



Fig. 2. Curve 1 (left ordinate) is a plot of  $C/a$  against  $C$  in which  $C$  is the concentration of antiovalbumin in the bulk phase and  $a$  is the amount (in milligrams) of antiovalbumin remaining per 100 mg of tissue after the third washing. Curve 2 (right ordinate) is a plot of the physiological response to 2 mg of antigen of tissues that had been incubated for 1 hour in antiovalbumin and washed three times before testing. The equivalent histamine concentration, a measure of the physiological response, is obtained graphically from histamine standardization curves carried out for each muscle strip. The vertical bars indicate standard errors of the respective means.

ing to this relationship, a plot of  $C/a$  against  $C$  should give a straight line having a slope of  $1/\beta$  and an intercept of  $1/\alpha\beta$ . Curve 1 of Fig. 2 shows that the data, in the range between 0.028 and 0.140 mg of  $I^{131}$ -labeled gamma globulin per milliliter are adequately described by a line with a slope of 206.7 and an intercept of 61.5, from which the values of  $\beta$  and  $\alpha$  are found to be 0.0483 and 3.35, respectively.

2) To determine the dependence of Schultz-Dale response on concentration of antibody used for incubation, tissues were incubated at 27°C for 60 minutes in nonradioactive antiovalbumin gamma globulin, the same range of concentrations being employed as in part 1. These tissues were washed three times at 27°C and then equilibrated at 37°C for 30 minutes before they were challenged with antigen in the Schultz-Dale test. Frontal writing levers were used to obtain kymographic records of muscular response. Each muscle was titrated with at least three doses of histamine before and after challenge with 2 mg of ovalbumin, the second histamine titration being carried out in a dosage range which was closely equivalent to the muscular response to antigen. The histamine equivalent of the specific contraction was obtained by interpolation from the histamine calibration curve of the particular strip of gut.

Curve 2 of Fig. 2 illustrates the dose-response characteristics of challenged

muscle strips that had been incubated in the same concentrations of the antiovalbumin preparation that had been used in the experiments with the radioiodinated gamma globulin. Each point is the average of 11 to 17 values, the modal number being 16. As can be seen from the magnitudes of the standard errors, the evaluation of response at the higher protein concentrations is subject to a great deal of error because the sigmoidal dose-response curve with respect to histamine tends to flatten out at higher doses, making interpolation uncertain at near-maximal responses to antigen; for this reason incubation experiments were not made in the range beyond 0.14 mg of antiovalbumin per milliliter. Muscles could not be sensitized in concentrations of antibody lower than 0.014 mg of antiovalbumin per milliliter at 27°C.

Taking 0.014 mg/ml as the limiting value for sensitization, we can, on the basis of certain assumptions, make an approximate calculation of the amount of antibody necessary to set off a minimal reaction. Since preliminary experiments showed that there is no difference in the binding properties of normal and immune radioiodinated gamma globulin, we can assume that the binding of antibody is not preferential—hence, that the amount bound will be determined by the proportion of antibody in the total protein preparation, in this case 32.5 percent. The amount of total protein taken up by 100 mg of tissue from a concentration of 0.014 mg/ml is found to be  $2.4 \times 10^{-4}$  mg; from this finding, a value of  $7.8 \times 10^{-5}$  mg of antibody per 100 mg of tissue is obtained as the minimal ratio for discharge of the Schultz-Dale reaction.

The studies discussed in this report have shown that rabbit antiovalbumin can be bound to the serosal surface of guinea-pig ileum in proportion to the concentration of the antiovalbumin in the bulk phase. Since the relationship conforms to a Langmuirian isotherm, we can infer that these molecules form a monolayer; this does not imply that the combining sites are limited to the cell surfaces or to the superficial layer of cells. The reaction to antigen is also dependent on the amount of antibody bound; whether or not this reaction involves the formation of toxic complexes cannot be determined from the experiments discussed here.

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#### References and Notes

1. F. G. Germuth and G. E. McKinnon, *Bull. Johns Hopkins Hosp.* 101, 13 (1957).
2. I. L. Trapani, J. S. Garvey, D. H. Campbell, *Science* 127, 700 (1958).
3. G. A. Feigen and C. B. Nielsen, *Federation Proc.* 17, 45 (1958).
4. H. H. Dale, *J. Pharmacol. Exptl. Therap.* 4, 167 (1913); J. L. Mongar and H. O. Schild, *J. Physiol.* 135, 320 (1957).
5. This research was supported by the Office of Naval Research and Stanford University [under contract No. NR 225 (46)] and by the U.S. Atomic Energy Commission.
6. R. A. Keckwick and R. K. Cannan, *Biochem. J.* 30, 227 (1936).
7. A paper describing the injection schedule is in preparation.
8. W. L. Hughes and R. Straessle, *J. Am. Chem. Soc.* 72, 452 (1950).
9. G. A. Feigen and D. H. Campbell, *Am. J. Physiol.* 145, 676 (1946).

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#### Variations in Style Number and Other Gynoecial Structures of *Lychnis alba*

**Abstract.** Consistent variations in style number (from the typical five), ranging from zero to ten, inclusive, are found in ovaries of *Lychnis alba* flowers. The number of carpels, placentae, and vascular strands of a given ovary usually varied directly with the number of its styles.

Ovaries of the pistillate flowers of *Lychnis alba* Mill. typically bear five styles. However, some descriptions read: "Styles 5 rarely 4" (1); "Styles 5 occasionally 4 or rarely 3" (2); "Styli 5, rarius 4, v. hinc inde 3" (3). Westergaard (4), Warmke and Blakeslee (5), and Warmke (6) published illustrations of artificially induced polyploid hermaphrodite and other flowers of *Melandrium (Lychnis)* with ovaries bearing differing numbers of styles but did not comment upon them.

Earlier I had found ovaries of *Lychnis alba* with two, three, four, six, and seven styles. These variations appeared so frequently and consistently that I initiated a statistical survey in 1958 to determine what variations in the number of styles actually occurred in the species. The preliminary results are described in this report.

Flowers were collected from plants (i) in a greenhouse; (ii) near Iowa City and West Liberty, Iowa; and (iii) near Bemidji, Minn. Thirty daily collections were made during May and June 1958 from greenhouse plants (7); 29 spaced, individual collections were made outdoors in Iowa, and 31 were made in Minnesota. All pistillate flowers were taken from every plant in every plot studied. Collections were total, and there were no special selections from any plant or plot suspected of producing, or known to produce, ovaries with differing numbers of styles. Collections in Minnesota clover-alfalfa fields covering several

acres were made from smaller subplots selected at random, or were made by collecting all *Lychnis* flowers encountered along lines walked edge-to-edge or corner-to-corner of a field.

The number of styles was counted for every flower collected. Each collection was tabulated, and cumulative records were kept, separately, for each plot from which repeated collections were taken. All ovaries with an atypical number of styles were preserved in formol-acetic-acid-alcohol as evidence of collection and for histological study.

Ovaries of most (66.9 percent) of the 21,669 flowers examined bore the typical five styles; style numbers for the remainder (33.1 percent) ranged as follows: 0, 1, 2, 3, 4, 6, 7, 8, 9, and 10 styles per ovary (Table 1). The number of carpels, placentae, and placental vascular strands of a given ovary, determined from paraffin sections, varied directly with the number of styles it bore, except for styleless ovaries and those with certain other irregularities.

Of 18 styleless ovaries sectioned, carpels ranging in number from three to seven, inclusive, were found. Similarly, the number of styles of a given ovary did not correspond always to the actual number of carpels. For example, ovaries with one to four styles, inclusive, could be expected to have, correspondingly, one to four carpels. However, any one of these might actually have five to eight (possibly more) carpels; this suggested that one or more styles of a particular ovary had failed to develop.

The typical, young, five-styled ovary of *Lychnis alba* has axile placentation with five carpellary septa connecting the ovary wall to the columnar placental region. Subsequently these septa break, and the whole central region (actually containing five placentae) thus detached

from the ovary wall becomes the free-central placenta.

Except for stated irregularities, one-styled ovaries have one carpel, one locule, and one marginal placenta. Two-styled ovaries have two carpels and two locules, the placenta becoming free-central when the two oppositely placed carpellary septa break. Ovaries with three to ten styles, inclusive, develop typically.

The placental column of a typical five-styled, five-carpellate ovary has five prominent, equally spaced, radially prolonged vascular strands (appearing starlike in placenta cross sections) extending platelike throughout its length. These vascular strands vary directly with the number of carpels and placentae but not always with the number of styles of a given ovary.

Ovaries of *Lychnis alba* may have more carpels, placentae, and vascular strands than styles, but the reverse situation was not found.

Significant variations in style number appeared in every larger collection of *Lychnis alba* flowers examined during 1958; smaller collections sometimes produced fewer variations, most (and occasionally all) ovaries bearing five styles. Twenty-nine individual plants observed daily in a greenhouse throughout their entire flowering period from October 1958 through January 1959 exhibited a similar variance (8). Twenty-five of these produced ovaries bearing significant variations in style number (four to nine, inclusive); one was an androhermaphrodite; in one most ovaries bore five styles; and in two all ovaries bore five styles, strictly.

In this respect, one plot of *Lychnis alba* (West Liberty plot No. 5) proved especially interesting because the most complete range of variation in style

numbers found in any one plot studied during 1958 (zero to ten, inclusive) occurred in the 4142 flowers collected from it. Two hundred and forty-one gynohermaphrodite flowers with styles ranging in number from three to eight, inclusive, were likewise found in this plot.

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#### References and Notes

1. M. L. Fernald, *Gray's Manual of Botany* (American Book Co., New York, ed. 8, 1950).
2. N. L. Britton and A. Brown, *Illustrated Flora of the Northern States and Canada* (New York Botanical Garden, New York, 1923).
3. G. Bentham and J. D. Hooker, *Genera Plantarum* (London, 1862), vol. 1.
4. M. Westergaard, *Dansk. Botan. Ark.* 10, 1 (1940).
5. H. E. Warmke and A. F. Blakeslee, *Am. J. Botany* 27, 751 (1940).
6. H. E. Warmke, *ibid.* 33, 648 (1946).
7. The plants were grown from seeds collected by Theodore VanBruggen near Indianola, Iowa, 21 June 1957.
8. The plants were grown from seeds collected from West Liberty plot No. 5 near West Liberty, Iowa, 23 July 1958.
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#### Incidence of Sex Chromatin in *Gallus domesticus*

**Abstract.** The presence of so-called sex chromatin has been demonstrated in the interphase nuclei of the cells of the domestic chicken (*Gallus domesticus*). Definite sex dimorphism was observed for the incidence of this nuclear component; the frequency of its occurrence in females was at least ten times that of its occurrence in the males, ranging from 22 percent in the duodenal muscle cells to 52 percent in the epidermal cells of a growing feather.

Considerable corroborative evidence for the existence of sex chromatin in mammals has been accumulating since Barr and Bertram (1) first drew attention to the "nuclear satellite" and its possible usefulness in establishing the genetic sex of an individual. The term *nuclear satellite* was subsequently (2) changed to *sex chromatin*. All the reports to date, of which those of Cantwell *et al.* (3) and of Osuchowska and Suminski (4) are among the latest, clearly indicate the sex-dimorphic nature of the distribution of sex chromatin. In mammals, it is the cells of genetic females that are richly provided with this strongly basophilic, Feulgen-positive material. In males, sex chromatin is either nonexistent or occurs only infrequently. One is not surprised, therefore, to find a generalization, suggested by Prince *et al.* (5), that sex chromatin is the heterochromatic portion of the 2 X chromosomes. More recently Klinger (6) came to the same conclusion. It should be recalled in this connection

Table 1. Data indicating the total number of *Lychnis alba* flowers collected from each general area, the spread of variations in style number, the number of examples and totals for each category of variation, and the grand total of all flowers examined during 1958.

| No. of styles | No. of flowers                      |                                |                               | Total No. of flowers of given style No. from the three areas |
|---------------|-------------------------------------|--------------------------------|-------------------------------|--|
|               | Iowa greenhouse, 19 May–23 June (7) | Iowa outdoors, 20 June–23 July | Minnesota outdoors, 1–27 Aug. |  |
| 0             | 0                                   | 8                              | 24                            | 32   |
| 1             | 0                                   | 13                             | 14                            | 27   |
| 2             | 0                                   | 13                             | 31                            | 44   |
| 3             | 4                                   | 46                             | 88                            | 138  |
| 4             | 149                                 | 218                            | 391                           | 758  |
| 5             | 2,607                               | 4,967                          | 6,933                         | 14,507   |
| 6             | 317                                 | 1,766                          | 2,454                         | 4,537  |
| 7             | 64                                  | 661                            | 712                           | 1,437  |
| 8             | 5                                   | 90                             | 68                            | 163  |
| 9             | 1                                   | 13                             | 10                            | 24   |
| 10            | 0                                   | 2                              | 0                             | 2  |
| Totals        | 3,147                               | 7,797                          | 10,725                        | 21,669   |