Table 1. Carbohydrate content (milligrams per 100 mg of protein) of protein TL and of the alpha heavy chain of a γ_{A1} myeloma globulin.

Total hexoses	Fucose	Sialic acid	Total hexosamines
Protein TL			
6.67	0.84	3.72	5.36
Alpha chain of γ_{A1} myeloma			
3.19	0.53	0.79	2.80

toward polymerization precluded purification of the "native" monomer.

Papain digestion of the purified TL polymers in the presence of 0.01*M* cysteine hydrochloride was ineffective. When carried out in the presence of 0.1*M* 2-mercaptoethanol, papain degradation resulted in the production of small peptides without any immunologically identifiable component.

With the use of appropriate antiserums, IgA myeloma proteins can be divided into two major subclasses γ_{A1} and γ_{A2} (Le and He) related to the α-chains (*5*). The TL protein was shown to be related to the γ_{A1} subclass since, in gel-diffusion experiments, it formed a spur over the γ_{A2} (deficient) protein (*5a*). To further characterize TL protein, we attempted to compare its antigenic properties to those of several γ_{A1} myeloma proteins and their subunits. All antiserums were made specific for IgA by absorption with light chains or serum of patients having selective IgA deficiency, or both. Since none of the studied IgA myeloma proteins have yielded Fc fragments after papain hydrolysis, we were unable to compare directly the TL protein to these fragments. No antigenic determinants common to TL protein and Fab fragments of these myeloma globulins could be demonstrated. Moreover several absorbed antiserums (which did not precipitate with light chains) showed a definite spur of heterologous IgA myeloma proteins and of normal IgA over the TL protein. With two of these antiserums, TL protein was found to be antigenically deficient when compared with all tested IgA myeloma proteins of type K as well as of type L (Fig. 1D). However, in view of the poor immunogenicity of the Fd piece and of the importance of the light-heavy chain interaction in establishing the antigenic structure of the immunoglobulin molecules (*6*), these findings do not conclusively indicate that the TL protein lacks Fd piece. Conformational specificity has not been ruled out as the source of these antigenic differences, because antigenic deficiency of TL protein with respect to purified myeloma α-chains could not be convincingly demonstrated. Although the high carbohydrate content of TL protein as compared to α-chains of γ_{A1} subclass (Table 1) suggests that it may represent mainly the Fc portion of the chain, further physicochemical studies are required in order to determine whether TL protein is a complete α-chain or a portion thereof.

Immunofluorescence studies with many different antiserums to κ and λ light chains showed no evidence of light-chain synthesis in the few lymphoid and plasma cells of the bone marrow which produced the TL protein. Samples of small intestine taken at biopsy were submitted to short-term culture in the presence of ^{14}C-labeled amino acids, and the cell extracts were studied by immunoelectrophoresis and subsequent autoradiography (*7*). The only immunoglobulin line labeled was the TL protein line (Fig. 1B). This TL protein synthetized in vitro did not react with antiserums to light chains and no free labeled light chains could be demonstrated in the cell extracts. Therefore it seems reasonable to assume that we are not dealing with an absence of light-heavy chain assembly but with an actual lack of light-chain synthesis in the proliferating cells.

This newly recognized abnormality of IgA is perhaps not uncommon in patients with this peculiar type of abdominal lymphoma, because an analogous protein has been recently detected in our laboratory in two other cases with a very similar clinical pattern. Alpha-chain disease may thus represent a well-defined condition with characteristic clinicopathological and biological features and possibly a genetic predisposition.

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8. We thank Dr. Y. Le Quintrec for referring the patient, Drs. J. P. Vaerman and J. F. Heremans for supplying the reference proteins Le and He, Dr. R. Van Rapenbusch for ultracentrifugal analysis, Mrs. Y. Signoret, A. Chevalier, and C. Masurel for technical assistance. Supported by grant CR.66.237 from the French National Institute for Health and Medical Research.

5 September 1968

Cucumber Sex Expression Modified by 2-Chloroethanephosphonic Acid

Abstract. *Application of 2-chloroethanephosphonic acid (120 to 240 parts per million) to monoecious cucumber plants when the first true leaf was 2 centimeters in diameter has resulted in as many as 19 continuous pistillate nodes. Control of the flowering habit simplifies the production of hybrid seed and offers the possibility of enhancing cucumber yields.*

Monoecious cucumbers (*Cucumis sativus*) treated with 2-chloroethanephosphonic acid have exhibited gynoecious characteristics for as many as 19 nodes. Other effects on growth regulation such

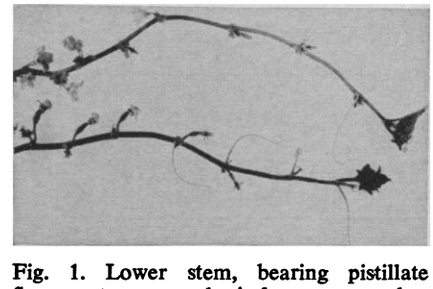

Fig. 1. Lower stem, bearing pistillate flowers at every node, is from a cucumber plant treated with 2-chloroethanephosphonic acid (240 parts per million) at first true-leaf stage. Upper stem, with staminate flowers at every node, is from an untreated plant.

as epinasty, root initiation, growth retardation, hastening of fruit ripening, and defoliation have been attributed to this chemical (*1*). More recently, treatment of pineapple with the same chemical has induced flowering (*2*).

Monoecious cucumbers of the cultivar "S.C. 23" were sprayed with one or more applications of a water solution of 2-chloroethanephosphonic acid at concentrations of 120, 180, and 240 parts per million (*3*). We applied the treatments to the point of runoff, beginning when the first true leaf was about 2 cm in diameter. Plants treated more than once were sprayed at 48-hour intervals.

The greatest number of continuous pistillate nodes was produced by plants treated with either two applications of 240 parts per million or four applications of 120 parts per million (Table 1). Lesser numbers of nodes were affected by the lower dose, suggesting an additive effect of the chemical. Untreated plants usually did not bear a pistillate flower until about node 9 and produced only 3.5 pistillate flowers on the first 20 nodes (Fig. 1).

It was also possible to enhance the yield of a monoecious variety with this chemical (*4*). The variety "Model," when sprayed with the chemical, produced essentially the same total yield as the five leading gynoecious varieties in a variety trial. Earliness of production was also improved.

The ability to control flowering in cucumbers will have far-reaching effects, for example, on the production of hybrid seed. To produce a hybrid, one need only plant the parental lines in separate rows, treat the female parent with 2-chloroethanephosphonic acid and let bees make the pollen transfer. If there is adequate isolation from other cucumber plantings, viable seed produced by the female parent should result from the cross.

Table 1. Effect of 2-chloroethanephosphonic acid on the production of pistillate flower-producing nodes by plants of the monoecious cucumber cultivar "S.C.23." Data were recorded for 20 nodes in all cases, and node No. 1 was the first true-leaf node; that is, the one immediately beyond the cotyledonary node.

Concentration (ppm)	Applications (No.)	Pistillate nodes	
		Continuous (No./plant)	Total (No./plant)
240	2	18.9	19.1
120	4	18.7	19.0
180	2	16.2	16.5
240	1	16.1	16.2
120	2	16.0	16.1
120	1	12.6	12.9
180	1	12.0	12.5
0	0	0.0	3.5

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1. Amchem Products, Inc., Information Sheet IS-37 (1967).
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3. Experiments were replicated three times on ten plants per plot for each treatment.
4. Plants were sprayed with the chemical once with a 120-ppm solution when the first true leaf was about 2 cm in diameter.
5. Published with the approval of the director of the North Carolina State University Agricultural Experiment Station. We thank Amchem Products, Inc., Ambler, Pennsylvania, for furnishing the chemical.

23 September 1968; revised 1 November 1968

Affinity Site Labeling of a Mouse Myeloma Protein Which Binds Dinitrophenyl Ligands

Abstract. *A mouse myeloma protein of the immunoglobulin A class, which specifically binds dinitrophenyl ligands, has been successfully affinity labeled (site-directed labeling) with several diazonium reagents, leading to inactivation of the combining sites. The labeling reagents reacted only with tyrosine residues of light chain origin. The 7S subunit of the myeloma protein appears to contain only one reactive site.*

Several human (*1*) and mouse (*2*, *3*) myeloma or Waldenström immunoglobulins which appear to be homogeneous and capable of binding ligands with high specificity have been described. They apparently have a homogeneous binding constant (*1*, *3*, *4*), and they satisfy all the operational criteria for chemically defining a single molecular form of antibody.

One mouse immunoglobin A myeloma protein (MOPC 315) which has been immunochemically characterized resembles normal mammalian antibodies to dinitrophenyl ligands in many of its characteristics (*3*). It binds ε-*N*(2,4 dinitrophenyl)-L-lysine as determined by equilibrium dialysis, and when the ligand is bound, it exhibits the characteristic shift in absorbancy and fluorescence quenching (*5*). Unlike induced antibodies which typically consist of molecules of varying affinities, the MOPC 315 protein appears to have uniform combining sites of high affinity ($K_A = 1$ to $2 \times 10^7 M^{-1}$) (*3*). Other mouse myeloma proteins of the immunoglobulin A class have been isolated which specifically combine with either dinitrophenyl or trinitrophenyl ligands, though these seem to have a lower combining affinity than MOPC 315.

In order to compare the combining sites of these proteins with each other and with those of specifically induced antibodies, we have applied the technique of affinity labeling (*6*). In this method, a modified ligand which is initially bound noncovalently to the combining site is used. The ligand contains a reactive group that can form a covalent bond with an amino acid residue side chain. In that it is preferentially localized in the combining site, the ligand reagent will preferentially react with side chains available in the combining site.

We now report results obtained when MOPC 315 was reacted with diazonium reagents under the conditions used to label rabbit antibodies to dinitrophenyl determinants (*7*, *8*).

The MOPC 315 tumor was grown in strain Balb/c mice; the myeloma protein was isolated from serum or ascites fluid by precipitation with ammonium sulfate at 5°C and dialyzed against water until a copious euglobulin precipitate formed. The euglobulin precipitate was either dissolved directly in the appropriate labeling buffer or in a 0.2*M* tris(hydroxymethyl)aminomethane-hydrochloric acid (tris) buffer (*p*H 8.6) prior to reduction and alkylation. The euglobulin (1 to 2 percent solution) was reduced with 0.01*M* dithiothreitol at room temperature for 1 hour, and the reduced protein was alkylated with iodoacetamide, in 10 percent molar excess over sulfhydril reagent, at *p*H 8.0 for 15 minutes at room temperature. The reduced alkylated preparation was then applied to a G-200 Sephadex column equilibrated with a *p*H 8.0 borate NaCl buffer and eluted at room temperature. Only the component having an effluent volume comparable to 7*S* γ-globulin was used.

A human γG_1 myeloma protein [γG(War)] was used as a control protein and was isolated by diethylaminoethylcellulose chromatography from a sodium sulfate precipitate of whole serum.

In all cases, the labeling reactions were conducted on protein solutions having an optical density of 4 at 280 nm. For analytical work, reaction volumes of 0.5 ml were routinely used. After reaction for various times, the