

TABLE 2
VITAMIN ASSAY OF DRIED CHLORELLA

Vitamin	Pilot plant* sample	Laboratory† sample
Carotene, mg/lb	—	218.0
Thiamin, mg/lb	11.0	4.5
Riboflavin, mg/lb	26.2	16.3
Niacin, mg/lb	54.0	109.0
Pyridoxine, mg/lb	—	10.4
Pantothenic acid, mg/lb	3.6	9.1
Choline, mg/lb	—	1370.0
Biotin, µg/lb	—	67.0
Vitamin B ₁₂ , µg/lb	45.0	10.0
Lipoic acid, acetate u/mg	1.5	—

* Microbiological assay by J. J. Mayernik and David Hendlin, of Merck & Co., Jan. 1952.

† Grown by Stanford Research Institute in continuous dilution culture apparatus; assay by Curtis and Tompkins, San Francisco. Data taken from unpublished report from Stanford Research Institute to the Research Corporation of New York, March 22, 1950, by permission.

acid, and choline chloride (group 4) did not improve the results. Chicks fed the same diet without Chlorella (group 5) attained slightly heavier weights. The last group of chicks (group 6) was fed the complete broiler mash, which not only contained adequate levels of all nutrients, but also an antibiotic, thus accounting for the additional growth improvement obtained.

The apparent growth-depressing action resulting from the inclusion of 10% dried Chlorella to the adequate supplemental diet is believed to be due to the hygroscopic nature of the Chlorella. Chicks fed this level of dried Chlorella exhibited an impacted beak condition. This mechanical impediment interfered with feed consumption and, consequently, resulted in a slightly lower total weight at 4 weeks of age. Three other groups of chicks not shown in the table were also included in this study. These groups received 2½, 5, and 20% Chlorella, respectively. Impacted beaks also were noted in the chicks that received 2½ and 5% algae. The chicks that received the 20% level developed impacted beaks and beak deformities, so

TABLE 3
EFFECT OF DRIED CHLORELLA ON BODY WEIGHT AND
FEED EFFICIENCY OF CHICKS

Group No.	Treatment	Av wt at 4 wks (g)	G feed required/g gain
1	Basal ration	135 (13)	3.1
2	As 1 + 10% chlorella	262 (16)	2.4
3	As 1 + 10% chlorella + 0.1% DL-methionine	298 (16)	2.3
4	As 1 + 10% chlorella + 0.1% DL-methionine + vitamin mixture*	292 (16)	2.3
5	As 1 + 0.1% DL-methionine + vitamin mixture†	316 (16)	2.2
6	Complete broiler mash	342 (16)	2.2

* Numbers in parentheses refer to surviving chicks.

† Vitamin mixture supplied the following per lb of diet: 6 µg vitamin B₁₂, 2 mg riboflavin, 10 mg niacin, 2 mg calcium pantothenate, 150 mg choline chloride, and 2500 IU vitamin A.

that their feed consumption and growth rate were greatly lowered. The hygroscopic nature of the Chlorella used in this trial is not considered a serious handicap for its use in feeds since it is expected that suitable processing methods can readily be developed which will eliminate this problem. No other harmful effects were observed in the chicks fed Chlorella. The consistency and appearance of the chick droppings were normal.

Although a mechanical difficulty was observed in the use of the vacuum-dried algae, the results demonstrate clearly that dried Chlorella may serve as a source of certain dietary nutrients for the chick. From the nutritional analysis of this material and the growth data obtained, dried Chlorella seems to supply important quantities of carotene and certain B-complex vitamins. Additional work is required to establish further its nutritional value. This information, combined with a determination of the cost of producing Chlorella on a large scale, will help decide to what extent Chlorella may be considered a new food source. Even the present limited study indicates that it may well be considered as a potential food source in areas of limited agricultural resources.

References

1. SPOEHR, H. A., and MILNER, H. W. *Plant Physiol.*, **24**, 120 (1949).
2. MYERS, J., PHILLIPS, J. N., and GRAHAM, J. R. *Ibid.*, **26**, 539 (1951).
3. *Nutrition Revs.*, **9**, 347 (1951).

Manuscript received April 13, 1952.

Concerning the Ability of Homing Pigeons to Discriminate Patterns of Polarized Light

K. C. Montgomery and Eric G. Heinemann¹

*Department of Psychology,
Cornell University, Ithaca, New York*

The search for the sensory basis of bird navigation is apparently far from ended, despite the large number of investigations devoted to this problem (1-3). The demonstration by von Frisch (4) that honeybees can discriminate among patterns of polarized sky light, and that they apparently utilize such patterns as cues in their homing flights, suggests the possibility that migratory and homing birds possess a similar ability.² The present paper reports an experimental test of this possibility. If homing pigeons do utilize polarization patterns of sky light as cues in their flights, they should be able to discriminate readily between two visual stimulus patterns, one of which consists of light polarized in one plane and the other of light polarized in a plane orthogonal to the first.

The subjects were three homing pigeons, 1-2 years

¹ The senior author is now at Yale University; the junior, at Harvard University.

² This possibility was mentioned by Donald R. Griffin in a talk attended by one of the writers about two years ago. The writers wish to express their appreciation to Dr. Griffin for his interest and cooperation in the present study.

old.³ The apparatus consisted of an unpainted box 12" × 12" × 14". The front panel of the box contained a ground-glass stimulus key 1" in diameter and a food-delivery magazine placed directly underneath the key. The key was illuminated from behind by virtually homogeneous blue light obtained by passing light from a mercury lamp through Corning filters Nos. 5113 and 3389. The light was conducted to the key through a lightproof metal tube 9" long and 1½" in diameter. Four in. from the key the tube was interrupted by a sliding panel (stimulus slide) containing 6 openings, each of which was covered with a piece of polaroid. Three polaroids were oriented in the same plane; the other three were oriented in a plane at an angle of 90° from the first. Light polarized in one plane was made the positive (rewarded) stimulus; light polarized in the other plane, the negative (unrewarded) stimulus. Three positive and 3 negative stimuli were used in order to eliminate the possibility of the birds' establishing a discrimination on the basis of an irrelevant cue—e.g., distinctive marks on the polaroids.

The response measured was key-pecking. A Harvard Cumulative Recorder and an electric counter were used to measure pecking behavior. Purina laboratory chow for pigeons was employed as a food reward; before and during the experiment this chow was part of the birds' standard diet.

The first phase of the experiment consisted of gradually reducing the body weight of the birds, over a period of about 2 weeks, to 80–85% of normal. This weight was maintained throughout the rest of the experiment.

The second phase consisted of preliminary training in which the birds were (a) adapted to the experimental box, (b) trained to approach and to eat promptly from the food magazine when it was opened, and (c) trained to peck the unilluminated key for food reinforcement. Both auditory and visual stimuli signaled the opening of the food magazine—the sound of the food-delivery motor and the flashing on of a light placed immediately over the magazine.

In the third phase of the experiment each bird was given 6 hr of discrimination training. One 30-min session was given on each of 12 days. During each session fifteen 1-min presentations of the positive stimulus and fifteen 1-min presentations of the negative stimulus were made. The order of stimulus presentation was randomized, subject to the restriction that neither the positive nor the negative stimulus appeared more than twice in succession. The stimulus was changed every minute by moving the stimulus slide to a predetermined position. All responses to the positive stimulus were reinforced with food; no responses to the negative stimulus were reinforced. During this training cumulative responses were recorded graphically. Careful study of these records yields no evi-

dence that any of the birds formed a discrimination.

The fourth phase consisted of extinguishing the birds to a criterion of 10 min of no responding. The procedure was exactly the same as during discrimination training except that no responses were reinforced. The total number of pecks emitted during each minute of exposure to the positive and to the negative stimuli was obtained from counter readings. These extinction data provide an additional, and perhaps a more sensitive, test of whether the birds developed a discrimination between the two planes of polarized light. The results are uniformly negative. Table 1 presents the total number of pecking responses emitted under the positive and negative stimulus conditions by each bird for 5-min extinction periods. The only bird that gave any evidence of having formed a discrimination is pigeon 3. For this bird the number of responses performed during each 1-min presentation of the positive and of the negative stimulus was tabulated. A *t*-test was run on the differences between the means based on these totals. The value of *t* is 1.40, which falls within the .10 fiducial limits. Thus, neither the acquisition nor the extinction data provide any evidence that homing pigeons can discriminate between planes of polarized light.

In the final phase of the investigation two of the birds were trained on a simple brightness-discrimination problem. Exactly the same apparatus and procedure were used as before except for one apparatus change: a second piece of polaroid was placed in the light-conducting tube and was so oriented that the positive stimulus was the same (except for a slight reduction in brightness) as in the original discrimination training and the negative stimulus consisted of very dim, if not the total absence of, illumination. One bird was given two 30-min sessions of training; the other, four 30-min sessions. This procedure provides a check on whether the birds (a) could "see" (i.e., learn to respond to) the discriminative stimulus provided by the blue light, and (b) were "intelligent" (i.e., capable of forming a simple discrimination). The results are summarized in Table 2. These data indicate clearly that both birds learned the brightness discrimination very rapidly, and that they were capable of responding to the blue light.

The results do not support the hypothesis that hom-

TABLE 1

5-Min period	Subject							
	3				4			
	+	-	+	-	+	-	+	-
1	127	88	8	29	91	79	66	84
2	88	68	9	5	69	81	61	42
3	51	58	24	19	0	3	47	57
4	70	67	12	6	0	0	80	58
5	44	45	21	16			4	7
6	56	20	2	11			0	0
7	56	39	0	0				
Total			568	470	160	163	258	248

³ Grateful appreciation is expressed to Otto Meyer, chief of the Pigeon Breeding and Training Center, Fort Monmouth, N. J., who lent the pigeons to us. The birds are carrier pigeons of the kind bred, trained, and used by the Signal Corps.

TABLE 2

5-Min period	Subject					
	3		6			
	+	-	+	-	+	-
1	170	54	95	50	68	13
2	187	82	62	3	109	30
3	206	54	51	1	102	32
4	160	43	88	47	84	2
5	167	7	87	25	117	2
6	158	12	79	40	79	0
Total	1048	252			1021	245

ing pigeons can discriminate among planes of polarized light. It is evident, however, that they can form a simple brightness discrimination very rapidly. Hence, it is concluded (a) that if homing pigeons can discriminate at all among patterns of polarized light, they can do so only with extreme difficulty, and (b) that it is highly unlikely that homing pigeons make use of patterns of polarized sky light as cues in their homing flights.

References

1. GORDON, D. A. *Science*, **108**, 710 (1948).
2. GRIFFIN, D. R. *Quart. Rev. Biol.*, **19**, 15 (1944).
3. WILKINSON, D. H. *Proc. Linn. Soc. London*, **160**, 94 (1949).
4. VON FRISCH, K. *Bees*. Ithaca, N. Y.: Cornell Univ. Press (1950).

Manuscript received April 25, 1952.

Extraction of Adrenal Cortex Hormone Activity from Placental Tissue

Richard H. Johnson and William J. Haines¹

Research Laboratories,
The Upjohn Company, Kalamazoo, Michigan

Several kinds of mammalian tissue have been examined in our laboratories in an effort to find sources of naturally occurring adrenal cortex hormones other than adrenal tissue itself. As a result of such studies, evidence has been obtained for the existence of this type of biological activity in human and equine placental tissue.

The investigation of the adrenal hormone content of human placentas² was initiated in conjunction with our preparation of crude placental extracts for studies by William H. Pearlman, of the Jefferson Medical College. A report on the latter work has recently appeared (1). In subsequent experiments, the placentas were collected at a local hospital, frozen quickly in solid CO₂, and processed by a standard procedure for isolating adrenal cortex hormones (2, 3). The resulting "neutral hormone concentrates" were analyzed for their content of adrenal cortex hormone activity.³

¹ We wish to acknowledge the helpful interest shown in this work by M. H. Kuizenga.

² We are indebted to Richard H. Barnes, of Sharp & Dohme, Glenolden, Pa., for the human placentas used in the preliminary investigation.

³ We are indebted to K. J. Olson, R. O. Stafford, and D. F. Kiel for the bio-assay data reported in this paper.

Two different methods of bio-assay were employed: the rat liver glycogen deposition test (4) and the rat survival growth test (5). The resulting data are presented in Table 1.

TABLE 1
ADRENAL CORTX HORMONE CONTENT OF EXTRACTS
OF HUMAN PLACENTAL TISSUE

Lot No.	Bio-assay procedure			
	Glycogen deposition* (u/kg tissue)	No. rats	Survival growth† (u/kg tissue)	No. rats
1	4.5	10	17.5	18
2	1.4	10	—	—
3	3.0	3	—	—

* One unit in the rat liver glycogen deposition test is the amount of bio-activity equivalent to 0.1 mg 17-hydroxycorticosterone (Kendall's compound F, hydrocortisone).

† One unit in the rat survival growth test is the amount of bio-activity that will cause 80% of the test animals to survive for 20 days and grow at an average rate of 1 g/day.

Unmistakable evidence for the existence of adrenal cortexlike hormone activity was observed for each of three individual lots of human placental tissue. It was of particular interest that the ratio of the two types of adrenal hormone activity observed for Lot 1 was approximately 4 survival growth units/glycogen deposition unit. This is essentially the same ratio of bio-activity that is observed for extracts of adrenal tissue, which indicates some similarity in the qualitative character of the extracts of these two tissues. The yield of bio-activity from human placental tissue, however, which was observed to vary from 1.4 to 4.5 units of glycogen deposition activity/kg in these experiments, is much less than that from adrenal tissue. It has been found in these laboratories that the usual recovery of this type of activity is about 25 u/kg from beef adrenals and 50-100 u/kg from hog adrenals.

Studies similar to those just described were also done using equine placentas. The two methods of bio-assay utilized in these experiments were: the rat liver glycogen deposition test and a modification of a test described by Grollman (6), involving the measurement of the gain in weight of adrenalectomized weanling rats. The yields of adrenal hormone activity from three individual lots of tissue are presented in Table 2, where it is seen that the glycogen deposition activity varied from 3.0 to 17.0 u/kg of tissue. A ratio of approximately 90 weight gain u/glycogen deposition unit was obtained in the case of Lot 1. Here, again, the ratio of these two types of bio-activity is in the general range of that observed for extracts of adrenal tissue. It is of interest that the yield of total hormone activity from equine placental tissue is about the same as from human placental tissue and thus much less than the yield from adrenal tissue itself.

The small amount of active material that has been obtained to date from placental tissue has prevented the characterization of the individual hormone com-