Santa Barbara, Calif. The Sandia projectile points have often been compared with the Solutrean points of France, but Hibben (*5*, p. 24) is right when he says: "This similarity to Solutrean material does not imply connection or contemporaneity with it, although the resemblance is remarkable." The Solutrean in France falls in a late period of the Hauptwürm stadial after 24,000 years before the present.

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Heterogeneity of Nuclear Ribonucleic Acid

There is an increasing body of evidence which supports the hypothesis that the ribonucleic acid (RNA) which is synthesized in the nucleus acts as a medium for the transfer of genetic information to centers of protein synthesis in the cytoplasm. Correlatively there has developed the concept that cytoplasmic RNA is derived from the nucleus (*1*, *2*). At the same time, there are at least two observations which do not appear to be compatible with such a hypothesis: (i) observed differences in the quantities of the bases in nuclear and cytoplasmic RNA isolated from the same cell type (*3*), and (ii) the differential incorporation of labeled precursor into nuclear and cytoplasmic RNA (*2*). The demonstration of heterogeneous populations of RNA molecules in the nucleus and cytoplasm would help to resolve these incompatibilities (*4*). In the experiments described here, the nuclear RNA of the starfish oocyte has been separated into two fractions which have striking differences in metabolic activity.

Starfish oocytes *in ovarii* were placed in sea water containing approximately 0.5 μc of P^{32} per milliliter. At the times indicated in Fig. 1, about 80 g of ovarian material was removed, and the nucleoli were isolated in sucrose solutions (*5*, *6*). The phosphorus-containing compounds were separated by a modification of the procedure described by myself (*6*) as follows: The "soluble P" fraction was removed by extracting with 0.1*N* HCl for 15 minutes at 0°C. The P found in this fraction was organic phosphorus, presumably in nucleotides. The precipitate was then extracted in hot alcohol-ether (3/1) by refluxing for 1 hour. "RNA_1" was separated by allowing the precipitate from the fractions to stand for 1 hour at 0°C in 1*N* HCl. This fraction contained the typical bases of nucleolar RNA as well as ribose and phosphorus in the proportions expected in RNA. "RNA_2" was extracted by heating the remaining precipitate in 1*N* HCl for 20 minutes at 90°C. The residue from these extractions contained only protein-bound P. "Cytoplasmic RNA" was separated by the same procedure except for the omission of cold 1*N* HCl extraction. The cytoplasm was prepared as an acetone-dried powder from the ovarian homogenate after the initial removal of nuclei and nucleoli by centrifugation (*5*). The extracts were plated and counted to 5-percent reproducibility. Phosphorus was determined by the method of Wiame (*7*), and by calculation from ultraviolet absorption at 260 mμ. Agreement between duplicate P determinations and ultraviolet-calculated P in the RNA fractions was within 8 percent.

As is readily seen from Fig. 1, RNA_1 and RNA_2, which differ in their solubility in cold 1*N* acid, demonstrate striking differences in specific activity with respect to time. Because the amount of the total nuclear RNA which is found in RNA_1 is small (*8*), the possibility exists that the early, high specific activity of this fraction may be the result of contamination. Although this possibility cannot be unequivocally denied, it appears to be unlikely on the basis of the following considerations. (i) At the time that the specific activity of RNA_1 is leveling off (5 to 8 hours), the specific activities of the other fractions are increasing at essentially linear rates. (ii) Agreement between the P found by direct analysis and that found by calculation from ultraviolet absorption is good. (iii) The prior exhaustive extraction of the nucleoli with cold dilute acid and alcohol-ether would remove compounds of low molecular weight which might be expected to contain phosphorus of high specific activity. (iv) When the RNA_1 fraction was submitted to paper electrophoresis, no radioactivity was found which was not associated with regions which were identified as nucleotides.

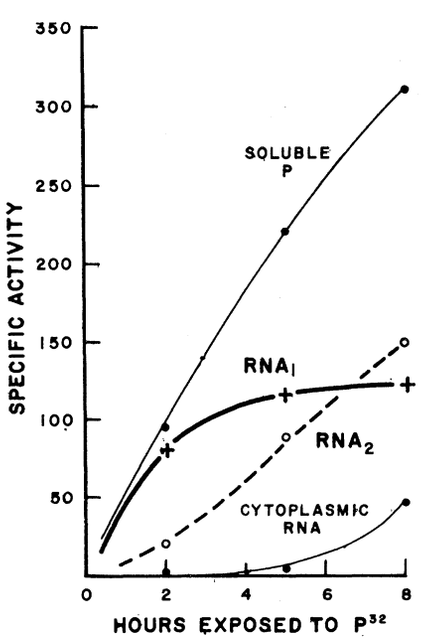

Fig. 1. Specific activity of various fractions of starfish oocytes calculated as counts per second per milligram of phosphorus.

The changes in specific activity with time of the RNA_1 fraction suggest, therefore, that this is a small, very active compartment which is saturated with labeled precursor in 5 hours. After this time, the radiophosphorus which is being incorporated into this pool must be disbursed as rapidly as it is incorporated. Two suggestions can be made about the fate of this material at this time: (i) it acts as a precursor for the RNA_2 fraction; and (ii) it is used in some other site in the cells. These alternatives are the subject of current investigations.

The experiments reported here serve to complement the earlier suggestions (*9*) that nuclear RNA must contain at least two populations of molecules which differ in their metabolic properties (*10*).

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4. A more complete analysis of the theoretical aspects of the interrelationships between nuclear and cytoplasmic RNA will appear in a paper by M. Ycas [*Proceedings of the Oak Ridge Conference on Applications of Information Theory to Radiobiology and Medicine* (Pergamon Press, New York, in press)].
5. The isolation procedure was modified from the technique of Schneider and Petermann [R. M. Schneider and M. L. Petermann, *Cancer Research* 10, 751 (1950)]. I am indebted to Alfred Marshak for his suggestions in using this medium for the isolation of oocyte nucleoli. (i) Ovaries were removed from starfish, rinsed in filtered sea water, and

then in 0.25*M* sucrose in deionized water made to 0.0018*M* with $CaCl_2$ (Ca-sucrose solution). The ovaries were allowed to stand for 20 min in 5 volumes of the Ca-sucrose, drained and homogenized in a Waring Blendor at slow speed (35 v) for 1 min in 4 volumes of Ca-sucrose. Four drops of caprylic alcohol were added to break the very stiff foam. (ii) The homogenate was filtered through two, and then four layers of coarse cheesecloth (20 by 30 mesh). The filters were washed with Ca-sucrose, and the washings were added to the filtrate to make 700 ml. The filtrate was centrifuged for 5 min at 150 *g* in 100-ml Lusteroid centrifuge tubes. (iii) The supernatant was discarded or saved for the isolation of cytoplasmic fractions, and the precipitate was resuspended in Ca-sucrose and filtered through eight layers of 40- by 40-mesh cheesecloth. The filtrate and washings were made up to 400-ml volume and centrifuged for 5 min at 150 *g*. (iv) The supernatant was discarded, and the precipitate was resuspended in 400 ml of 0.34*M* sucrose in ion-free water and centrifuged for 5 min at 150 *g*. The supernatant was discarded, and the precipitates were combined, made up to 100-ml volume with 0.34*M* sucrose solution, and centrifuged for 5 min at 150 *g*. (v) The supernatant was discarded, and the precipitate made up to 15-ml volume with 0.34*M* sucrose, then the nucleoli were concentrated, and the final contaminants were removed as described by Vincent (*6*). For nucleic acid and phosphorus studies, the sucrose was removed from the final pellet by two washes each in 70 percent, 95 percent, and absolute alcohol. The washed nucleoli were then vacuum dried and stored at – 20°C until used. All procedures were carried out at 2° to 4°C. The average time for isolation was 2 hours. The average yield has been about 1 mg of dry nucleoli from 10 g (wet weight) of ovaries.

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10. These studies were supported in part by AEC grant AT(30-1)1343.

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Table 1. Homing performance of displaced bats.

Distance (mi)	Number released	Percentage that returned at speed (mi/hr) indicated*					
		> 15	10–15	7–10	5–7	< 5	lost†
				Experiment A, 20–21 Oct.			
5	59			3.4	6.8	33.9	55.9
10	58			1.7	13.8	25.9	58.6
15	57			3.5	17.5	7	71.9
20	60		1.7	6.7	8.3		83.3
				Experiment B, 23–24 Oct.			
30	50	4	2	12	10	8	64
40	50		8	8	6		78
50	50			4	4		92
60	50	2	2	2			94
				Experiment C, 23–24 Oct.			
Blindfolded	25‡		7.2	21.6	21.6	21.6	28.1
Controls	25		12	32	8	12	36

* The figures in the last two columns of experiment A and in the last three columns of experiment B were variously influenced by inequalities in the time that was available for returning before the termination of the experiment. Therefore, they should not be used in comparing the performances of the various distance-groups.

† Netting was terminated at dawn on the first night. Any animals that did not return on the first night were designated lost.

‡ Of the 25 blindfolded bats that were released, 18 were recovered at the mine entrance, and, of these, eight had scratched the covering from one or both eyes. Figures in the table are based on the ten bats which were still blindfolded on their return, multiplied by a factor of 25/18 to render the figures directly comparable with the return percentages of the other experiments.

Homing in Bats

Homing ability has been demonstrated in a variety of terrestrial vertebrates. Of the many theories advanced for explaining the sensory basis of this phenomenon, only sun compass orientation has survived critical experimentation (*1*). The navigational problems of homing generally involve more than compass orientation, however (*2*). Matthews (*3*) has proposed a complete celestial navigation theory, based on vision, for certain birds, but others have questioned his interpretations (*2, 4*). Lindenlaub (*5*) has recently demonstrated definite homing tendencies in mice in an apparatus which eliminated any possibility of visual or olfactory clues.

A survey of the literature on homing in bats is given by Cockrum (*6*). Past work has consisted largely of displacing bats from a cave and then attempting to find them in the intricacies of the cave some days, or even months, later. This technique has yielded little information on the homing speeds or the percentage of bats that returned. In 1954 Cockrum released 68 bats (*Myotis velifer*) 28 mi from a cave in Arizona and recaptured two of these as they left the cave the following evening. On the assumption that these bats did not fly during the daytime, it appears that they must have traveled these 28 mi in something less than 4 hours.

During the fall of 1956 we performed a few simple homing experiments with bats which revealed a remarkably accurate navigational ability, even in blindfolded individuals (7). A total of 484 bats were removed from their hibernating roosts in an inactive lead mine in southwestern Wisconsin during the early hours of darkness on the nights of 20–21 and 23–24 October and transported to the east to points at distances that varied between 5 and 60 mi, for release. Distinctive markings were painted on the wings of the animals for each release point, and the animals were grouped in carrying cages for transport; all had fully recovered from the torpor of hibernation before release. Recaptures were made in a modified Japanese mist net as the bats entered the mine, and the exact time of each arrival was noted.

Homing performance was measured in terms of (i) the percentage of bats that returned to the mine during the same night, and (ii) the over-all homing speed of these returning animals, from the time of release to the time of entering the mine. Return records were obtained for all releases and percentages ranged from 6 percent in the 60-mi group to 72 percent in one of the 5-mi groups (Table 1). Homing speeds ranged up to 19 mi/hr and exceeded 10 mi/hr in 14 cases. Light tail-winds favored most of these animals (*8*), but the performance is regarded as indicating a rapid orientation and a direct homeward course, since flight speeds for bats of this genus have been measured as only 10 to 11 mi/hr (*9*).

Although vision is poorly developed in bats (*10*), its demonstrated importance for distance orientation in other animals indicated that a blindfolding experiment should be performed. Accordingly, 25 bats, blindfolded with eye caps of lampblack in collodion, were released, together with 25 controls, at a point 5 mi east of the mine. Although some of the blindfolded animals had scatched away all or part of their masks before reaching home, others retained theirs, and the fact that they reached home as rapidly as the controls did demonstrated that obstruction of vision had no gross effect on homing performance (Table 1C).

Since bats are able to detect obstacles in their flight path by echolocation (*10*), it is interesting to speculate on the possibility that they are aided by auditory mechanisms in distance orientation. Bats are also capable of learning landmarks and complex spatial patterns within a room by auditory means (*11*), and it is possible that whole landscapes may be memorized. However, evidence presented by Griffin (*12*) and Möhres (*13*) suggests that the maximum effective distance of echolocation for bats of this genus is less than 10 m, and it is difficult