tors in the field, although it is supported more or less strongly by a considerable body of evidence.[1,2,3,4] We have now carried out some experiments which correspond so well in their results with the predictions of this theory as to leave little doubt of its correctness.

We have synthesized a substance which gives a specific precipitate with a mixture of two different antisera, but gives no precipitate with either antiserum alone. The substance contains two different haptenic groups, R and X, to which the two antisera are homologous: the anti-R serum was made by injecting rabbits with azoprotein containing R groups, and the anti-X serum by injecting with azoprotein containing X groups. The R and X groups were respectively the *p*-azophenylarsonic acid group and the *p*-azobenzoic acid group, and the RX substance used in most of our work was 1-amino-2-*p*-(*p*-azophenylazo)phenylarsonic acid-3,6-disulfonic acid-7-*p*-(*p*-azophenylazo)benzoic acid-8-hydroxynaphthalene. Similar results were also obtained with 1,8-dihydroxy-2-*p*-azophenylarsonic acid-3,6-disulfonic acid-7-*p*-(*p*-azophenylazo)benzoic acid-naphthalene.

The experimental results show that in the formation of the precipitate both of the two haptenic groups of the molecule enter into specific reaction, the R group with an anti-R antibody molecule and the X group with an anti-X antibody molecule. This is shown by the fact that the RX substance precipitates only with a mixture of the two antisera, and not with either one alone; the explanation of the failure of precipitation with only one antiserum given by the framework theory is that with respect to either antiserum the RX molecule is only monohaptenic and hence only univalent, and so can not act as the link between antibody molecules in the formation of a framework.

With the effective bivalence of the precipitating antigen thus proved, knowledge of the antibody-antigen molecular ratio for the precipitate provides the value of the average valence of the antibody molecules. The molecular ratio was found by analysis to be 0.7, which corresponds to $2/0.7 = 2.8$ for the average antibody valence.[5] If the antibody were univalent the molecular ratio would have the value 2, which is far greater than the experimental value. Further evidence for the effective multivalence of antibody is provided by the observation that the RX precipitate is soluble in excess of the mixed antisera; this solubility can not be explained on the basis of univalent antibody.

A detailed account of this work will be published in the *Journal of the American Chemical Society.*

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5 Similar values of the antibody-antigen molecular ratio have been previously reported (footnote 4) for precipitates of anti-R antisera and simple substances containing two or more R haptenic groups.

THE PRODUCTION OF MULTIPOLAR MITOSES IN NORMAL EMBRYONIC CHICK CELLS

THE sporadic occurrence of an abnormal type of cell division, namely, multipolar mitosis, in normal somatic cells remains unexplained. Kemp[1] reported finding a single tripolar mitosis in approximately 10,000 dividing cells in tissue cultures of tissues from normal chick embryos and adult fowls. A later study[2] of dividing cells from a double monster (Cephalopagus) revealed five triasters among 500 mitotic cells. The fact that multipolar mitoses with resultant unbalance of chromosome number in daughter cells occur during the growth of certain types of malignant cells is too well known to be emphasized.[3, 4, 5, 6] However, the significance of their occurrence in relation to malignancy has not yet been established. Attempts to produce this type of cell abnormality in normal somatic cells have met with varied degrees of success.[7, 8, 9, 10]

In the present experiments 4 series of cultures (total 92) of eight-day normal embryonic chick heart muscle were made by the usual hanging drop technique. A total of 12 to 14 chick hearts were used. The culture medium consisted of 1 drop of fowl plasma (dil. 1:1 with Tyrode solution) and 2 drops of a mixture of embryonic juice from 8- and 11-day chick embryos (dil. 1:1 with Tyrode solution). The

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medium was renewed on the cultures of series 1 and 2 on the sixth day of cultivation; on those of series 3 and 4 on the seventh day. Cultures of series 1 and 2 were fixed for cytological study on the seventh day of life *in vitro;* those of series 3 and 4 on the eighth day. The temperature for incubation was constant at a given point in the incubator but varied from 42+° C. at the back side to 37½° C. at the front. The heating units of the incubator consisted of two electric light bulbs (carbon). Consequently, light of a very low intensity was emitted intermittently. All cultures placed in the back half of the incubator were killed.

Microscopic examination of cultures of series 1 revealed the presence of numerous multipolar mitoses (chiefly triasters) in 8 of the 12 cultures fixed and stained. In cultures of series 2, 3 and 4 frequent triasters and other aberrant forms of mitosis have been found in at least 2 cultures of each series. The abnormal division figures consist chiefly of: triasters (ana-, telo-, and reconstruction phases); cells with 3 poles and two spindles. "Resting" cells with two or more nuclei are of frequent occurrence; chromosome vesicles (16 in one cell) have been observed. The unusually large size of cells exhibiting these abnormalities is noteworthy. The cultures presented a very vigorous growth. Over 500 dividing cells were counted in culture 2 of series 1.

A detailed study of the cultures at hand is in progress and the results will be published elsewhere at a later time. From the results already obtained it is obvious that the possibility of repeated production of multipolar mitoses in normal somatic cells *in vitro* is an important point of departure for determining the significance of this phenomenon of cell growth in relation to malignancy. A program of experimental work designed to analyze the factors responsible for the production of these modified cells has been planned.

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THE ROLE OF NIGHT TEMPERATURE IN PLANT PERFORMANCE[1]

IN the course of observations of the responses of some 240 varieties or species of plants to different temperatures and photoperiods[2] chance evidences became increasingly suggestive that the temperature during the night rather than the daytime level largely determines the type of response which the plants make to temperature. Hamner and Bonner[3] have reported this is true in the case of *Xanthium* (cocklebur).

This past winter several species of plants were grown in four greenhouses at different temperatures, approximately as follows: cool (55° F.); cool/warm (55° F. at night and 75° F. in the daytime); warm/cool (75° F. and 55° F.); and warm (75° F.). Each house had provision for both long- and short-day treatments. Some of the reactions of the plants given warm nights and cool days will be reported briefly.

They were of a pale color, particularly those in short days. "Warm climate" plants as Proso millet, corn, hemp, Biloxi soybean and sorghum were particularly yellowish, some being near the color "Grapefruit."[4] "Cool climate" plants as oxeye daisy, timothy, rye, nasturtium, brome grass and blue grass, on the other hand, developed a relatively good green color in cool days following warm nights.

The plants with pale color because of cool days following warm nights made relatively little growth when compared to those with warm days, either with warm or cool nights. They did, however, have practically normal blossom induction reactions. This was true for such short-day plants as Biloxi soybean, hemp, pigweed, cocklebur, poinsettia, Refugee bean and Jimson weed as well as the long-day species alfalfa, beet and snapdragon. Indeterminate types, as tomato, Russian dandelion and Alaska pea, had a time schedule like the plants in the warm house.

The warm nights with cool days had an effect like the continuously warm environment of delaying or inhibiting the flowering of snapdragon, poinsettia and beets. Other effects which were comparable in these two temperature environments were a reduction in the setting of seed of Alaska peas, alfalfa and yellow sweet clover, delayed tuberization of potatoes, reduced root formation by Russian dandelions and a masking of potato virus symptoms.

Some cool temperature plants, as bluegrass, oats and barley, tend to head better in the cool day house with warm nights than when these are kept warm both day and night, but do not set seed well as in the cooler houses. It would appear possible to use such types and secure evidence that the daytime temperature is most important. Such a conclusion should await the results from carrying the day temperatures at a lower level than was used this season.

These limited preliminary observations indicate that for a number of plants at least, the temperature during the dark period of the day is an important factor affecting blossom induction as well as some other reactions.

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[1] Published with the approval of the director of the Agricultural Experiment Station.

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