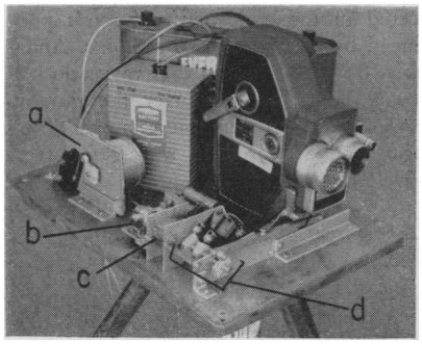

Fig. 1. Camera and timing mechanism showing (*a*) clock and timing cam, (*b*) electric motor, (*c*) gear reduction case, and (*d*) shutter tripping and reset cams.

a study, the U.S. Geological Survey obtained two such cameras and coupled them to battery-operated timing devices designed and constructed by Ernest Parshall of the Geological Survey. Each of these devices is operated by a small electric motor that rotates a cam, which in turn trips the shutter mechanism of the camera (Fig. 1) once every 15 minutes. The timing cam on the clock can be changed easily, and the present range of the interval cam is from 2 minutes to 1 hour. Slight modification of the reset cams could increase the interval to one frame every 24 hours.

We decided to apply the technique to a study of glacier motion. Emmons Glacier in Mount Rainier National Park, Washington, was chosen because of its accessibility and because it is known that some glaciers on Mount Rainier have been advancing since 1946.

The objectives of the initial study were to photograph the forward movement of a glacier terminus and the ablation of the glacier, together with the resulting movement and accumulation of debris on and in the ice.

On 21 June 1958, members of the Geological Survey and personnel of the National Park Service under the supervision of Vernon R. Bender, park naturalist, back-packed the disassembled shelters and cameras to sites adjacent to Emmons Glacier. Mark M. Meier, of the Geological Survey, surveyed the front of the glacier at the time the cameras were installed and erected control stakes for precise measurements of glacier movement. One camera was placed on debris-covered stagnant ice, to one side and about ¼ mile away from the terminus of the glacier. The site was abandoned for reasons of safety on 16 August, because of excessive melting of the stagnant ice during the summer. The other camera was installed on debris-covered stagnant ice about 300 feet in front of the glacier (Fig. 2) and was not removed until 9 October. During the summer the cameras and timers were serviced once every 10 days by Park Service personnel.

Fig. 2. Camera shelters and front of Emmons Glacier.

The results of the first season's operation were disappointing, owing to mechanical breakdowns of the shutter mechanisms of both cameras and to failure of one electric motor. Unseasonably warm weather, moreover, resulted in less forward movement of the glacier than had been anticipated. The film obtained does confirm the view that exterior time-lapse photography can be applied to the study of glacier motion, but the results of the summer's study are incomplete. After modifications had been made in the timing mechanisms and in the construction of light, easily portable shelters, the cameras were reinstalled in new locations near Nisqually Glacier on Mount Rainier in June 1959 (*1*).

ROBERT D. MILLER
DWIGHT R. CRANDELL
U.S. Geological Survey,
Denver, Colorado

Notes

1. Publication of this report was authorized by the director of the U.S. Geological Survey.

30 April 1959

Mammalian Cytochrome b

Abstract. Digestion of a preparation of cytochromes *b* and c_1 with pancreatic protease followed by ammonium sulfate precipitation resulted in a soluble cytochrome *b* uncontaminated by cytochrome c_1. This preparation, which was free of succinic dehydrogenase and cytochrome oxidase activity, had an estimated $\Delta E_{1\,cm}^{1\,g/ml}$ of 102 for its alpha-peak. In the reduced form absorption maxima were found at 560 to 562, 530 to 532, and 427 to 428 mμ, and in the oxidized form, at 413 mμ.

Since 1948, when Wainio and his coworkers (*1*) first reported on the use of 4-percent sodium deoxycholate to solubilize cytochrome *b* from mammalian heart muscle, there have been several attempts to purify this material. By lowering the deoxycholate concentration, Eichel *et al.* (*2*) were successful in removing most of the cytochrome oxidase contaminant and characterizing the cytochrome *b* spectrum. The major absorption maxima in the reduced form were demonstrated at 560, 530, and 428 mμ, and in the oxidized form, at 414 mμ. In 1954 Hubscher, Kiese, and Nicolas (*3*) reported on a soluble preparation of cytochrome *b*, showing reduced absorption maxima at 564, 530, and 431 mμ. However, this material contained a contaminating component absorbing at 554, 525, and 418 mμ which these investigators considered a denaturation product of cytochrome *b* but which is now recognized as cytochrome c_1. In addition, considerable succinic dehydrogenase activity was present. Widmer, Clark, Neufeld, and Stotz (*4*) purified cytochrome

b by fractionation with sodium cholate and subsequent ammonium sulfate precipitation. This preparation also contained succinic dehydrogenase activity and a 554 contaminant. More recently, in 1956, Sekuzu and Okunuki (*5*) reported on the preparation of a "crystalline" cytochrome *b* from ox heart muscle. However, little information was presented on the preparation and properties of this material, and inspection of the gamma band of the dithionite-reduced spectrum indicates some absorption in the 418-to-420 mμ region.

In our attempts to isolate and purify cytochrome *b* free of any spectral contaminant and extraneous catalytic activity, use was made of the finding of Wainio, Eichel, and Cooperstein (*6*) that the component which most rapidly sedimented in the ultracentrifuge was cytochrome *b*. It was felt that this might be a useful technique for removing contaminating components.

Preparations of cytochrome *b*, made by the method of Bernstein and Wainio (*7*) from successive extractions of an insoluble preparation of beef heart muscle with sodium deoxycholate, were centrifuged at 168,380*g* in the "C" rotor of the Spinco analytical ultracentrifuge for 4 hours. A red gelatinous pellet was obtained which contained most of the cytochrome *b* component of the original fraction as assayed spectrally before and after centrifugation. This red pellet was solubilized by digestion with phospholipase A (*8*), and a red precipitate was obtained after ammonium sulfate precipitation at approximately 30-percent saturation. The absorption spectrum of this precipitate dissolved in 0.1*M* phosphate buffer at *p*H 7.4 showed maxima in the dithionite-reduced form at 560 to 562, 530 to 532, and 428 to 430 mμ. However, there was obviously a contaminant absorbing at 553, 525, and 418 mμ. Ultracentrifugal studies of this preparation of cytochromes *b* and c_1 indicated a single component with an uncorrected sedimentation constant of 13.9×10^{-13} sec. This compares with the previously reported value of 6.5×10^{-13} sec for cytochrome *b* (*6*). It would appear that at this stage we probably were dealing with a cytochrome b–c_1 "particle."

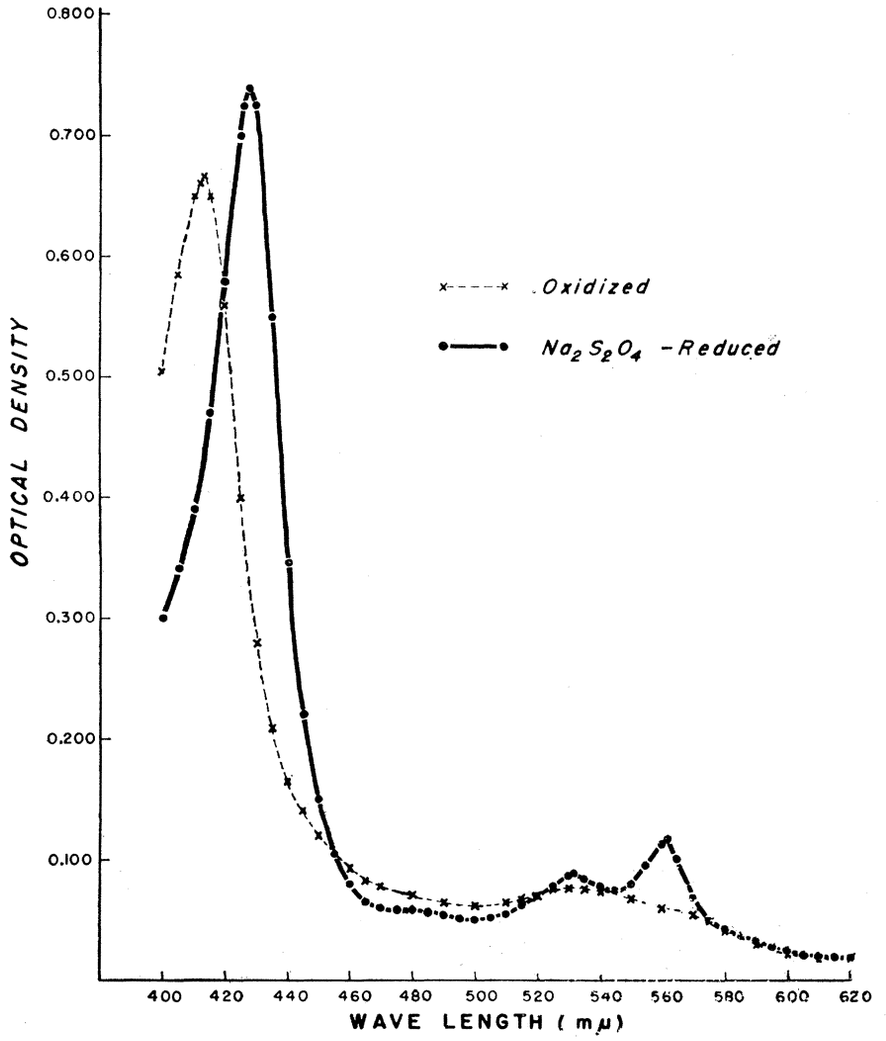

Fig. 1. Oxidized and dithionite-reduced absorption spectra of cytochrome *b* after protease treatment.

Successful removal of the 553 mμ component was accomplished by digestion of the soluble cytochrome b–c_1 "particle" with protease (*9*) at room temperature for 2 hours and subsequent precipitation at *p*H 7.4 with ammonium sulfate at approximately 20-percent saturation. A bright red precipitate was obtained which could be dissolved in 0.1*M* phosphate buffer at *p*H 7.4. The oxidized and dithionite-reduced spectra are shown in Fig. 1. Maxima in the reduced state are at 561, 531, and 428 mμ. The range of values for several preparations was 560 to 562, 530 to 532, and 427 to 428 mμ. In the oxidized state, a prominent peak is seen at 413 mμ.

This preparation could not be reduced with sodium succinate and showed no absorption at 553, 525, or 418 mμ. In addition, no succinic dehydrogenase or cytochrome oxidase activity could be detected (*11*). The 553-mμ component (cytochrome c_1) was found to be present in the supernatant after ammonium sulfate precipitation at approximately 20-percent saturation. The cytochrome *b* preparation at this stage still contained trace amounts of sodium deoxycholate, and attempts to remove this component by dialysis have led to denaturation.

The extinction coefficient ($\Delta E_{1\,cm}^{1\,g/ml}$) for this cytochrome *b* preparation was calculated for the maximum at 561 mμ relative to the isosbestic point at 575 mμ and was found to be 102. This should be compared with the value of 63 calculated by Slater (*10*) for the preparation of Sekuzu and Okunuki (*5*).

As the protease preparation was grossly impure, it is not yet possible to state with any certainty what fraction(s) of this crude enzyme were responsible for hydrolyzing the cytochrome *b* from the cytochrome b–c_1 "particle." It is of interest, however, that a series of other proteolytic enzymes from animal, bacterial, and plant sources, as well as a variety of phospholipases and esterases, were not effective. No definite conclusions can be drawn as to the nature of the cytochrome b–c_1 linkage.

DONALD FELDMAN
Medical Research Laboratory, Veterans Administration Hospital, East Orange, New Jersey

W. W. WAINIO
Bureau of Biological Research, Rutgers University, New Brunswick, New Jersey

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8. Rattlesnake venom (from *Crotalus adamanteus*) was obtained from Ross Allen's Reptile Institute, Silver Springs, Fla. Bee venom was even more effective at this stage.
9. A crude protease from bovine pancreas obtained from Nutritional Biochemicals Corp., Cleveland, Ohio, was used.
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14 May 1959

Albinism in the California Hagfish Eptatretus stoutii

Abstract. The discovery of an albino specimen of the hagfish, *Eptatretus stoutii* (Lockington), is reported, together with general notes regarding collection of this species for research purposes. The extreme abundance of hagfish in the area described refutes the generally accepted notion that they are of infrequent occurrence in southern California waters.

True albinism occurs but rarely among fishes, and it is believed that this is the first record of such a natural freak to be reported among the most primitive of craniate chordates, the hagfish (Fig. 1). There is a report of an albino lamprey (*Petromyzon marinus*), the other division of the class Cyclostomata, which was collected some years ago at the mouth of the Salmon River on Lake Champlain in New York (*1*).

During a routine collection for specimens of *Eptatretus stoutii* (=*Polistotrema stoutii*) on 5 May 1959, using 5-gallon-can traps, I obtained an albino individual in a total collection of some 500 fish. Over the past 2 years I have taken about 5900 hagfish of this species from the same general vicinity (latitude, 32° 31'N; longitude, 117° 18'W). This area is the floor of a submarine canyon, a tributary of the San Diego Trough, where the bottom is green mud and the depth is 210 fathoms. The traps remained on the bottom for 7 hours and they were baited with fish oil and mackerel.

Normally, the body color of this species is pinkish to purple grey, lighter ventrally than dorsally. Of the several thousand hagfish studied, about 20 were piebald (Fig. 2), with certain areas pale pink because of reduced pigmentation.

The skin of the albino is white save for a pinkish cast imparted by the blood. This coloration is most pronounced in the eye regions, the barbels, and the caudal area. The pulsations of the caudal heart are plainly visible beneath the translucent skin.

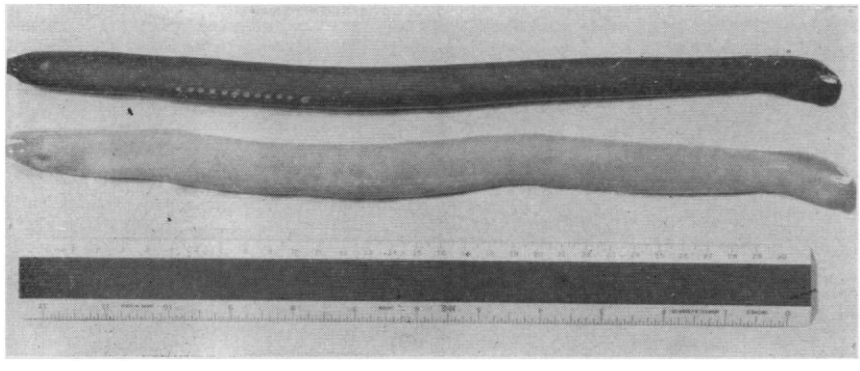

Fig. 1. Living albino *Eptatretus stoutii* with normally colored individual for comparison (anesthetized). Note the slime glands beneath the skin in the albino (especially prominent posterior to the gill openings), not readily evident in the pigmented specimen.

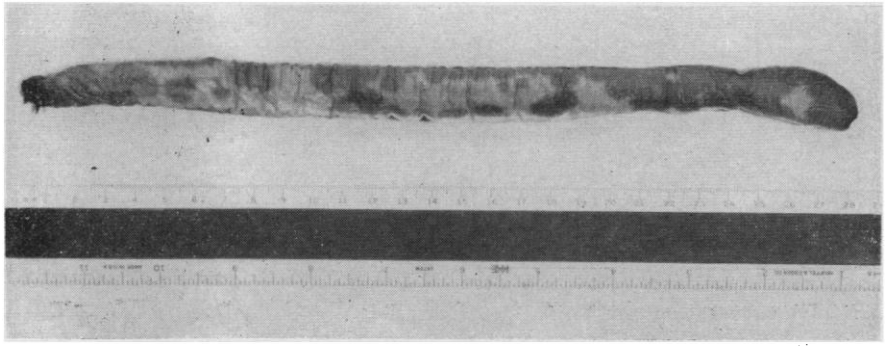

Fig. 2. Piebald specimen of *Eptatretus stoutii* (preserved), collected 7 Nov. 1958.

Measurements of the albino specimen (*2*) were as follows: body length, snout to tip of tail, 38.1 cm; weight, 55 g; external gill openings, 12 left, 12 right. Measurements of the piebald specimen were as follows: body length, 28.3 cm; weight, 36 g; gill openings, 11 left, 11 right.

DAVID JENSEN
Scripps Institution of Oceanography, University of California, La Jolla

References and Notes

1. F. H. Wilson, *Lebanon Valley College Alumni News* (January 1954).
2. This specimen was obtained during the course of research supported by a grant from the San Diego County Heart Association, to which grateful acknowledgment is made.

10 June 1959

Identification of a Growth Inhibitor from Extracts of Dormant Peach Flower Buds

Abstract. A growth inhibitor in dormant peach flower buds was identified as naringenin (5,7,4'trihydroxyflavanone) from infrared and ultraviolet spectra, through determination of melting point, through paper chromatography, and from activity in the bioassay test.

A growth-inhibiting substance has been reported to occur in resting peach flower buds by a number of investigators (*1–3*). This substance has been closely correlated with the emergence of peach buds from rest (*1*); however, its identity is unknown.

In an investigation of the seasonal fluctuation in quantity of growth substances in resting peach flower buds, several attempts were made to obtain a purification step which would separate the inhibiting substance from oils, fats, and waxes present in the plant extract. The inhibitor could not be separated by means of a sodium bicarbonate solution; therefore, the ether or methanol extract was dissolved in 0.1*N* NaOH. The *p*H was adjusted downward, one unit at a time, with 0.1*N* HCl, and after each adjustment of *p*H the aqueous solution was extracted with peroxide-free diethyl ether and the ether extract was assayed for activity. It was noted that the intense straw color at *p*H 9.0 changed to a less intense straw color at *p*H 8.0. It was found that the inhibitor was very soluble in the ether phase at *p*H 8.0. There was a small amount of the inhibitor in the ether phase at *p*H 9.0 and 7.0. This provided a means whereby the inhibitor could be recovered free from fats, oils, and waxes and from many of the acidic substances which still remained in the salt form at this *p*H.

A number of attempts were made to crystallize the inhibitor from resting