in contrast to that obtained in studies of adenine nucleotide binding to skeletal muscle actin (33-35), in which AMP bound much more weakly than ADP or ATP. This implies that there may be an AMP binding site on the D. discoideum membrane-associated actin which is distinct from the well characterized ADP-binding site. Since the labeled actin avidly binds to DNase I, the interaction of these two proteins appears to be unaffected by covalent incorporation of the photoaffinity ligand. This suggests that the binding of actin to DNase I is not accompanied by removal of bound nucleotide.

ATP binds to G-actin and upon polymerization to F-actin the bound ATP is hydrolyzed to ADP, which remains associated with the F-actin (38). If the photoaffinity ligand binds to this adenine nucleotide site, then the nucleotide must be exchangeable. Some fraction of the membrane-associated actin is in a form identical to F-actin, as indicated by decoration with the myosin subfragment S1 (37). Rabbit muscle F-actin is also photoaffinity-labeled by 8N₃-AMP (Fig. 5G). Although the rate of exchange of nucleotide bound to F-actin is much slower than that bound to G-actin (39), exchange does occur (34). Furthermore, exchange of AMP for the ADP of F-actin has been accomplished with appropriate experimental conditions (35), thus directly demonstrating that AMP can bind to F-actin at the ADP site. In the experiment with muscle actin, the amount of label incorporated indicates that 0.2% of the actin molecules were labeled. Thus, only a small amount of nucleotide exchange need take place in order to incorporate enough photoaffinity ligand to account for the labeling observed in these experiments. However, the experiments do not rule out the possibility that photoaffinity labeling identifies an AMP-regulatory site distinct from the ADP site of D. discoideum membrane-associated actin.

The soluble unpolymerized actin of many nonmuscle cells appears to form a complex with a protein called profilin which inhibits actin polymerization (38). Profilin seems to bind to G-actin in such a way as to prevent nucleotide binding and at the same time stabilize the unpolymerized nucleotide-free form (39). If such a complex is found in D. discoideum and is stable during dialysis of cytosolic fractions against pyrophosphate- or sucrose-containing solutions, it would account for the observed lack of incorporation of [³²P]8N₃-AMP into soluble actin. Furthermore, the degradation of actin during dialysis against phosphate buffer may be accompanied or
preceded by dissociation of the profilin-actin complex, thus unmasking the nucleotide binding site. Studies to determine the factors that influence incorporation of the photoaffinity label into soluble actin could provide pertinent information for the elucidation of the mechanisms regulating the physical state of actin in the cell.

Changes in the cellular distribution and synthesis of actin occur during the developmental cycle of *D. discoideum*. In vegetative amoebae, the protein is uniformly distributed throughout the entire cell, while in aggregating cells actin is localized under the advancing regions of membrane (40). Furthermore, during the first few hours of differentiation, actin accounts for about 20% of total protein synthesis, and this newly synthesized actin becomes preferentially associated with the cell membrane (41, 42). Thus, a mechanism must exist for regulating the localization of actin within the cell and the association of the protein with the membrane. In addition, actin and associated contractile proteins (38) are certainly required for cell motility. The regulation of this contractile apparatus by the cAMP receptor which functions in chemotaxis to direct cell movement represents an interesting case of the general problem of transmembrane regulation of contractile proteins. Labeled actin, which can be obtained from membrane preparations of *D. discoideum* by methods described in this communication, should provide a useful tool for identifying actin binding sites of the plasma membrane which may be directly or indirectly coupled to the cAMP receptor. The identification of these actin binding sites in the membrane is an important step in the elucidation of the mechanism by which activation of CAMP receptors leads to vectorial cell movement.

**Acknowledgments**—We thank Dr. Mark Willard for rabbit muscle actin and Drs. M. Willard, C. Frieden, H. Waterston, and D. Bray for advice and helpful discussion.

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