dent, and the data substantiated the evidence obtained from the differences in their precipitation curves.

The multiplicity of antigens was also indicated by layering various concentrations of the β-lipoprotein preparations over antiserum in agar according to the Oudin procedure as described by Munoz and Becker (*11*). By this technique 5 bands were readily revealed in BS0 and 6 bands in BS1.

No γ-globulin could be detected in BS1 with rabbit antiserum vs human serum γ-globulin; BS1 contained about 0.2% human serum albumin as determined immunochemically (*10*). The rabbit antiserum vs β-lipoprotein revealed no demonstrable antibodies vs human serum albumin, human serum γ-globulin, or crystallized human serum β-metal-combining globulin (*12*). There was some cross reaction between rabbit anti-β-lipoprotein and the α-lipoprotein fraction, IV-1-1 (*1*), but, owing to the demonstrated heterogeneity of this fraction electrophoretically, this reaction could not be interpreted satisfactorily.

It has been found in this study, therefore, that the β-lipoprotein fraction of normal human plasma represents a class of β-lipoproteins differing in their immunochemical characteristics. Since several preparations of β-lipoprotein obtained from different pools of plasma were studied, the data suggest that the immunochemically reactive components were present in varying ratios in the individual plasmas. The data also indicate that normal human plasma contains a lipid-poor protein that is immunochemically related to the protein moiety of β-lipoprotein; the evidence is insufficient to state this relationship any more positively.

It has been suggested that definite distributions of physical heterogeneity of circulating lipoproteins as demonstrated by the ultracentrifuge is intimately associated with certain pathological states such as atherosclerosis (*6*). The relationships between immunochemical heterogeneity and physical heterogeneity await further investigation, but analysis of the data presented here suggests that immunochemical heterogeneity exists within the various flotation "classes."

The correlation between circulating lipoprotein and human disease is extremely interesting in view of the observation that β-lipoprotein is a component of the cell nuclei of almost all human tissues (*13*). Since the level of individual plasma proteins is a reflection of tissue metabolism, the finding of an altered pattern or level of a particular protein may be only a secondary factor in a disease state rather than a primary or etiologic factor. It is also true, however, that the altered level of the plasma protein, although secondary to tissue metabolism, can itself give rise to characteristic disease as evidenced in hemophilia, afibrinogenemia (*14*), and agammaglobulinemia (*15*). The possible relationships between tissue metabolism and hyperlipoproteinemia in disease await further study.

Kunkel (*16*) fractionated human serum with zephiran and ultracentrifugation and obtained a β-lipoprotein which proved antigenic in rabbits; the rabbit antiserum was then used to estimate lipoproteins in normal and pathological sera. For the estimation of an antigen immunochemically, it must be ascertained that the antigen is either immunochemically homogeneous or that the ratio of its reactive components remains constant. Although the β-lipoprotein employed as an antigen in this study was prepared differently from that of Kunkel, the basic immunochemical criteria for use of antiserum in quantitative estimations were not fulfilled for the β-lipoproteins. By proper absorption techniques, however, it may be entirely feasible to render such antisera specific.

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The Effect of Indoleacetic Acid and Amount of Solar Radiation on Heterosis in the Snapdragon (*Antirrhinum majus* L.)[1]

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In the course of studies with hybrid snapdragons *Antirrhinum majus* L., it became apparent that light was a factor in determining the degree of heterosis and the degree of reaction of the plants to indoleacetic acid. Heterosis is the difference between the mid-parent mean and F_1.

Since auxins are believed to be essential to cell elongation and since one of these substances, indoleacetic acid, is inactivated by riboflavin in the presence of light (*1*), it is possible that a difference in auxin level may account for the phenomenon of heterosis, and that these phenomena may be influenced by the amount of light received by the plant.

In order to test this general hypothesis, two inbred lines of *Antirrhinum majus* L. and their hybrid were chosen as the experimental test subjects. Three experiments were conducted under different light conditions to obtain data on height and dry weight of the parents and of the F_1.

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TABLE 1

AV HEIGHT AND DEGREE OF HETEROSIS OF 350 PLANTS EACH OF INBRED (P_1 AND P_2) AND THE HYBRID (F_1) AND SEGREGATING POPULATION (F_2) OF *Antirrhinum majus* GROWN UNDER DIFFERENT AMOUNTS OF LIGHT

Experiment	Total solar radiation (g cal.)	Parent 1 (P_1) height (cm)	Parent 2 (P_2) height (cm)	Mid-parent mean height (cm)	Hybrid (F_1) height (cm)	F_2 height (cm)	Heterosis* height (cm)
I†	5,512.9	7.3	12	9.65	10.38	7.91	0.73
II‡	13,433.8	5.9	8.93	7.415	10.1	9.9	2.685
III§	16,018.1	4.7	9.2	6.95	10.9	6.9	3.95

* Heterosis is the difference between mid-parent mean and F_1.
† Dec. 1, 1951, through Feb. 5, 1952.
‡ Feb. 3, 1952, through Apr. 6, 1952.
§ Mar. 28, 1952, through May 8, 1952.

In order to obtain plants grown under low amounts of light, seeds were sown on December 1, 1951, and 350 seedlings in each group were transplanted 45 days later.

In another trial, under higher amounts of light, seeds were sown on February 2, 1952, and 350 seedlings each (P_1P_2 and F_1) were transplanted on March 28, 1952. In the final test, under still higher amounts of light, seeds were sown on March 28, 1952, and 350 seedlings from each parent were transplanted on April 19, 1952.

The seedlings of the inbreds (P_1 and P_2) and their hybrid (F_1) were grown on a greenhouse bench containing a suitable soil mixture and maintained under uniform greenhouse conditions. Height measurements were made as soon as the tallest population approached 10 cm. Dry weights were recorded after the response to indoleacetic acid was noted.

It was consistently found that heterosis, as detected by these two criteria, increased with seasonal increase in solar radiation as shown in Tables 1 and 2; for example, the degree of heterosis, as measured by dry weight in g/plant, increased from 0.115 under low amounts of light to 0.46 under higher amounts of light.

The plants in the three experiments were sprayed with indoleacetic acid after height measurements were recorded.

Plants grown from December through February under low light conditions were sprayed with indoleacetic acid at 100 ppm and exhibited typical epinasty. However, it was found that on plants grown from March through May, when light intensities were high, a stronger concentration of indoleacetic acid was needed to produce the same degree of response. Concentrations of 1000 ppm were used in the later two trials. Heterotic effect was more evident in the consistently longer and more intense response of the F_1 than in either parent.

TABLE 2

AV DRY WEIGHT IN GRAMS AND DEGREE OF HETEROSIS OF INDIVIDUAL PLANTS OF PARENT 1 (P_1), PARENT 2 (P_2), AND HYBRID (F_1)
(Individual averages based on the av of 350 plants)

Experiment	Parent 1 (P_1)	Parent 2 (P_2)	Mid-parent mean	Hybrid (F_1)	Heterosis*
I†	0.216	0.357	0.286	0.401	0.115
II‡	0.231	0.206	0.216	0.570	0.354
III§	0.12	0.16	0.14	0.6	0.46

* Heterosis is the difference of the F_1 from the mid-parent mean.
† Dec. 1, 1951, through Feb. 5, 1952.
‡ Feb. 3, 1952, through Apr. 6, 1952.
§ Mar. 28, 1952, through May 8, 1952.

TABLE 3

EFFECT OF A SIMULTANEOUS APPLICATION OF AQUEOUS EXTRACTS OF EITHER P_1 OR P_2 OR F_1 AND INDOLEACETIC ACID AT 25 PPM ON ROOT GROWTH OF CUCUMBER SEEDLINGS
(Seedlings germinated in dark)

Treatment	I Root growth in mm av 16 seedlings	II Root growth in mm av 16 seedlings	Av
Distilled water	5.8	5.5	5.2
IAA*	0.49	1.0	0.74
Parent 1 + 25 ppm IAA*	1.95	2.33	2.14
Parent 2 + 25 ppm IAA*	3.7	4.3	4.00
Hybrid + 25 ppm IAA*	0.47	0.56	0.51

* Indoleacetic acid.

Because the F_1 showed a greater sensitivity to indoleacetic acid, a biological test was employed in an attempt to prove the hypothesis that the F_1 differed from its parents in an indoleacetic-acid-inhibiting or -inactivating mechanism. Alamercery (*2*) has shown that cucumber seedlings are very sensitive to indoleacetic acid. He found that the elongation of hypocotyls and roots is in proportion to the concentration of indoleacetic acid. Accordingly, cucumber seeds were germinated in a distilled water solution of 25 ppm of indoleactic acid with and without a tissue extract of both parents and hybrid snapdragons. The tissue extract was prepared by using a 10-g sample of fresh leaves and stems from the P_1, P_2, and F_1 plants when the F_1 was approximately 10 cm tall. The plants were macerated in a Waring Blendor and made up to a volume of 100 cc. To this slurry indoleacetic acid was

added to make a final concentration of 25 ppm. Sixteen cucumber seeds were placed on a filter paper in a Petri dish and 5 cc of this solution was added to each dish. In addition, a distilled water solution of indoleacetic acid at 25 ppm and distilled water alone were used as controls. Each treatment was replicated five times. The cucumber seedlings were allowed to germinate at constant temperature in darkness. Four days after treatment root measurements were recorded.

It was repeatedly observed that the inhibitory effects of indoleacetic acid on cucumber roots were reduced much less by the extract from the F_1 than by the extracts from either parent. This may suggest that the parents inactivated indoleacetic acid more effectively than did their hybrid (Table 3).

The degree of heterosis in the experimental plants of *Antirrhinum majus* L. was greatly influenced by the amount of solar radiation. Heterotic ability of the F_1 to retain the indoleacetic acid has been demonstrated. The greater ability of the hybrids to retain and utilize growth substance under high light conditions permits greater expansion of plant tissue and thus gives the additional growth increment that can cumulatively result in heterosis.

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The Prevention of Dental Caries by Rock Phosphate in the Diet of the Rat

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The trend toward the fluoridation of communal water supplies offers little for caries prevention in areas where fluoridation of water is impracticable. As a means of supplying extra fluorine in a vehicle other than water, rock phosphate was chosen by us for testing because of its additional high calcium and phosphorus content.

The cariogenic diet, Diet I, which was used contained 800 g cracked yellow corn, 30 g alfalfa meal, 60 g linseed meal, 250 g sucrose, 80 g corn oil, 500 g active dry yeast, and 10 g sodium chloride.

All the caries prevention diets were the same and similar to Diet I except that they also contained Tennessee brown rock phosphate in varying amounts from 5 to 35 g. In several experiments, it was found that the addition of 15 g or less of rock phosphate had little or no caries prevention effect; but above 15 g, the caries prevention effect progressively increased until an optimum effect was reached with approximately 35 g. For this reason the results of only two diets are reported here, namely, Diet II which contained 20 g and Diet III which contained 34 g rock phosphate.

In these experiments 210 Wistar rats were used, one half of each litter being fed one of the diets. The rats were placed on these diets 20–22 days after birth and observed until they died naturally.

Of 105 rats on Diet I, caries appeared in 382 teeth or 61%, mostly lower molars; of 49 rats on Diet II (containing 20 g rock phosphate) caries appeared in 62 or 21% of the lower molars; of 56 rats on Diet III (containing 34 g rock phosphate) caries occurred in 19 or 5% of the lower molars (Table 1).

TABLE 1

DENTAL CARIES IN LITTER MATE RATS ON DIETS CONTAINING BROWN ROCK PHOSPHATE

Diets	No. of rats	Number carious lower molars	Percentage
Diet I	105	382	61
Diet II	49	62	21
Diet III	56	19	5

The upper incisors of the rats on the caries prevention diets were overgrown and curved. Growth lines were seen on many of these teeth. Calcification not only of the jaw bones, but also of the entire skeleton was better in the rats on Diets II and III than in rats on Diet I. The death rate of the rats on the caries prevention diets was normal whereas all the rats on Diet I died between the 4th and 7th months of age.

The rock phosphate used in these diets contains 5.9% calcium fluoride. The concentration of fluorine in Diet II was approximately 350 ppm and in Diet III, it was 700 ppm. Compared to 1–2 ppm of sodium fluoride recommended for use in communal water supplies, this seems like an extraordinarily excessive amount of dietary fluorine for the prevention of caries. But in the rat, it has been established in numerous experiments summarized by Hodge and Sognnaes (*1*) that at least 125–200 ppm of the more soluble sodium salt of fluorine must be present in the rat's diet before an appreciable caries resistant effect can be obtained. Miller (*2*) used 500 ppm dietary calcium fluoride in order to prevent dental caries in the rat. The fluorapatite used in our experimental caries prevention diets is far more insoluble than sodium fluoride, but we preferred to use fluorine in this form because of the extra benefit that might be derived from the additional calcium, phosphorus and other trace minerals present in brown rock phosphate.

Successful cariostasis in the posteruptive phase of dental development as recorded here already has been documented by McClure (*3*).

In experiments on humans, McClendon and Foster (*4*) each ingested daily 2.5 g of rock phosphate containing 75.5 mg of fluorine for three weeks and then reduced the daily intake for three years to 1 g rock phosphate containing 31 mg fluorine. Measuring the