

BEHAVIORAL AND ELECTRICAL RESPONSES ASSOCIATED WITH EXPOSURE TO EXTREME COLD OR HEAT IN THE SALAMANDER¹

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In order to obtain a clearer understanding of the influence of extremes of temperature on the responsiveness of organisms, the present study applied a series of neurological tests to salamanders exposed to extremes of cold or heat. These tests have been employed successfully to investigate the action of drugs on the nervous and muscular systems of salamanders (Peters, Vonderahe, and Palmisano, 1956; Peters, Vonderahe, and Powers, 1958). They yield information about patterns of behavior, reactions to stimuli, the function of definite tracts and reflex arcs, and electrical activity. Even in a motionless animal, recordings of electrical activity provide clues to the alertness of the animal, condition of nervous and muscular systems, and the frequency and regularity of the heart beat. The salamander, *Triturus viridescens*, was selected for this experiment because it is poikilothermous and does not undergo hibernation; at moderate temperatures it is active and responds readily to a variety of stimuli; and finally the careful studies of Coghill (1930) and Herrick (1948) are available for correlation of its anatomy and behavior.

MATERIALS AND METHODS

About 100 salamanders, the aquatic phase of *Triturus viridescens*, were used for the present study. The animals were maintained for one or more days at a temperature of $24 \pm 1^\circ \text{C}$, then subjected to high or low temperatures of 44° or 0°C . Tele-Thermometers (Yellow Spring Instrument Co.) equipped with hypodermic and other types of small probes were used for measuring temperatures, both internal and external, with an accuracy of plus or minus one degree C. A moist chamber containing the salamander was suspended in an incubator wherein the temperature was changed from 24 to 44°C by a surrounding heating coil and a variable Powerstat transformer. Lower temperatures were achieved by lowering or raising the moist chamber in a larger container immersed in a frozen 10 percent NaCl solution. The salamanders were kept at the extreme temperatures for 30 minutes. However, to avoid excessively sudden changes, 30 minutes were consumed in making the gradual transition from 24°C to either extreme of temperature, and another 30 minutes allowed for the return to 24°C . At approximately 10 minute intervals during the transitional periods, and at the end of 30 minutes of exposure to 44 or 0°C , the salamanders were given a series of neurological tests. These neurological tests involved walking, righting, corneal and abdominal reflexes, optokinetic nystagmus, response to touch, recording of electrical activity of the neuromuscular systems and heart beat, and reaction to the stimulant, Metrazol (Peters, Vonderahe, and Palmisano, 1956). For electrical studies, four pairs of stainless steel needles were inserted under the skin so as to span four regions of the body (fig. 2), namely, the head, chest and heart, the lumbar, and the pelvic regions of the trunk. A Grass Model III-D 4-channel electroencephalograph was used for recordings. The electrical activity is interpreted as being of muscular origin with modifications by the nervous system

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and superimposition of an electrocardiogram, especially in recordings from the chest. Metrazol (pentylenetetrazol) at a dosage of 1 mg per gm of body weight was injected into the abdominal cavity during the 30 minutes of exposure to extreme temperatures. In the salamander at moderate temperature this dosage of Metrazol induces a violent seizure, as manifested by external behavior and electrical activity (Peters and Vonderahe, 1954). During the transitional periods the temperature within the abdominal cavity typically lags behind that of the external environment by about 2° C. At the end of 30 minutes exposure to extreme cold the internal temperature reaches 0° C. However, at the end of 30 minutes exposure to 44° C the temperature within the abdominal cavity is at 40° C. Unless stated otherwise, the observations and illustrations (figs. 1 and 2) refer to temperatures of the external environment. Separate animals were used for behavioral tests, electrical recordings, reaction to Metrazol, measurement of

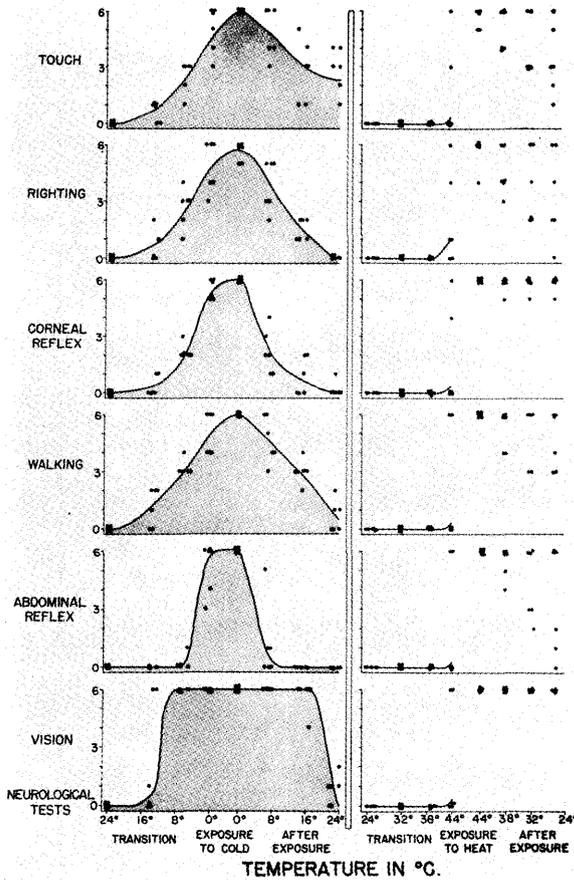


FIGURE 1. Graph illustrates the modification of neurological responses of salamanders subjected to extremes of temperature indicated on the lowest line of the horizontal axis. On the vertical axis, 0 signifies a normal response to various neurological tests, 1 to 6 degrees of depression, with 6 indicating complete extinction of response. Each dot represents the result of a test on a single salamander. Under the influence of cold, the responses gradually disappear, but return to normality with moderate temperature. Under the influence of heat, all responses are lost suddenly and simultaneously; some are regained temporarily before the death of the organism.

internal temperature with a hypodermic probe, and exposure to extremes of heat or of cold.

RESULTS

Although a more detailed verbal report of observations is given below, the reader may find it helpful to refer immediately to figures 1 and 2 in which the neurological changes are graphically summarized. In figure 1 temperature changes are indicated on the horizontal axis; different behavioral tests and various degrees of depression accompanying the changes in temperature are indicated along the vertical axis. Zero indicates a normal response, and the numbers 1 to 6 refer to degrees of depression, in which 6 refers to complete extinction of response. Twelve animals were used to compile figure 1, and each dot represents the result of a single test on a single animal. Modifications of electrical activity of neuromuscular system and heart which accompany changes in temperature are illustrated in figure 2.

Neurological Changes during a Lowering of Temperature

As the temperature drops from 24 to 0° C, the salamander exhibits a sequential loss of neurological responses. The first influence of cold is evident at 14° C when walking, righting, and the responses to touch are performed more slowly. As the temperature drops to 7° C, walking and righting become more labored; the corneal reflex is less prompt; the response to touch is negative unless a strong stimulus is applied; and attempts to evoke optokinetic nystagmus are unsuccessful. When 0° C is reached, the abdominal and corneal reflexes disappear, and touching is followed by an occasional weak response. When placed on its back, the salamander makes a few feeble attempts to right itself by movements of trunk and limbs. Eventually the forelimbs lose the ability to support the body, and finally the effort to walk degenerates into an erratic placement of limbs without any forward movement of the animal. If the salamander is transferred to water of the same temperature (0° C), it neither swims nor rights itself. At the end of 30 min at 0° C the salamander is limp and shows no response to the various stimuli employed in this study. In some superficial blood vessels, with the aid of a microscope, blood cells can be seen to be moving rapidly while in other vessels they appear motionless. Recordings of electrical activity at 0° C indicate that a lowering of temperature is accompanied by a decrease in the amplitude and frequency of waves from the neuromuscular system. The frequency of the heart beat has diminished from about 45 beats per minute at 24° C to 5 beats per minute, and at lowered temperature each heart cycle consumes a longer time. At 0° C neither behavior nor the electrical record showed changes following an injection of Metrazol.

As the temperature is raised gradually from 0 towards 24° C, the salamander regains responsiveness to various stimuli in the following sequence (fig. 1). At 7° C the abdominal and corneal reflexes are sluggish but definitely present. The animal recovers the ability to right itself and to walk slowly. The response to touch is small and inconsistent. The test for optokinetic nystagmus remains entirely negative. The electrical record shows a slight increase in amplitude and some acceleration of the heart rate. As the temperature approaches 12° C salamanders previously receiving an injection of Metrazol of 0° C begin to show gasping, some spastic movements and opisthotonus. With a further rise in temperature clonic behavior becomes more conspicuous.

At 14° C the corneal and abdominal reflexes are prompt; spontaneous movements of trunk and limbs occur more frequently; and walking and righting are less labored. However, the response to touch remains only slight and the visual test is negative. The electrical record shows a further increase in amplitude.

When a temperature of 24° C is reached, the salamander shows positive re-

sponses to all the neurological tests, but there is still some lack of promptness especially in the response to touch. The electrical activity of neuromuscular system and the heart rate return to the amplitude and frequencies recorded at the outset of the experiment dealing with cold. Within 24 hours at a temperature of 24° C the salamander appears normal in all respects, and observations over the next few days reveal no abnormalities due to the exposure to 0° C.

Neurological Changes with a Raising of Temperature

The neurological changes which accompany a rise in temperature are graphically summarized in figures 1 and 2. The sudden depression of all responses at 44° C, and the individual differences between salamanders during the transition from 44° to 24° C did not permit the plotting of a curve for these observations. This fact is a noteworthy difference between the effects of extreme heat and cold on the behavior of the salamander. Modifications of electrical activity of the neuromuscular system and the heart during extreme heat are illustrated in figure 2.

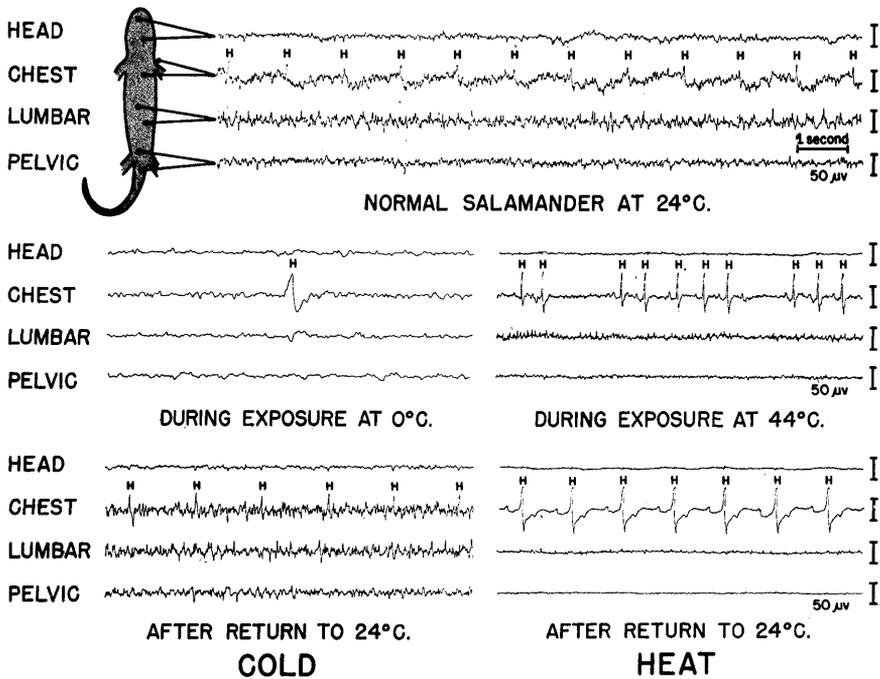


FIGURE 2. Electrical activity recorded from specified regions of salamanders at the temperatures indicated beneath each record. During exposure to either extreme of temperature the electrical activity of the neuromuscular system diminishes in amplitude. Cold slows the heart rate; heat induces acceleration and irregularity. After a return of 24° C., salamanders exposed to cold regain a normal electrical record while those exposed to heat show continued low amplitude and a modified electrocardiogram (H).

As the temperature rises from 24 to 34° C the salamanders manifest no conspicuous abnormalities, except increased rapidity in attempts to escape. Above 34° C these escaping reactions appear somewhat spasmodic. At 41° C activity diminishes, but the salamander gives prompt responses to the various stimuli employed in the neurological tests (fig. 1). When the animal is motionless, the electrical records from the head and trunk show waves of moderate amplitude with

a frequency of about 30 to 40 per second; the amplitude can be temporarily increased by touching the animal. At 44° C the heart rate may reach a frequency of 120 beats per minute. At some time during the exposure to 44° C, the exact moment is not consistent, the salamanders typically exhibit a cloniclike lashing of the vertebral column, lasting about 30 seconds, followed by slow serpentine movements of the trunk, lasting several minutes. When this episode of violent activity ceases, the salamander enters a stage of inactivity and offers no response to any of the stimuli employed in this study. At this time the electrical record shows a decrease in the amplitude of the electrical activity of the neuromuscular system (fig. 2); the heart rate is accelerated but irregular. Blood cells can be seen circulating in peripheral blood vessels. If Metrazol is injected into the abdominal cavity after the clonic-writhing episode described above, this stimulant does not evoke a change, either in the electrical record or in the behavior of the salamander. For about 50 percent of the salamanders exposed to high temperatures the events just described above culminate in a permanent disappearance of all vital signs; for them the electrical record persists at the noise level of the apparatus. For the remaining 50 percent the gradual lowering of temperature is associated with a temporary and depressed return of some of the responses to the stimuli used in the neurological tests. When a salamander regains these responses, they typically return in the following order: the response to tactile stimulation returns first, then righting, followed by walking. The abdominal reflex appears next and finally the corneal reflex. None of the salamanders showed a return of optokinetic nystagmus. The return of responsiveness is accompanied by some increase in the amplitude of the electrical record and an occasional resumption of a regular heart beat (fig. 2). Whatever responses are present at 24° C lack the promptness found in normal salamanders. The regaining of activity described above is of brief duration, for the animals become sluggish and die within 24 hours after the exposure to 44° C. Death is associated with a swelling of the body, vascular dilation, and some hemorrhage in the oral and nasal cavities.

DISCUSSION

During the transition to and from cold temperatures, the successive loss and return of various responses may find an explanation in different resistances of various nerve fibers to certain degrees of cold, as suggested by Brooks (1956). The exposure for 30 minutes to either 44 or 0° C leaves the salamander immobile and irresponsive to stimulation. However, the results of the neurological tests immediately before and after exposure to these extreme temperatures indicate that the reasons for this condition are quite different in the two experiments. The lower range of temperature produces a motionless animal presumably because it raises the threshold of receptors and effectors, delays conduction of nerves (Tasaki and Fujita, 1948) and contraction of muscles, and diminishes the ionic activity (Crescittelli, 1957) associated with the electrical discharges by the neuromuscular system. The lowered frequency of heart rate and the protracted time for individual beats suggest that lowered temperatures decrease the irritability of the pacemaker and slow conduction in the auriculoventricular bundle. The deep depression associated with cold is temporary as proved by the return to normal behavior and normal electrical activity when the organism is returned to an environment of 24° C. Apparently the salamanders suffered no permanent injury when exposed to 0° C for 30 minutes.

The simultaneous disappearance of all responsiveness to tests (fig. 1) and the drop in amplitude of the electrical record of the neuromuscular systems in some salamanders after an exposure to 44° C suggest that heat has a tendency to cause sudden and widespread changes in the organism. In all the salamanders the injury caused by heat was permanent and lethal, even though individual differences were manifested in the resistance to heat and in the temporary return of some of the responses.

The cloniclike lashing and subsequent writhing induced in the salamander by exposure to extreme heat permits several conclusions. This behavior may be regarded as a convulsant type of activity induced by heat similar to the convulsions induced by heat in mice (Swinyard and Toman, 1948). Clonus, a tonic phase, and writhing are conspicuous phenomena in convulsions induced in salamanders by both electrical and chemical agents (Peters and Vonderahe, 1953, 1954). Since clonic activity may be associated with the striatum or diencephalon (Peters, Vonderahe, and Hehman, 1959) it is probable that these centers of the brain have a lower threshold to stimulation by heat than the spinal cord which is responsible for writhing.

It is noteworthy that the heart continues to beat and show electrical activity for some time after the neuromuscular system has lapsed into electrical silence. This persistence of the electrocardiogram may find an explanation in the greater resistance of heart tissue to heat or to its protected location deep within the organism.

The results of this investigation offer further evidence for the view that not merely the range of temperature and the rate of change but the duration of exposure is an important factor in the modification of behavior of the survival of an organism, especially at higher temperatures (Orr, 1955). At the onset of the exposure to 44° C the neurological tests and the electrical records from the neuromuscular system showed no conspicuous abnormalities; longer exposure finally resulted in drastic and lethal changes within the organism.

SUMMARY

1. The salamander, *Triturus viridescens*, was subjected to a series of neurological tests during exposure to extreme cold, 0° C, or heat, 44° C. These tests involved walking, righting, corneal and abdominal reflexes, optokinetic nystagmus, response to touch, recordings of electrical activity of the neuromuscular system and the heart, and the reaction to the nervous stimulant, Metrazol.

2. Exposure for 30 minutes to either extreme cold or heat results in the loss of responses to external stimuli, eventual reduction in amplitude of the electrical record from the neuromuscular system, and no activation after an injection of Metrazol. The heart rate is decreased by exposure to cold, accelerated and made irregular by exposure to heat.

3. During the exposure to cold all responses become slower; as temperature falls, optokinetic nystagmus disappears first, corneal and abdominal reflexes last; as temperature rises they return in reverse order. All animals recover completely.

4. During the exposure to heat, after a seizurelike episode, all responses are lost suddenly and simultaneously. As temperature returns towards 24° C some responses are regained temporarily. All salamanders exposed to heat die within a few days.

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Landscapes of Alaska. *Howel Williams, Editor.* University of California Press. xii+148 pp. \$5.00.

The U. S. National Park Service joined with the U. S. Geological Survey to present the geological history of Alaskan landscapes, and the Press of University of California "did themselves proud" in publishing their report. Illustrations are superb, starting with a frontispiece in color of Mount McKinley, including a series of aerial photographs in black and white, and six maps (some in color) of the physiographic provinces of the new State. Chapters deal with these provinces one by one, each chapter written by an authority.

Three figures are of outstanding interest. One shows the evolution of OKmoh caldera on Umnak Island, another presents a glacial and permafrost map of Alaska, and the third includes four sketches which show the development of the Bering Sea Land Bridge.

An appendix includes a Geologic Time Scale and a glossary, and there is an excellent index. The study was well conceived and the report was well prepared. It makes good, interesting reading.

THOMAS H. LANGLOIS

The Gulf Stream. *Henry Stommel.* University of California Press. xiii+202 pp. \$6.00.

The purpose of this book is stated to be "to describe and explain what is known about the Gulf Stream in a way which will interest physical scientists, for but few scientists have any knowledge of the nature of this grand natural phenomenon." The author tries by means of this book to communicate the facts and theories concerning the Gulf Stream to a wide scientific audience.

The book presents records of early ideas and explorations and brings the history to date, then discusses methods of observation and instruments used. The amount of the Gulf Stream begins with a statement of the balance of forces at work with discussion of schematic density and pressure fields. Then follows a description of the large-scale features of the North Atlantic circulation and the hydrography of the Gulf Stream. A technical discussion of the wind system over the North Atlantic leads to the Linear and non-linear theories of the Gulf Stream, meanders in the stream, fluctuations in the currents, and the role of thermohaline circulation.

The concluding remarks present significant facts for the layman, such as the following:

"The Gulf Stream flows along the western boundary of the warm Sargasso Sea surface water. As the stream turns toward the east, off the Grand Banks, it acts as a kind of dynamic barrier, or dam, which, by virtue of Coriolis forces, restrains the warm Sargasso water from overflowing the colder northern water of the North Atlantic. The water in the stream is not significantly different in temperature from the large mass of warm water which lies to the right of its direction of motion. The Gulf Stream is not an ocean river of hot water. The intensity of flow of the Stream, the Stream's direction, and its temperature are not primary climatic factors in determining the climate of Europe; but the role which it plays in determining the northern boundaries and average temperature structure of the Sargasso Sea must be of critical climatic importance. . . . It is really a flow along the very edge of the juncture of a mass of cold water and a mass of warm water. . . . It is a boundary phenomenon . . . as a recognizable part of a larger physical system. . . . Therefore, one is led to inquire whether the primary physical phenomena to discuss are not the origin and nature of the two contrasting water masses, and whether it is not proper to regard the Gulf Stream current system as merely a secondary feature associated with their zone of contact."

THOMAS H. LANGLOIS