gray area of the midbrain (*17*), a region frequently associated with punishment and aversive behavior (*18*). Such considerations focus attention on serotonergic synapses in this central gray area as possible sites of action for the anxiety-reducing effects of benzodiazepines and related tranquilizers.

C. DAVID WISE
BARRY D. BERGER
LARRY STEIN

Wyeth Laboratories,
Philadelphia, Pennsylvania 19101

References and Notes

1. K. M. Taylor and R. Laverty, *Eur. J. Pharmacol.* **8**, 296 (1969).
2. H. Corrodi, K. Fuxe, T. Hökfelt, *ibid.* **1**, 363 (1967); H. Corrodi, K. Fuxe, P. Lidbrink, L. Olson, *Brain Res.* **29**, 1 (1971).
3. T. N. Chase, R. I. Katz, I. J. Kopin, *Neuropharmacology* **9**, 103 (1970).
4. B. B. Brodie and P. A. Shore, *Ann. N.Y. Acad. Sci.* **66**, 631 (1957); C. D. Wise, B. D. Berger, L. Stein, *Dis. Nerv. Syst. GWAN Suppl.* **31**, 34 (1970); K. Fuxe, T. Hökfelt, U. Ungerstedt, *Int. Rev. Neurobiol.* **13**, 93 (1970); S. S. Kety, in *The Neurosciences: Second Study Program*, F. O. Schmitt, Ed. (Rockefeller Univ. Press, New York, 1970), pp. 324–336; L. Stein, *J. Psychiat. Res.* **8**, 345 (1971).
5. C. D. Wise, B. D. Berger, L. Stein, *Biol. Psychiat.*, in press; L. Stein, C. D. Wise, B. D. Berger, in *Benzodiazepines*, S. Garattini, Ed. (Raven Press, New York, in press).
6. I. Geller and J. Seifter, *Psychopharmacologia* **1**, 482 (1960); I. Geller, J. T. Kulak, Jr., J. Seifter, *ibid.* **3**, 374 (1962); L. Cook and A. B. Davidson, in *Benzodiazepines*, S. Garattini, Ed. (Raven Press, New York, in press).
7. F. G. Graeff and R. I. Schoenfeld, *J. Pharmacol. Exp. Ther.* **173**, 277 (1970).
8. R. C. Robichaud and K. L. Sledge, *Life Sci.* **8**, 965 (1969); I. Geller and K. Blum, *Eur. J. Pharmacol.* **9**, 319 (1970).
9. M. H. Aprison and C. B. Ferster, *J. Neurochem.* **6**, 350 (1961).
10. C. D. Wise and L. Stein, *Science* **163**, 299 (1969).
11. W. P. Koella and J. Czicman, *Amer. J. Physiol.* **211**, 926 (1966).
12. F. F. Weight and G. G. Salmoiraghi, in *Advances in Pharmacology*, S. Garattini and P. A. Shore, Eds. (Academic Press, New York, 1968), pp. 395–413.
13. M. E. Goldberg, A. A. Manian, D. H. Efron, *Life Sci.* **6**, 481 (1967).
14. D. L. Margules and L. Stein, *Psychopharmacologia* **13**, 74 (1968).
15. If benzodiazepines act only to reduce the turnover of norepinephrine and serotonin, one would expect decreases rather than increases in the concentrations of their metabolites. However, it is probable that benzodiazepines also interfere with the transport of these metabolites out of the brain, as Chase *et al.* (*3*) have demonstrated for 5-hydroxyindoleacetic acid.
16. Although the reduction of serotonin turnover decreased slightly after repeated doses of benzodiazepines, the anxiety-reducing activity actually increases (*14*). Such increase may be due partly to progressive unmasking of the anxiety-reducing action as tolerance to the depressant action develops, and partly to supersensitivity to norepinephrine, which may develop as a result of drug-induced disuse of noradrenergic synapses.
17. G. K. Aghajanian and F. E. Bloom, *J. Pharmacol. Exp. Ther.* **156**, 23 (1967).
18. A. F. deMolina and R. W. Hunsperger, *J. Physiol. London* **160**, 200 (1962); J. Olds and M. E. Olds, *J. Comp. Neurol.* **120**, 259 (1963); L. Stein, *J. Comp. Physiol. Psychol.* **60**, 9 (1965).

19. We thank N. S. Buonato, W. J. Carmint, H. Morris, and A. T. Shropshire for technical assistance.

21 December 1971; revised 21 March 1972

Locomotion: Control by Positive-Feedback Optokinetic Responses

Abstract. *Several species of arthropods perform forward locomotory movements when restrained in place and exposed to a pattern of stripes moving backward at normal locomotory velocities. Locomotory effort varies directly with stripe velocity. In nature such locomotory reactions would increase the visual stimulus that elicits them; hence, the reactions represent a new class of optokinetic responses employing positive visual feedback. Stabilizing mechanisms include response decrement during constant stripe velocities.*

When an animal is placed within a rotating, vertically striped cylinder, it turns in the same direction as the cylinder (*1*). The stimulus, or input, for this optokinetic reaction is relative motion in the animal's visual field. The response, or output, is eye or body movement that tends to stabilize the stripes in the visual field. In terms of control theory (*2–4*), the output of this optokinetic system reduces the visual input, and hence the feedback is negative. In nature this response is presumed to help maintain an animal on a "desired" locomotory course despite external disturbances and bilateral asymmetries within the organism (*1*).

In contrast, we now describe a new class of optokinetic responses in which the visual feedback is positive. That is, the output (locomotory movement) increases the input (relative motion in the visual field), and thus the response is self-reinforcing. Our data suggest that positive-feedback optokinetic responses play an important role in controlling normal locomotory activity and that such responses are of widespread occurrence in the animal kingdom.

The positive-feedback optokinetic responses were elicited by exposing an animal to a pattern of stripes painted on the moving belt of a treadmill (Fig. 1A). Specimens were restrained in place above the treadmill and separated from it by a transparent Plexiglas platform; they could therefore see the moving stripes, but were unable to feel the movements of the belt. Lateral mirrors, mounted at an angle of 45°, reflected the ventral stripes to provide additional visual stimulation from both sides of the animal. The treadmill was bolted to a thick plate of stainless steel that rested on rubber feet within a Plexiglas aquarium. The aquarium was in turn enclosed within a Faraday cage that was supported on a table by vibration-proofed mounts. The treadmill was attached by a long, flexible drive shaft to a variable-speed electric motor outside the cage. During the experiments, which lasted from one to several hours, the animals were maintained at constant temperatures and viewed under constant illumination through a one-way mirror mounted in the front of the Faraday cage.

Species studied include the lobster (*Homarus americanus*), crayfish (*Procambarus clarkii*), locust (*Shistocerca vaga*), cockroach (*Periplaneta americana*), blowfly (*Sarcophaga bullata*), and sphinx moth (*Manduca sexta*). In all of these animals, movement of the stripes from front to rear induced forward locomotory movements [however, see (*5*)].

The detailed properties of the positive-feedback optokinetic responses have been examined in the lobster. Here, forward walking typically began within 1 or 2 seconds following the onset of stripe movement, increased in strength during the first few stepping cycles, and then showed marked decrement (Fig. 1, B and C). These responses could be eliminated by covering the moving belt with a stationary pattern of stripes, showing that the moving visual stimulus caused the responses. The optokinetic responses were often labile, but ones like those shown in Fig. 1 were obtained from the majority of the 35 lobsters used in this study.

The optokinetic responses involve more than one motor system, each recruited at a specific and relatively stable optokinetic threshold. In Fig. 1D, for example, walking movements were elicited at a treadmill speed of 2.5 cm/sec, while rhythmic, locomotory movements of the abdominal swimmerets were not initiated until the stripe velocity reached 11 cm/sec. Each time the treadmill speed was increased the responses of both locomotory systems increased and then decreased simultaneously to about the same degree. The decrement may have resulted from adaptation in the visual input common to both motor systems (*6*).

In the lobster not only are several motor systems involved in the optokinetic responses, but in addition the activity within a given motor system varied directly with the movement velocity of the stripes. In the walking system, for example, increasing the treadmill speed increased the stepping frequency (that

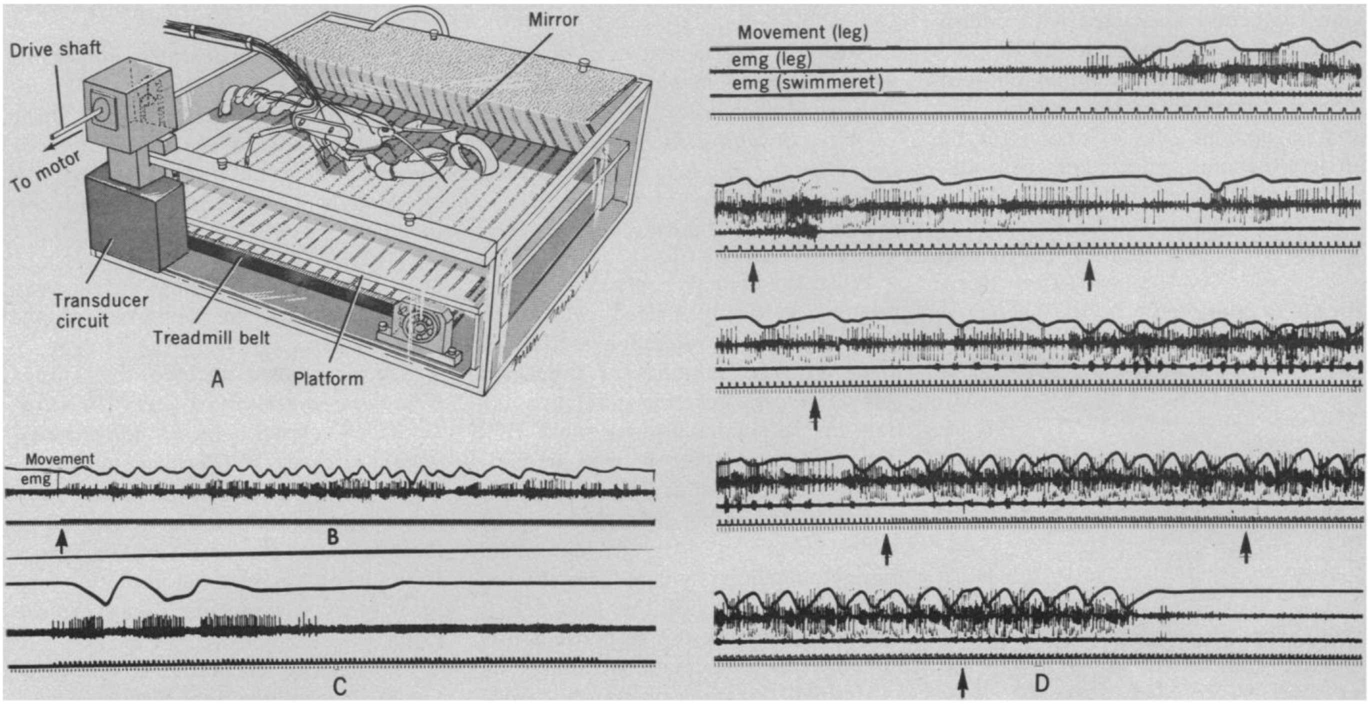

Fig. 1. (A) Treadmill apparatus with a lobster clamped in place above the striped belt and separated from it by a transparent platform of Plexiglas. The stripe width was 0.7 cm, well within the computed theoretical limit of resolution of the lobster eye (*10*). Movement of the belt was monitored by a photocell unit and an appropriate transducer circuit, calibrated to produce one pulse per centimeter of belt movement (see B to D). The mirror on the animal's right side has been removed for purposes of illustration. Wires attached to the carapace clamp are for recording limb movements and electromyograms. See text for further description. (B) Typical response of a walking leg following onset of treadmill movement (arrow), showing initial facilitation followed by decrement. (C) Rapidly diminishing response of a walking leg. (D) Response of a walking leg and swimmeret during stepwise increases in the treadmill velocity (arrows). In B to D the upper trace shows the movement of the ischiopodite of the right third walking leg, recorded with a linear movement transducer (*11*). Upward deflection corresponds to forward movement of the limb. In all records the lower trace shows the treadmill speed (above the continuous horizontal line; one vertical mark per centimeter of belt movement) and a time base (beneath the horizontal line; one vertical mark per 100 msec). Leg electromyograms (emg's) were recorded differentially with fine, Teflon-insulated silver wires implanted into the flexor and extensor muscles of the meropodite (B and D) or the flexor alone (C). Swimmeret emg's (D) were recorded from the powerstroke muscle of the right swimmeret on the third abdominal segment. The records in D are continuous.

is, decreased the stepping period) and increased the length of steps made by individual walking legs (Fig. 2). This finding eliminates the possibility that the locomotory activity was simply initiated by the visual stimulus and then maintained exclusively by proprioceptive feedback from the leg movements.

Many properties similar to those described above have also been identified in the optokinetic responses of the other arthropods that have been examined. In both the lobster and the crayfish, for example, the direction of attempted walking can be reversed by reversing the direction of movement of the stripes, and the walking rate is directly related to the treadmill speed (graphs similar to Fig. 2 have also been produced for the crayfish). In the blowfly, walking is elicited at low treadmill speeds (less than 3 cm/sec), while flying is induced at higher speeds (10 cm/sec).

What is the normal role of the demonstrated optokinetic responses? Forward locomotion normally causes relative motion from front to rear in the visual field. Our experiments demonstrate that front-to-rear motion in the visual field is itself an effective stimulus for forward locomotion. Thus, under natural, "closed loop" conditions, the visual feedback is positive and would tend to reinforce and sustain the locomotory activity once it is initiated.

Positive-feedback control systems are inherently unstable because they are subject to uncontrolled cascading, in which the output increases the input, which further increases the output, and so on (*2–4*). What prevents the positive-feedback optokinetic responses described here from causing uncontrolled, "runaway" locomotion? Four interrelated, limiting features can be identified. First, in the lobster, at least, the responses diminish after the first few stepping cycles (Fig. 1, B to D). Second,

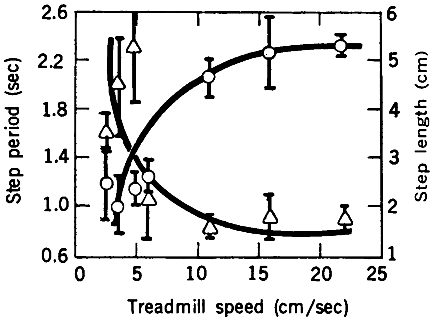

Fig. 2. Step period (triangles) and step length (circles) plotted against treadmill speed (that is, stripe velocity). Step period is defined as the time between positions of maximal limb protraction during walking, step length as the distance within a given walking cycle between the positions of the tip of the walking leg at maximal protraction and retraction. Both parameters were determined from data like those shown in Fig. 1, B to D. Each point on the graph is the average from all walking cycles during a 10-second period following an increment in treadmill speed (5 to 12 measurements per point). Vertical bars show standard deviation. All the data in the figure were obtained from a single, representative specimen.

both the inertia of the locomotory appendages and the resistance to their movement in a viscous medium are power functions of the velocity of limb movement. Such nonlinear mechanical constraints are presumably especially significant at higher velocities of locomotion. Third, under natural circumstances the optokinetic control system can probably be overridden by other sensory and central influences (*7*). Finally, the gain of the optokinetic control system, defined as the ratio of the output velocity (the speed of attempted locomotion) to the input velocity (the treadmill speed) and computed from the data presented in Fig. 2, is less than unity (*8*). Under such circumstances the output of a positive-feedback control system is stable (*4*).

We have demonstrated a new class of optokinetic responses, distinguished from previously known optokinetic responses by their utilization of positive rather than negative visual feedback. Our data show that within a given species the responses involve more than one motor system and are sufficiently strong to play a major role in sustaining and reinforcing normal locomotory behavior. Our studies on several species of arthropods, as well as suggestive findings of other investigators (*9*), indicate that positive-feedback optokinetic responses are of widespread occurrence in the animal kingdom.

WILLIAM J. DAVIS
JOSEPH L. AYERS, JR.
Division of Natural Sciences-I,
University of California,
Santa Cruz 95060

References and Notes

1. J. Loeb, *Forced Movements, Tropisms and Animal Conduct* (Lippincott, Philadelphia, 1918); W. Reichardt, Ed., *Processing of Optical Data by Organisms and by Machines* (Academic Press, New York, 1969).
2. L. E. Bayliss, *Living Control Systems* (Freeman, San Francisco, 1966).
3. J. H. Milsum, *Biological Control Systems Analysis* (McGraw-Hill, New York, 1966).
4. L. Stark, *Neurological Control Systems: Studies in Bioengineering* (Plenum, New York, 1968).
5. Smyth and Yurkiewicz report that moving the visual field from front to rear beneath the blowfly *Phaenicia sericata* (Meigen) suppresses the activity of flight muscles, a result opposite to ours [T. Smyth and W. J. Yurkiewicz, *Comp. Biochem. Physiol.* **17**, 1175 (1966)]. It is possible that under their experimental conditions the landing reflex, which involves a suppression of flight [L. J. Goodman, *J. Exp. Biol.* **37**, 854 (1960)], was unknowingly activated.
6. Adaptation in the visual system is known to contribute to behavioral habituation in other organisms, for example, the mollusk *Pleurobranchaea* [W. J. Davis and G. J. Mpitsos, *Z. Vergl. Physiol.* **75**, 207 (1971)].
7. To generalize the second and third arguments, the positive feedback which we have demonstrated may occur within an inner loop that is stabilized by negative-feedback outer loops, a common arrangement in biological control systems [see (*3*, p. 321) and (*4*)].
8. The overall gain of the optokinetic control system over the entire range of input velocities in Fig. 2 is 0.387. To obtain this value we assumed that the retraction velocity of a walking leg of a restrained lobster is equivalent to the locomotory (output) velocity. The average output velocity corresponding to each treadmill speed (input velocity) could then be computed directly from the data in Fig. 2. The mean gain (output velocity/input velocity) was then determined for each input velocity, and these were averaged to obtain the overall gain.
9. H. Bangert, *Z. Tierpsychol.* **17**, 143 (1960); K. G. Götz, *Kybernetik* **4**, 199 (1968).
10. K. G. Götz, *Kybernetik* **2**, 77 (1964); D. J. Rutherford and G. A. Horridge, *Quart. J. Microsc. Sci.* **106**, 119 (1965).
11. D. C. Sandeman, *Comp. Biochem. Physiol.* **24**, 635 (1968).
12. We thank Paul Ponganis, who performed the comparative studies on insects; Doreen Davis M. for the artwork; and Drs. L. Stark, R. Berger, B. Le Boeuf, and H. Wang for discussion and criticism of the manuscript. Supported by NIH grant NS-09050.

23 March 1972; revised 17 May 1972

Daily Variation in Concentration of Cortisol in Plasma in Intact and Hypophysectomized Gulf Killifish

Abstract. *A daily variation in concentration of cortisol in plasma is synchronized by a 12-hour daily photoperiod in intact as well as in hypophysectomized fish. The daily rhythm in concentration of the adrenal steroid does not depend on a daily rhythm in the concentration of pituitary adrenocorticotropic hormone.*

The adrenocortical (interrenal) tissue of teleost fish is stimulated by adrenocorticotropic hormone (ACTH) secreted by the anterior lobe of the pituitary gland. Hypophysectomy results in a decrease in the plasma concentration of cortisol, the major corticosteroid in poeciliid fishes (*1, 2*), and a regression of the adrenocortical tissue which can be alleviated or prevented by administration of mammalian ACTH (*3*). Daily rhythms in the concentration of plasma glucocorticoids have been reported for teleosts (*4*), as they have been for mammals (*5*) and birds (*6*). Although it was not investigated in teleosts, many of the daily rhythms of adrenal steroids in mammals are synchronized by the photoperiod (*5*). The rhythms in concentration of the glucocorticoids in plasma are thought to be direct consequences of daily rhythms of release of ACTH. If this assumption is correct for teleosts, hypophysectomy should not only cause a reduction in overall concentrations of cortisol in plasma, but it should also result in a loss of the daily rhythm. We demonstrate here that the presence of the pituitary is not required for the maintenance of a daily rhythm in the concentration of cortisol in plasma, nor for the photoperiodic synchronization of the rhythm in a teleost fish.

We studied intact and hypophysectomized male Gulf killifish *Fundulus grandis* (*7*). One group of intact fish and one group of hypophysectomized fish were maintained on a 12-hour daily photoperiod with light beginning at 0800 (series 1), whereas another group of intact fish and a group of hypophysectomized fish (series 2) were subjected to an inverted photoperiod of 12 hours, with light beginning at 2000 on 2 November 1971. After 15 days of acclimatization, the fish were killed at one of several times of day (4, 8, and 12 hours after the onset of light). The times were chosen on the basis of direct (*8*) and indirect evidence (*9*), which

Table 1. Concentration of cortisol in plasma of intact and hypophysectomized adult male *F. grandis*. Results are expressed as means ± standard error. Numbers in parentheses indicate number of fish studied. The quadratic curve of results for each of the four groups of fish of the two series follows a similar temporal pattern, whether a comparison is made between intact and hypophysectomized fish of series 1 versus intact and hypophysectomized fish of series 2, or between intact versus hypophysectomized fish in each series.

Group	Cortisol in plasma (micrograms per 100 ml) (hours after onset of light)		
	4	8	12
		Series 1; light from 0800 to 2000	
Intact	7.43 ± 2.23 (7)	19.50 ± 5.88 (6)	8.70 ± 3.32 (5)
Hypophysectomized	5.19 ± 1.21 (8)	14.29 ± 4.06 (7)	8.14 ± 3.32 (7)
		Series 2; light from 2000 to 0800	
Intact	19.42 ± 3.85 (6)	49.17 ± 8.04 (6)	21.08 ± 4.66 (6)
Hypophysectomized	5.0 ± 1.41 (6)	13.20 ± 2.42 (5)	7.75 ± 2.78 (4)