Combined use of the reference electrode and single interemitter capacitor enables stable, independent operation of the input transistors so that base-emitter voltages need not be matched. Transistors with matched gains or balance controls are not mandatory, as there is no first-stage discrimination against the interference signal. Circuit stability allows the signal inputs to be maintained close to a zero-potential so that very little current flows through the electrodes.

The tabulated performance features of Figs. 1 and 2 afford a meaningful comparison except in common mode capabilities which, due to the limitations of routine measurement techniques, are better compared under actual operating conditions. In examining a human scalp for brain potentials with a 1000-ohm impedance at all electrode junctions (save for one signal electrode of 5000 ohms) and with the subject placed in a uniform electrostatic field (50 mv of 60 hz interference impressed upon his body), the standard amplifier produced an interference output of 560 mv, while the modified amplifier produced 30 mv. In this practical test, an 18-fold reduction in susceptibility to interference and the effects of unequal electrode impedance is obtained by the modification.

With the modified amplifier circuit, low-noise electroencephalograms, electromyograms, electronystagmograms, and abdominal lead fetal electrocardiograms have been recorded. However, with signal impedances less than 10,000 ohms, any extracellular potential recording in the frequency range of 0.01 to 10,000 hz will be aided by the modified circuit.

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Noncollagenous Nature of the Proteins of Shark Enamel

Abstract. *The proteins, soluble and insoluble at neutral* p*H, of relatively mature enamel of two different species of sharks were distinctly different from both collagen and embryonic enamel proteins and similar to the proteins isolated from mature human and bovine enamel.*

The amino acid compositions of the proteins of the organic matrix of embryonic and developing enamel from a number of different species have been characterized by their relatively high contents of proline, leucine, histidine, and glutamic acid (*1–3*). During the maturation and development of enamel, more than 95 percent of the proteins, especially components rich in proline and histidine, are lost from the enamel (*4, 5*), so that the overall amino acid composition of mature enamel proteins is distinctly different from that of the embryonic proteins (*5, 6*). Although both the embryonic and mature enamel proteins contain small amounts of both hydroxyproline and hydroxylysine (*2–4, 7*), their amino acid composition, molecular structure, and physical chemical properties (*8, 9*) distinguish them from collagens. In contrast with these findings, however, it has been reported that the organic matrix of enamel from the mature shark consists principally of an ectodermal collagen (*10*). Because of the significance of this latter finding for the comparative biology of enamel (*10*), and because of the difficulties (*4, 5*) encountered in obtaining "pure," mature enamel free of dentinal or cemental collagen, we felt that these results should be independently confirmed, preferably on freshly obtained teeth, rather than on teeth obtained from commercial sources, which are usually treated with chlorine-containing bleaches and other strong reagents in order to improve their appearance. Consequently arrangements were made to obtain the teeth of several sharks immediately after their death.

The jaws of hammerhead sharks (2.4 to 3.0 m) were dissected free immediately after decapitation of live animals caught in the waters off the Florida Keys. The jaws were frozen with dry ice and kept frozen until used. After the jaws were thawed, the first three rows of unerupted teeth, which were adjacent to the already erupted teeth, were dissected from the mandible and maxilla. All soft-tissue attachments were carefully scraped and dissected free under the dissecting microscope, and the coronal surfaces were wiped clean with gauze. The teeth were placed in 0.3*M* EDTA (ethylenediaminetetraacetate), *p*H 7.7, at 2°C for 24 hours, and very small fragments of tissue were removed from the surfaces of the crowns. The surfaces of the crowns were brushed with gauze in EDTA and placed in fresh EDTA for an additional 12 hours. The surfaces were again cleaned, and the crowns were rinsed with water and immersed immediately in 0.5*M* EDTA, *p*H 8.3, at 25°C, with toluene or thymol crystals added for bacteriostasis. After 24 to 36 hours, small, partially decalcified, thin fragments of the enamel could be dissected free, with a small amount of enamel still present on the tooth. These small, partially demineralized fragments of enamel were placed in dialysis bags in 0.5*M* EDTA, *p*H 8.3 and dialyzed against EDTA at 2°C for 3 days, and then against 0.05*M* NH_4HCO_3, *p*H 7.7, at 2°C for 3 days. The clear supernatant representing the neutral-soluble, nondiffusible proteins was lyophilized. The fraction insoluble at neutral *p*H was washed with water and dried.

In addition to the hammerhead sharks, six near-term embryonic blacktip Bermuda sharks were obtained live, frozen immediately with dry ice, and kept frozen until used. The first and second rows of teeth, which were partially erupted, were dissected free and carefully cleaned of soft tissues. Because the teeth were so small, it was not possible to dissect the separate tissue layers. Therefore the teeth were placed in 0.3*M* EDTA, *p*H 7.7 at 25°C for 24 hours, the surfaces and pulp chambers were cleaned, and the teeth were washed with water. The teeth were then placed in fresh 0.3*M* EDTA, *p*H 7.7 at 2°C for 1 week and then fragmented in the EDTA solution with mortar and pestle. The entire contents including the EDTA solution were transferred to a dialysis bag and dialyzed for 3 days against 0.3*M* EDTA, *p*H 7.7 at 2°C. The nondiffusible proteins soluble at neutral *p*H were obtained as described. Portions were hydrolyzed in constant boiling 6*N* HCl, at 105°C for 24 hours and analyzed on an automatic amino acid analyzer.

Small fragments of the organic matrix, insoluble at neutral *p*H, of the enamel of the hammerhead shark teeth were washed with 0.15*M* NaCl, *p*H 7.4, rinsed quickly in water, and dried on siliconized glass slides. Thin sheets of the dried material were removed with a razor blade, and x-ray diffraction

Table 1. The amino acid composition of the proteins of shark enamel (*Elasmobranchii*). The results are expressed as the number of residues of amino acid per 1000 total amino acid residues.

Amino acid	Amino acid residues		
	Neutral-soluble proteins		EDTA insoluble from mature hammerhead
	Unerupted teeth, mature hammerhead	Partially erupted teeth, embryonic Bermuda black tip	
Cystine (one half)			2
4-Hydroxyproline			4
Aspartic acid	148	159	96
Threonine	44	35	47
Serine	197	104	84
Glutamic acid	161	87	119
Proline	56	59	59
Glycine	174	156	180
Alanine	54	58	72
Valine	23	30	45
Methionine	4	14	17
Isoleucine	19	26	40
Leucine	33	50	73
Tyrosine	11	29	25
Phenylalanine	9	24	26
Hydroxylysine			8
Lysine	21	94	48
Histidine	32	39	14
Arginine	19	19	43

analysis was carried out in a Phillips Microcamera fitted with a glass capillary (100-μ bore) collimator; CuK_α radiation (wavelength, 1.54 Å) was obtained from a copper target tube equipped with a nickel filter and operated at 35 kv.

The amino acid compositions of the neutral-soluble nondiffusible proteins and the proteins insoluble at neutral *p*H of shark enamel are given in Table 1. It is perfectly clear from these results that the nondiffusible, soluble enamel proteins of unerupted hammerhead shark teeth have a composition unlike that of either mesodermal or ectodermal collagens, or of embryonic enamel proteins. They are distinguished by their relatively high concentrations of aspartic and glutamic acids, serine, and glycine, similar to the nondiffusible proteins of mature human (*11*) and bovine (*4, 6*) enamel. This is also true of the insoluble residue obtained from the enamel of the unerupted hammerhead shark teeth, which, in addition to an amino acid composition distinctly different from collagen, showed only diffuse broad lines and no reflections of collagen by x-ray diffraction. The small amount of hydroxyproline in the insoluble residue probably represents, for the most part, contamination with dentinal collagen, although this amino acid appears to be present in trace amounts in bovine enamel proteins (*3, 4, 7*). Hydroxylysine, reported present in both porcine (*2*) and bovine (*3, 4, 7*) enamel proteins, occurs in much greater quantity than can be accounted for by the hydroxyproline present, and it is, therefore, in part at least, probably a true constituent of shark enamel proteins. The amino acid composition of the neutral-soluble embryonic shark enamel proteins, which may have included material from tissue other than the enamel because of the method of preparation, nevertheless were also distinguished by their high content of aspartic acid and glycine, with somewhat lower concentrations of serine and glutamic acid, and somewhat higher concentrations of proline and leucine than the enamel proteins of the unerupted teeth of adult hammerhead sharks. They are also characterized by their very high content of lysine and are distinctly different in amino acid composition from mesodermal or ectodermal collagens.

The finding that the shark enamel proteins were more similar in composition to the adult or mature bovine and human enamel proteins rather than to the embryonic or developing enamel proteins, is consistent with the findings that the enamels of both the unerupted teeth of mature animals and the partially unerupted teeth of embryonic animals were very well developed and highly mineralized, and, like mature bovine and human enamel proteins, were partially soluble in EDTA. These findings are consistent with the histological evidence that maturation and calcification of shark enamel occurs very early in development (*12*).

Since our findings, which are consistent with previous studies of the mature enamel of other species (*4, 6, 11*), are in distinct contrast with those reported by Moss, Jones, and Piez (*10*), it is necessary to try to explain the divergence. Examination of the methods and of the specimens used by Moss *et al.* offers a likely explanation for the discrepancy. First, the teeth used by Moss *et al.* were obtained commercially from a biological supply company which processes shark teeth by immersion in chlorine containing bleaches and other strong reagents that may remove considerable amounts of the organic matrix. Second, the mineralized shark enamel was isolated by Moss *et al.* from the underlying dentin by a flotation method (*13*) based on the difference in the densities of enamel and dentin. However, even when this method was repeated five or six times with mineralized bovine teeth, considerable quantities of dentinal collagen were always present in the enamel samples. Third, the mineralized enamel sample isolated by Moss *et al.* was decalcified in EDTA, and only the insoluble residue was used for analysis. Even with meticulous microdissection, small amounts of insoluble and soluble dentinal or cemental collagen are usually present in samples of mature enamel (*7*). Since most of the mature enamel proteins are soluble and diffusible (*4, 6*), the proportion of the dialyzed, insoluble dentinal collagen in the decalcified residue may be markedly increased, to the extent that a relatively "pure" sample of dentinal collagen is obtained. In our study, careful direct microdissection of shark enamel produced an EDTA-insoluble residue, which, in contrast to that obtained by Moss *et al.*, contained only a very small amount of collagen (about 5 percent), insufficient to generate an x-ray diffraction pattern in highly oriented samples. The major constituents of the insoluble residue were noncollagenous proteins. It seems likely, therefore, that the insoluble collagen obtained by Moss *et al.* from mature shark teeth originated primarily from tissue other than the enamel proper, probably the adjacent layers of dentin.

Further evidence that the matrix of shark enamel is similar to bovine and human enamel and is not collagenous comes from recent histochemical studies by Moss (*14*). Sections of decalcified embryonic shark teeth revealed that the enamel matrix, unlike the dentin, does not have the characteristic staining properties of collagen.

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Periodicity of Desert Rodent Activity

Abstract. *The radiation dose detected by microthermoluminescent dosimeters attached to pocket mice,* Perognathus formosus, *indicated the amount of time these animals were active on the surface of the ground. Radiation was from an elevated, partially shielded source in the center of the 8-hectare enclosure. The rodents are almost entirely inactive in midwinter but spend 30 to 40 percent of their time above ground in the summer months. Periods of activity increase gradually through the spring. These results support laboratory findings that members of this genus undergo periods of torpor in response to low ambient temperatures or food shortage. That this adaptation may enhance survival is indicated by the longevity of marked individuals of a related species.*

A microdosimeter of lithium fluoride has been developed to determine the amount of radiation to which small animals have been exposed in studies of radiation ecology (*1*). Thermoluminescent dosimeters made from this material are particularly valuable because they respond almost linearly to amounts of radiation ranging from a few milliroentgens to many kiloroentgens, and because their energy dependence is small (*2*). Microdosimeters are well suited to measuring radiation exposure and the dose absorbed by wild rodents living in irradiated areas (*3*). We have measured exposure of wild desert rodents (the pocket mouse, *Perognathus formosus*) to radiation from an artificial source, and with this information have evaluated seasonal changes in the daily activity cycle.

An 8-hectare circular area in the Mojave desert at the U.S. Atomic Energy Commission's Nevada test site is irradiated by a ^{137}Cs source supported 15 m above ground at the center of the area. The source is differentially shielded in order to reduce radiation intensity at ground level near the tower supporting the source. With increasing distance from the tower there is less shielding between the source and the ground. In this way the variation in dose rate with distance is reduced to approximately sixfold along the radius of the study area from 50 m to over 160 m (Fig. 1). Below the surface of the ground radiation decreases rapidly. At a depth of 15 cm the dose rate is less than 10 percent of the surface dose rate. At 30 cm it is less than 1 percent. Farther from the center of the area, the dose rate decreases more rapidly with depth because of the greater angle of incidence between the source and the ground. Rodents in their burrows are therefore shielded from the radiation.

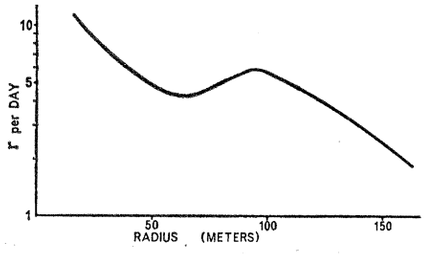

Fig. 1. Radiation dose rate at ground level along the radius of circular 8-hectare plot.

The microdosimeter, a sealed glass capillary tube 0.8 by 6.0 mm containing 0.6 mg of LiF crystals, is placed in a small piece of polyethylene tubing, and that in turn is placed into a piece of black electrical spaghetti. The assembly is fastened to the neck of the rodent with a single suture. Rodents are trapped at monthly intervals, and dosimeters are removed from the containers and replaced with unexposed units. Our results are based on the recovery of 136 dosimeters.

Radiation exposure is determined by the total time an animal spends on the surface of the ground, as well as by the location of the plot where it lives. These rodents normally move about an area approximately 20 m in diameter. The trapping records for an individual rodent indicate where it lives, and the plot of radiation dose rate tells the degree of exposure in that location. The fraction of the exposure period during which the animal was on the surface is obtained by comparing the total ground exposure with the exposure of the dosimeter (Fig. 2).

The average dose rate in roentgen per day received by rodents in the study area between 1964 and the present shows strong seasonal fluctuations (Fig. 2). Exposure varies from nearly zero in midwinter to over 1.25 r/day during the summer months. Integrated exposure in the first half (January through June) of 1966 was 50 percent higher (148 r) than for the same period in 1965 (95 r). The average annual radiation exposure to the rodents is approximately 350 r. These dose rates are not known to affect the natural behavior of animals in any measurable way. Comparison of each exposure with the surface dose rate where the animal was trapped shows the activity time of the animal. The variation in exposure between years, as well as the seasonal variation, resulted from changing activity of the animals, that is, the amount of time they spent on the surface. The average fraction of time on the surface (Fig. 2) indicates almost complete inactivity during 1 month in winter, whereas they spent 30 to 40 percent of the time on the surface in summer.

The rodents were 50 percent more active in the spring of 1966 than during the corresponding time in 1965. The cooler, wetter spring of 1965 probably suppressed surface activity of the animals. These conditions resulted in greater production of leaves, flowers, and fruits by plants. Reduced animal activity may have been a consequence of the abundant food supply.

Bartholomew and Cade (*4*) reported

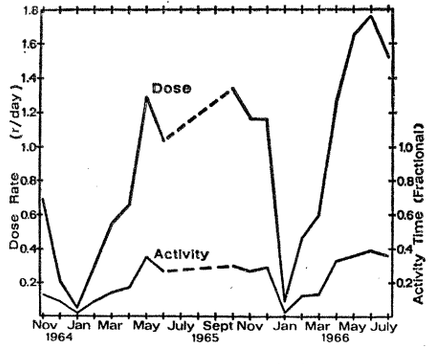

Fig. 2. Monthly mean radiation exposure to pocket mice, and fractional time active on the ground surface. The fraction 1.0 equals 100 percent of the time. The dotted lines cover a 4-month period during which no dosimeters were retrieved.