At all passages of MSV-FeLLV in cat cells, cellular susceptibility was restricted to cat cells and produced no foci in mouse cells. The new, specific host range of the virus obtained by aggregation of MSV with FeLLV prompted attempts at reentry of MSV-FeLLV back into the murine cells by a second aggregation procedure now combining MSV-FeLLV and Moloney leukemia virus. The MLV (1×10^7 LVHU) was sedimented with 2×10^5 FFU of MSV-FeLLV in saline. The resulting interviral pellets were then used to infect mouse cells insusceptible to pure MSV-FeLLV. After 6 days of infection, typical foci were seen in Swiss mouse embryo cells, and viral progeny of these foci did not transform murine cells chronically infected with MLV. The sarcoma focus-forming genome apparently became reenveloped with an MLV coat.

The production of focus-forming MSV-FeLLV was readily reproducible by the centrifugation procedure. In contrast, attempts at transspecies rescue of defective MSV genome from MSV-induced hamster tumors, by a cultivation of cat cells, hamster tumor cells, and a superinfection with FeLLV were negative (*7*). Partial purification of MSV-FeLLV by sedimentation and by sucrose density gradient banding revealed that, at equilibrium, the peak of MSV-FeLLV focus-forming activity is localized where the buoyant density is 1.16 g/cm^3. The apparently easy crossing of the species barrier by the fusion techniques in vitro may have been facilitated by common antigens found between the mouse and the cat leukemia viruses (*8*). Indeed, we have on occasion observed that a coinfection of cat embryo cells with competent MSV and FeLLV could, after a second tissue culture passage, spontaneously produce limited amounts of MSV-FeLLV. The scope of this aggregation technique is under investigation with reference to crossing of other species barriers where appropriate C-type leukemogenic viruses are available. Modifications of this technique may eventually provide delivery systems for the introduction of various genetic messages into receptive cells.

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Bird Migration: Influence of Physiological State upon Celestial Orientation

Abstract. *By means of photoperiod manipulation, the physiological states of spring and autumn migratory readiness were induced in indigo buntings. The orientational tendencies of these two groups of birds were tested simultaneously in May 1968, under an artificial, spring planetarium sky. Birds in spring condition oriented northward; those in autumnal condition, southward. These results suggest that changes in the internal physiological state of the bird rather than differences in the external stimulus situation are responsible for the seasonal reversal of preferred migration direction in this species.*

Migration, by definition, refers to a two-way journey. For most birds residing in North Temperate areas, this migration consists of a southward trip each autumn followed by a northward return the following spring. Distances covered in such round-trip journeys are considerable, frequently exceeding 4000 to 6000 miles (6400 to 9600 km) (*1*).

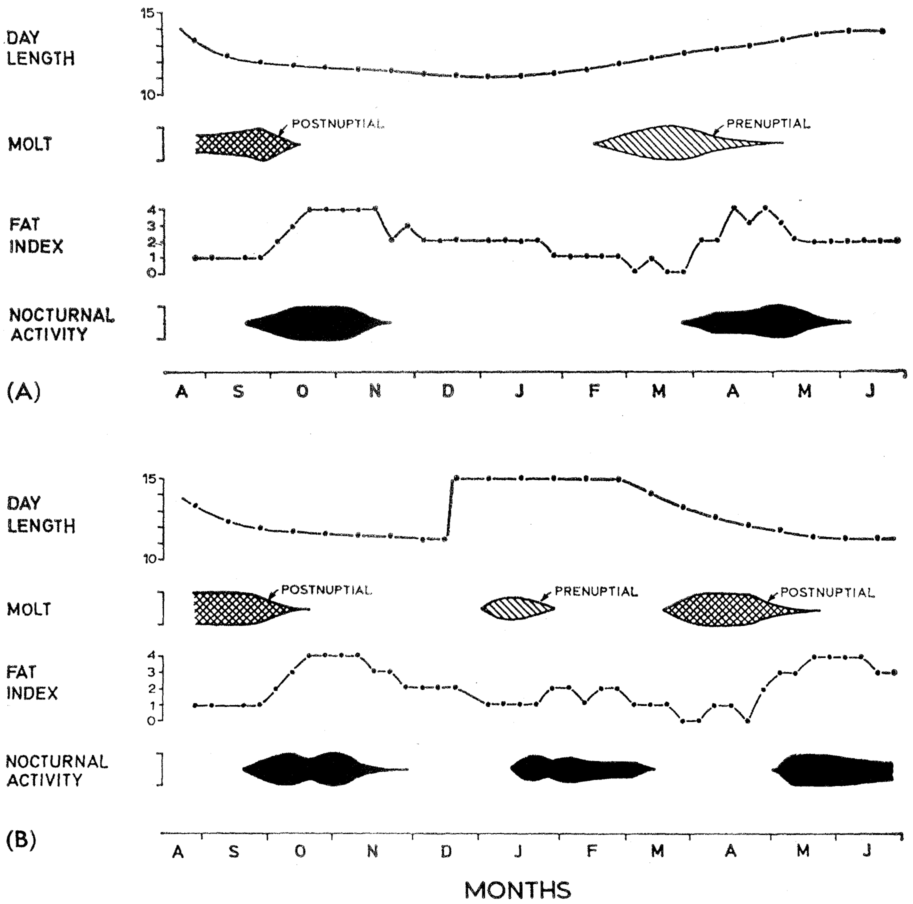

Fig. 1. The occurrence of molt, fat deposition, and nocturnal activity in indigo buntings (*7*). (A) Control birds on a natural photoperiod. (B) Experimental birds exposed to an accelerated photoperiod regimen.

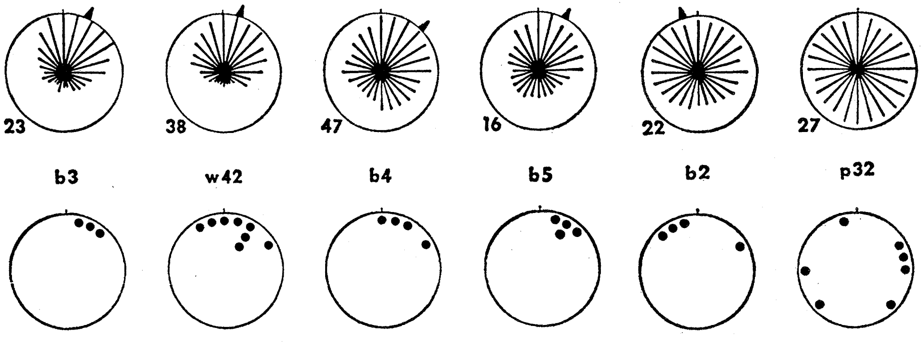

Fig. 2. Orientation of indigo buntings in spring migratory condition tested under a spring planetarium sky. (Upper) Vector summaries of total nocturnal activity. Arrows denote mean direction. (Lower) Mean directions adopted on different nights of testing. Vector diagrams are plotted such that the radius equals the greatest number of units of activity in any 15° sector. The number that this represents is presented at the lower left of each diagram. In all diagrams, 0° or 360° is north.

Bird migrants have been the subjects of intensive study by both physiologists and ethologists. Physiologists have studied hormonal and biochemical changes associated with the physiological state of migratory readiness, whereas ethologists have concentrated upon the navigational problems posed by these long-distance flights. Neither group has focused its attention upon the proximate factors underlying the seasonal reversal of preferred migration direction.

The similarity in the physiological events leading up to migratory departure (particularly the deposition of large quantities of subcutaneous fat and the appearance of migratory activity) has suggested that the internal state of migratory readiness is a general one which recurs in similar form twice each year (*2*). Experiments involving the hormonal induction of fat deposition and nocturnal restlessness support this view. Injections of prolactin and adrenocortical hormone produced these two effects in white-crowned sparrows *Zonotrichia leucophrys* in both vernal (photostimulatory) and autumnal (photorefractory) conditions (*3*). This type of study has led to the hypothesis that changes in internal physiology cause a migrant to become highly receptive to external stimuli that can serve as orientational cues, and that the seasonal reversal of migration direction is not dependent upon internal changes in the physiological state of the bird but rather upon seasonal changes in the external stimuli themselves (*4*).

Many birds which migrate at night are able to determine their direction from the stars (*5*). Because of the inequality of the solar and sidereal day, the temporal positions of stars change with the seasons, with the result that very different stellar information is available from an autumn night sky in contrast to that of a spring night sky. This raises the possibility that migrants might possess a specific northward directional response to the stellar stimuli of the spring night sky, and a different, southerly, response to the different stellar stimuli present in the autumn sky. Indeed, experiments performed on European warblers supported this hypothesis (*4*).

Experiments on indigo buntings *Passerina cyanea* suggested that a very different mechanism might be operative in this species (*6*). Directional tendencies of caged buntings were recorded when the birds were exposed to various correct and "incorrect" planetarium skies during the spring of 1965. These birds, which oriented in a generally northward direction under an artificial spring sky, continued to display northerly preferences under a sky advanced 12 hours from local time—a sky that normally would be present in the autumn. Thus, the visual stimuli typical of the opposite migration season failed to evoke any change in directional response.

Experiments designed to determine which star patterns are essential for orientation in buntings suggested a second dichotomy with European warblers. Evidence obtained by blocking various portions of the artificial sky from view suggested that the northern circumpolar area may be of special importance to orientation during both migration seasons (*6*). Although this finding requires confirmation, it raises the possibility that migrants may not use different cues during their northbound and southbound travels. This would indicate that it is not the external stimulus situation but rather some feature of the internal physiological state of the animal that dictates directional preferences.

To study this in further detail, I brought two groups of indigo buntings into spring and autumn migratory condition simultaneously by means of

Table 1. Orientation of indigo buntings in spring (control) and autumn (experimental) migratory condition. Vector analysis of the total nocturnal activity (upper row) and of the nightly mean headings (lower row) are presented for each bird. All tests were conducted in May 1968 under a spring planetarium sky (30°N); ISS, insufficient sample size.

Bird	Activity (hr : min)	Nights active (No.)	Units of activity (total *N*)	*P*	Mean direction	Mean angular deviation
			Control			
b3	11 : 50		288	<< .001	23°	66°
		3			ISS	
w42	20 : 15		540	<< .001	16°	66°
		8		<< .001	12°	31°
b4	11 : 15		828	.019	45°	75°
		4			ISS	
b5	8 : 45		246	.016	29°	73°
		4			ISS	
b2	15 : 5		413	.026	342°	76°
		4			ISS	
p32	20 : 40		581	Random (.4)		
		7		Random (.4)		
			Experimental			
w95	20 : 0		594	<< .001	146°	55°
		6		.007	144°	27°
b62	10 : 40		311	<< .001	153°	68°
		5		.007	147°	19°
b63	17 : 30		882	<< .001	161°	72°
		6		.007	156°	36°
p38	15 : 40		561	<< .001	207°	71°
		6		.003	202°	26°
b64	8 : 35		337	Random (.4)		
		3			ISS	

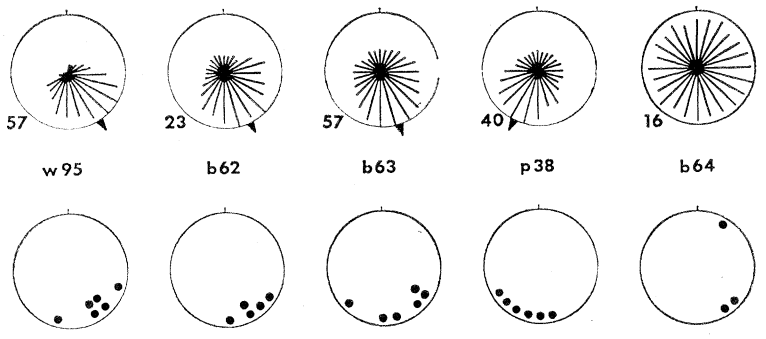

Fig. 3. Orientation of indigo buntings in autumn migratory condition tested under a spring planetarium sky. (Upper) Vector summaries of total nocturnal activity. (Lower) Nightly mean headings.

photoperiod manipulation. The directional preferences of both groups then were tested at the same time under identical planetarium skies.

Fifteen adult male indigo buntings were captured with mist nets in the vicinity of Ithaca, New York, during the autumn of 1967. They were housed in two identical 8 by 12 foot (2.4 by 3.6 m) flight rooms and given free access to food and water. Weight, fat level, and molt status were recorded weekly for each bird (*7*). Nocturnal activity was monitored continuously on an Esterline-Angus event recorder by means of microphone transducer.

As controls, eight birds were subjected to a photoperiod regimen simulating that encountered in nature. An astronomical-time clock maintained a day length equivalent to that on the wintering grounds at 18°N. These birds underwent normal, prenuptial molts between February and early April (Fig. 1). After this molt, large quantities of subcutaneous fat were deposited. Nocturnal activity first appeared in early April.

The experimental group of seven buntings was subjected to an accelerated photoperiod regimen. These birds were exposed to a long day length (15 hours light and 9 hours dark) after the termination of the refractory period in early December. This long day length was maintained until 1 March when the control of photoperiod was transferred to an astronomical timer set for 42°N and for a date 5 months advanced from local time. Hence, for example, the birds received the daylight equivalent of August on the breeding grounds during March. Each bird in this group initiated the prenuptial molt during the first 3 weeks of exposure to this long day length, but in all cases this molt was arrested before completion (*8*). During March and April these birds underwent normal postnuptial molts, attaining the brown winter plumage during late April. After this, fat deposition occurred, and nocturnal activity commenced.

As a consequence of this photoperiod manipulation, both groups of birds were available for experimentation during May of 1968. Both were in a state of migratory readiness, the controls in preparation for a spring flight, the experimentals for an autumnal flight.

The orientational tendencies of these birds were tested under an artificial planetarium sky mimicking that present at 30°N on 1 May (*9*). Each bird was placed in an individual, circular, funnel-shaped cage, and its directional preference was recorded by the "footprint technique" (*10*) (Figs. 2 and 3, Table 1). For each data distribution the null hypothesis of randomness was tested by the "v" modification of the Rayleigh test (*11*), with the prediction that control birds would orient northward and experimentals, southward. Mean direction and mean angular deviation were calculated by vector analysis (*12*). Four buntings failed to display nocturnal activity and hence could not be tested. Of the six active control birds, one was random while the five others each demonstrated a significant north to northeasterly tendency that was fairly consistent from night to night. One of the five active birds in the experimental group also failed to show any directional preference. Four birds, however, did display significant directional tendencies which ranged from southeast to south-southwest.

In short, the behavior of these two groups of birds differed markedly when tested simultaneously under the same planetarium sky. Individuals in spring condition oriented in a direction appropriate for spring migration, while those anticipating an autumnal migration took up directions appropriate for that season.

These findings indicate that important differences exist between the physiological states of migratory readiness in the spring and autumn. Results of planetarium blocking experiments mentioned earlier raised the possibility that the same celestial cues were used during both migration seasons. It thus seems plausible that differences in hormonal state may determine the manner in which an orientational cue will be used.

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7. Fat was recorded on an increasing 1 to 4 scale, according to C. M. Weise [*Ecology* **37**, 274 (1956)]. Molt was assessed as absent, slight, moderate, or heavy, according to the index of M. L. Morton and L. R. Mewaldt [*Physiol. Zool.* **35**, 237 (1962)].
8. Possibly the arrest of molt was caused by the rapid increase in gonadal hormones, stimulated by the long days.
9. The Cornell research planetarium is a 30-foot (9 m) diameter, air-supported dome equipped with a Spitz A-3-P star projector. Experimental cages are placed 3 feet off-center to minimize distortion of the artificial sky.

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