10. This survey included arboreal, prehensile-tailed species of 20 genera distributed among the orders Marsupialia, Rodentia, Edentata, Pholidota, Carnivora, and Primates.
11. R. E. Sloan [in *The Encyclopedia of Paleontology*, R. W. Fairbridge and D. Jablonski, Eds. (Dowden, Hutchinson & Ross, Stroudsburg, Pa., 1979), p. 492] reviews the diversity of Multituberculata.
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The Effects of Direct-Current Magnetic Fields on Turtle Retinas in vitro

Abstract. *Direct-current magnetic fields of 10 to 100 gauss cause a significant short-term reduction of the in vitro electroretinographic b-wave response in turtle retina. This response compression is not accompanied by the usual reduction in retinal sensitivity that occurs with background illumination. Furthermore, this effect is obtained only briefly after the offset of ambient lighting in the diurnal light-dark cycle of nonhibernating animals.*

The magnetic susceptibility of the human visual system is manifested by magnetophosphenes induced in the retina by alternating-current (a-c) magnetic fields (*1, 2*). Direct-current (d-c) magnetic fields do not elicit magnetophosphenes, but do influence certain spatially ordered biological systems, including the visual photoreceptors (*3–7*), through a physical realignment of diamagnetically anisotropic molecules, cellular elements, or both. For instance, in an aqueous suspension, isolated rod photoreceptor outer segments will realign themselves with their long axes parallel to an external d-c magnetic field of several thousand gauss (1 G = 10^{-4} tesla). This diamagnetic anisotropy (*8, 9*) arises in the photopigment rhodopsin (*10–12*). We have used an in vitro retinal eyecup preparation to study the electrophysiological effects of acute exposures to d-c magnetic field intensities known to influence rhodopsin in solution.

Our studies assessed the functional capacity of the vertebrate retina to undergo visual adaptation (operationally defined here as the change in retinal sensitivity resulting from an altered level of background illumination) and contrasted this capacity with the effects of applied d-c magnetic fields. We examined whether or not d-c magnetic fields can produce changes in retinal sensitivity and function similar to those that follow normal photoisomerization.

On-line electroretinogram (ERG) recordings from an in vitro turtle eyecup preparation (*13*) before, during, and after brief d-c magnetic field exposures were used; all of the results presented here refer only to b-wave ERG data. Both cone-dominant (*Pseudemys scripta elegans*) and mixed, rod-cone (*Chelydra serpentina*) turtle retinas were used to determine whether differences in photoreceptor cell type exist in magnetic field susceptibility. The animals were maintained under a rigorous diurnal light-dark (LD) cycle (lights turned on at 6 a.m. and off at 6 p.m.) for a minimum of 2 weeks before the study. Although experiments were conducted at different times throughout the diurnal cycle, all results are grouped into one of four different LD cycle phases: (i) light (L), 8 a.m. to 6 p.m.; (ii) light-to-dark transition (L-D), 6 p.m. to 8 p.m.; (iii) dark (D), 8 p.m. to 6 a.m.; and (iv) dark-to-light transition (D-L), 6 a.m. to 8 a.m.

Extracellular microelectrodes (25- to 50-μm tip diameters, 350- to 500-kilohm resistance) were placed in the thin layer of vitreous humor remaining in the eyecup after dissection and drainage. The eyecup was placed in a continuously gassed (95 percent O_2 and 5 percent CO_2) chamber within the 10-inch pole piece separation of an SCR-controlled d-c electromagnet. Magnetic field profiles mapped at this point showed a maximum d-c field inhomogeneity of 0.2 percent over the dimensions of the eyecup (7 to 10 mm) and a maximum a-c magnetic field ripple component of 10 mG in the

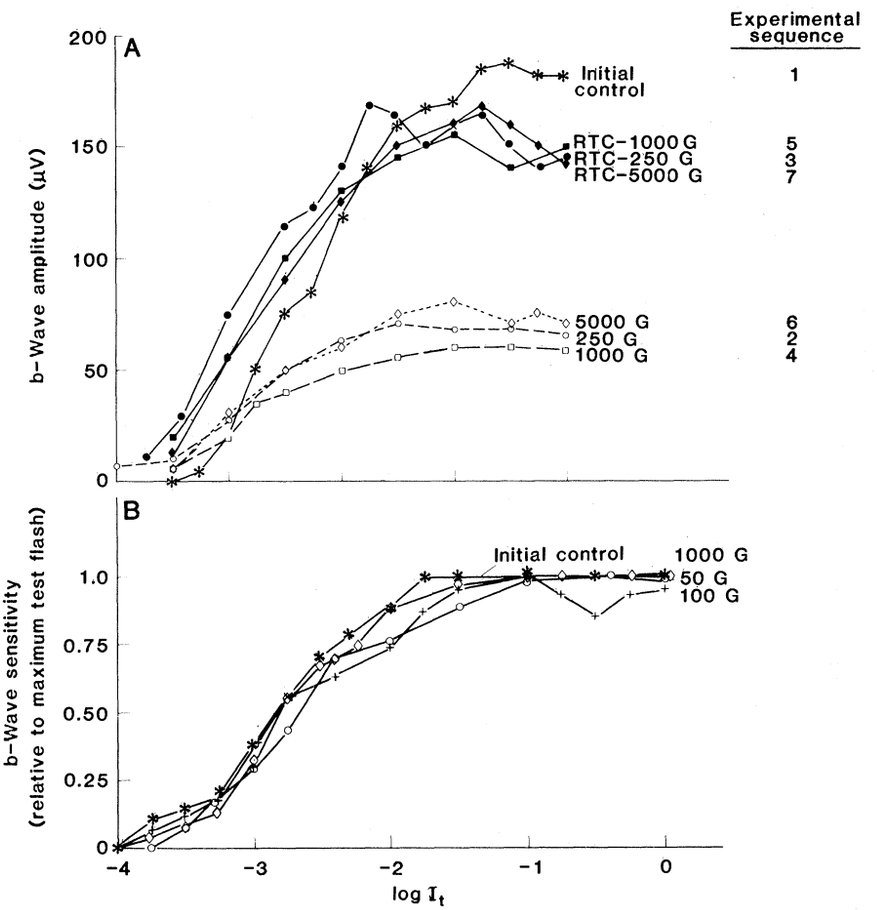

Fig. 1. (A) Retinal response of the turtle to photic stimuli during transition from light to dark. Intensity-response functions obtained with diffuse light flashes in an in vitro retinal eyecup (*Chelydra serpentina*) during d-c magnetic field exposures. Each curve is labeled with the d-c magnetic field intensity at which it was obtained. Note the extensive, but similar, amount of response amplitude decrement seen with each magnetic field intensity. (B) Normalized intensity-response functions that show no curve shifting as a result of retinal desensitization.

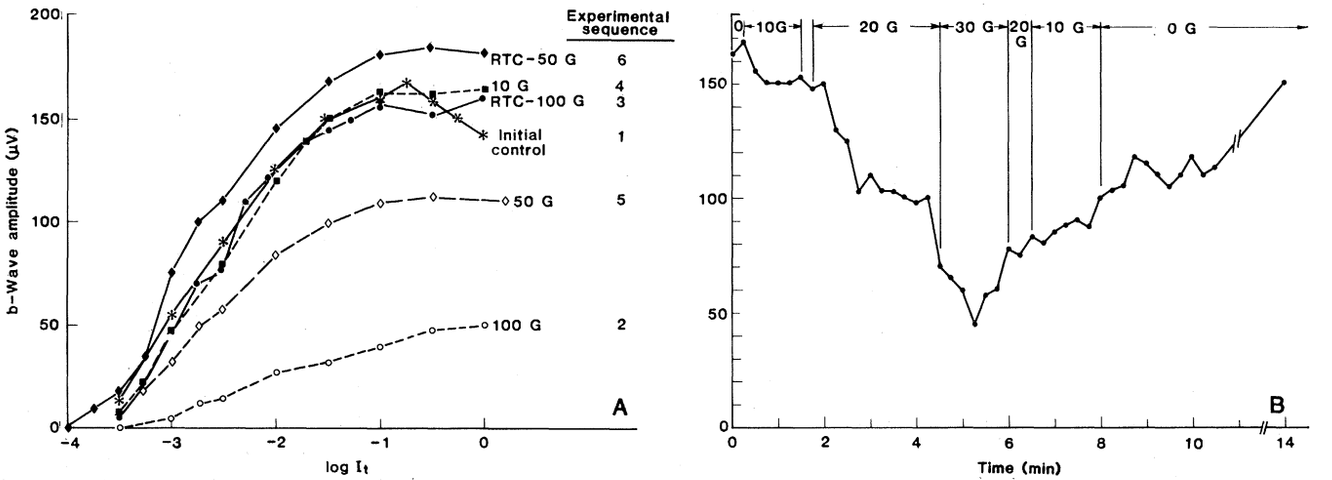

Fig. 2. (A) Intensity-response functions obtained with d-c magnetic fields near the threshold intensity. Response compression becomes evident (relative to control curves) by 50 G. The amount of response compression saturates at 100 G. (Compare with curves in Fig. 1A.) (B) ERG b-wave response amplitudes to a constant intensity probe light flash (that elicited half-maximum responses) during short-term exposures to d-c magnetic field intensities. The interflash interval was 15 seconds.

range of SCR driving currents used in these experiments. The upper cutoff frequency of the a-c–coupled recording system (time constant, 1.0 second) was 300 Hz (*14*) with additional notch filtering at 60 and 180 Hz. A standard calibration pulse was introduced through the recording electrode before each trial to monitor any electronics changes. Intensity response functions, or *V*–log *I* curves (*15*), were obtained for diffuse, 100-msec light flashes delivered through fiber-optic light guides to the retinal eyecup preparation. The amplitude of the ERG b-wave was measured from the a-c–coupled base line to the peak of the response.

As a control, light adaptation was first measured; this demonstrated that increasingly brighter backgrounds result in two major effects: (i) a reduction (or compression) of the b-wave response amplitudes to probe light flashes, which is seen as a downward shifting of the *V*–log *I* curves; and (ii) a rightward shifting of the normalized *V*–log *I* curves representing the retinal desensitization (*15*). The d-c magnetic field was then energized and adjusted to a selected magnetic field intensity at the recording chamber. A 5-minute waiting period ensured total stability of the applied field. A complete *V*–log *I* series was obtained at this stable d-c magnetic field level. The field was then reduced to the control level (0.5 to 1.0 G), another 5 minutes allowed to pass, and the return-to-control (RTC) *V*–log *I* series obtained. This sequence was repeated at every magnetic field intensity studied.

When these procedures were carried out with retinas just after the offset of ambient lighting, a large compression was seen in the ERG b-wave response amplitudes to probe light flashes presented during the short-term magnetic field exposures. Although they were not studied in detail, these same response compression effects were also seen in the ERG a wave. These effects were not observed at any other time of the diurnal cycle in these nonhibernating turtles, but were seen in both cone-dominant and rod-cone retinas.

This response compression was seen to some degree in every retina successfully tested ($N = 22$) during the L-D phase transition period of the diurnal cycle (Fig. 1A). The extent of response compression varied from retina to retina and ranged from 25 to 80 percent of the control responses. Compression can be seen by comparing the *V*–log *I* curves obtained during magnetic field exposures of 250, 1000, and 5000 G with their respective RTC curves (Fig. 1A). Because each retina was used as its own control, the statistical significance of the effects is represented by the differences in absolute response magnitudes between control and magnetic field curves. For example, the difference between the averaged means of the control and exposed conditions (Fig. 1A) was statistically significant [$t(2) = 9.925$, $P < .01$] for all stimulus intensities at, or brighter than, −2.0 log units. Another major characteristic of this effect was that its dose dependence had a narrow dynamic range in each retina and saturated at fairly low magnetic field strengths (100 to 250 G). This is also seen in Fig. 1A by the clustering of the different magnetic field curve maxima at amplitudes of 50 to 70 μV. This saturation effect resembles the dependence at low magnetic field strengths of the primary photochemical reaction centers of photosynthetic bacteria, in which the molecular mechanism is characterized by a perturbation of charge transfer processes (*7, 16*).

Since real light backgrounds also result in a comparable response compression (*17*), these data suggest that d-c magnetic fields mimic the effects of normal photoisomerization. However, the normalized data in Fig. 1B demonstrate that, unlike background lights, which cause a rightward shift (desensitization) of the normalized *V*–log *I* curves, d-c magnetic fields have no comparable effect on b-wave sensitivity. The normalized *V*–log *I* curves obtained during short-term d-c magnetic field exposures show no rightward shift and indicate that even though the absolute magnitude of the ERG photocurrent is reduced, there is no corresponding reduction in retinal sensitivity. A similar independence between response compression and retinal sensitivity has been reported after the removal of bicarbonate-CO_2 from the retinal superfusate (*18*). This deletion also lowered guanosine 3′,5′-monophosphate (cyclic GMP) concentrations in rod outer segments.

No d-c magnetic field effects were seen in any retina (either cone or rod-cone) tested during the L ($N = 17$), D ($N = 4$), or D-L ($N = 6$) periods. The diurnal asymmetry observed between the two phase transition periods (L-D, D-L) indicates that the d-c magnetic field–induced response compression depends on processes that occur after the offset of ambient diurnal illumination. Another process that is similarly activated by light offset (or darkness) is the synthesis of retinal melatonin (*19–21*). The kinetics of this circadian rhythm are rapid enough to play a role here, but any causal relationship remains obscure.

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The possible role of rhodopsin's diamagnetic anisotropy in this compression effect is addressed by the data in Fig. 2A. A 10-G field was subthreshold, a 50-G field produced a half-maximum compression, and a 100-G field is at the saturation level for this retina (compared with higher field strengths in Fig. 1A). The lowest threshold level seen was 10 to 20 G. These d-c magnetic field strengths are two orders of magnitude lower than those required for outer segment realignment (*3*) and at least one order of magnitude lower than those strengths that produce interaction energies exceeding the thermal energy level (*22*). These data argue that rhodopsin's anisotropy did not cause this response compression. Although experiments with altered magnetic field orientations would be helpful in addressing this issue, they were not technically feasible because of the restraints in positioning our in vitro eyecup preparation within the electromagnet gap. The ERG, however, is a voltage response summed across the entire curvilinear retinal surface. Thus, a considerable photoreceptor orientation already exists relative to the horizontal magnetic field used in these experiments.

Several additional experiments indicate that this effect can be demonstrated in nonhibernating animals only briefly after the offset of ambient lighting (L-D, 6 p.m.). Unlike the responses at low field strengths seen in retinas tested during the L-D phase of the diurnal cycle, strong d-c magnetic fields ($\geq$ 5000 G) were required to elicit any response compression in any retina prepared later than approximately 2 hours after the light offset. This effect is photodependent and not entirely caused by an internal circadian clock, because leaving the lights on at 6 p.m. and carrying out the same experiment did not result in any b-wave response compression. Dark adaptation per se was not involved, because protracted dark adaptation in vivo during any other period of the LD cycle did not bring about the effect. Concurrent light backgrounds did not prevent the magnetic field-induced response compression, but, instead, seemed to add their effects to that of the magnetic field. Our most recent experiments confirm the initial data obtained during the previous winter hibernation season, suggesting a significant seasonal variation in this diurnal effect. During the winter season (November through March), we did not always see a magnetically induced response compression during the L-D period; when this effect was present, the threshold magnetic field strengths required were usually high (> 1000 G). This observation is in line with the reported loss of diurnal retinal melatonin synthesis in turtles during winter (*19*).

These data indicate that d-c magnetic fields have a significant, but brief, suppressive effect on the extracellularly monitored, light-elicited ionic current fluxes in the in vitro turtle retina. These suppressive effects disappear after the field is removed. In vitro measurements of visual sensitivity are not likewise reduced; this suggests that the locus of this effect is not on any gain control mechanism in the retina. Our results do not resolve the question whether this response compression occurs in the photoreceptors themselves or in the synaptic processes involved in ERG b-wave generation. The importance of the diurnal LD cycle implies a hitherto unsuspected aspect of metabolic activity in the retinal outer plexiform layer. The effect on both rod and cone photoreceptor types and the very low threshold magnetic field intensities required suggest that the diamagnetic anisotropy of disk membrane rhodopsin is not crucial for these response compression effects.

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Single-Channel Fluctuations in Bimolecular Lipid Membranes Induced by Rat Olfactory Epithelial Homogenates

Abstract. *Chemosensitive single-channel fluctuations were observed to be induced in essentially solvent-free lipid bimolecular membranes by the addition of sonicated homogenates of rat olfactory epithelium. The chemosensitive channels were not observed when respiratory epithelium homogenates were used instead. Ionic selectivity is consistent with potassium ions as the charge carrier. These channels may be associated with the initial events of chemoreception in the rat olfactory epithelium.*

The initial events in olfactory chemoreception are poorly understood compared to analogous processes in bacterial chemotaxis (*1*). We report here a model system for the initial chemosensory events in the mammalian olfactory epithelium, the incorporation of rat olfactory epithelial homogenates into planar, essentially solvent-free bimolecular lipid membranes (BLM's).

Olfactory epithelia from three or four male Sprague-Dawley rats (200 to 225 g; Blue Spruce Farms) were pooled and minced in 10 m*M* 3-*N*-morpholino propanesulfonic acid (MOPS), *p*H 7.40, K^+ counterion, containing 15 μM adenosine triphosphate, 10 μM guanosine triphosphate, and 50 m*M* sucrose. The suspension was processed (4°C) in a Teflon-in-glass homogenizer (one or two up-down strokes, 200 rev/min) with a loose clearance, filtered through four layers of cotton gauze, and then was sedimented at 120,000*g* (30 minutes). The resulting pellet was twice resuspended and twice resedimented at 100,000*g* (20 minutes) in the same buffer. The final pellet was suspended in 250 μl of the buffer and was sonicated (30 to 40 seconds, 4°C). This suspension was sedimented at 12,500*g* (3 minutes), and the supernatant was retained as the homogenate for reconstitution studies. Light microscopic examination revealed vesicles (30 to 50 μm in diameter) which presumably arose from the resealing of ultrasonically disrupted