

## CENTRAL RESPONSE TO INFRA-RED STIMULATION OF THE PIT RECEPTORS IN A CROTALINE SNAKE, *TRIMERESURUS FLAVOVIRIDIS*

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### INTRODUCTION

The facial pits of boid and crotaline snakes are unique in the animal kingdom as infra-red receptors. The pits of the crotaline snakes, in particular, have excited the interest and imagination of naturalists from the time that rattlesnakes were first encountered in the early stages of colonization of the American continents (Klauber, 1956).

The snake family Viperidae is divided into two subfamilies by the presence or absence of pit receptors. The Viperinae, found throughout Europe, Africa and Asia, apparently lack any form of infra-red receptor (Bullock & Barrett, 1968). The Crotalinae, instead, are characterized by the presence of a highly specialized pit receptor in the loreal region of either side of the face. Five principal genera make up this subfamily: *Crotalus* (the rattlesnakes),\* *Bothrops* and *Lachesis*, all confined to North and South America; *Trimeresurus*, distributed widely in Asia; and *Agkistrodon*, which is found both in America and throughout Asia as far west as the eastern part of Europe. There are no crotalines in Africa or Australia.

The pits are found in a much more primitive form in the family Boidae; in fact, some boids possess thermoreception without any external pits at all (Bullock & Barrett, 1968).

The function of the boid pit is essentially the same as that of the crotaline pit (Noble & Schmidt, 1937; Warren & Proske, 1968). However, most research on snake thermoreceptors has been confined to crotaline snakes. The structure and innervation of these pits have been described in considerable detail by the following workers: Lynn (1931), Noble & Schmidt (1937), Bullock & Fox (1957), Bleichmar & De Robertis (1962), Terashima, Goris & Katsuki (1970).

That these pits are thermoreceptors responding to changes in background heat flux has been demonstrated amply by Noble & Schmidt (1937), using behavioural experimental techniques, and by Bullock & Cowles (1952), Bullock & Diecke (1956) and Goris & Nomoto (1967) using electro-physiological recording techniques. Terashima, Goris & Katsuki (1968) recorded the generator potential from the sensory membrane of crotaline receptors. Terashima *et al.* (1970) also determined the terminal nerve structure, and Meszler (1970) showed how changes in the densely packed mitochondria of the nerve endings indicate that these mitochondria are involved as transducers of heat energy to electrical potentials.

\* The pigmy rattlesnakes of North America, genus *Sistrurus*, are now included in the genus *Crotalus* (Underwood, 1967).

To date, however, all published works, with the exception of Harris & Gamow (1971), who recorded evoked potentials from the boid brain, have been concerned with peripheral responses. Much obscurity remains as to how the central nervous system receives these responses and how it utilizes them.

The work reported here was undertaken to study the response of the crotaline central nervous system to stimulation of the infra-red receptors; and to utilize this response to elucidate further the function of these receptors.

Three problems were posed:

- (1) To discover an area of the snake brain from which response potentials to infra-red stimulation could be recorded reliably and consistently.
- (2) To investigate the vertical and horizontal field of response of the pit receptors.
- (3) To investigate the possibility that the snake has stereoscopic perception of an infra-red stimulus source.

#### MATERIALS AND METHODS

The habu, *Trimeresurus flavoviridis*, was chosen as the most suitable experimental animal. It is the largest pit viper in Asia, and was available in quantity at moderate cost. *Trimeresurus okinawensis* and *Agkistrodon halys* were also considered, but were deemed unsuitable because of their high resistance to the immobilizing agent curare.

About 100 snakes were utilized, averaging *ca.* 130 cm in total length and 300 g in weight. The snakes were immobilized with tubocurarine chloride injected intramuscularly. Usually 0.3–0.6 mg was sufficient to immobilize snakes of the size used within 30 min. The amount did not seem to be critical, but we tried to obtain the lightest degree of immobilization possible. With this amount of curare the snake retained muscle tone and was capable of tail movements and other slight muscle twitches, but was unable to right itself when placed upside down. Under bright light the pupils contracted completely, although sluggishly. Respiration, evinced by opening and closing of the glottis, and by partial inflation of the lungs, took place about once a minute. We noted that as long as good pupil contraction persisted, good response could be obtained; but when immobilization progressed to the point that pupil contraction ceased, nerve potentials also became erratic or ceased. Therefore we used the pupil reflex as an indicator of the degree of immobilization and of the reliability of the recordings: once the pupil reflex became weak, the animal was no longer used.

Recordings were made with vinyl-coated tungsten electrodes and with glass micropipettes filled with 3 M-KCl. The tungsten electrode tips averaged about 5  $\mu$ m in diameter. The micropipettes were uniformly of 0.5  $\mu$ m in diameter, with a resistance varying between 20 and 50 M $\Omega$ . The potentials obtained were amplified in a conventional manner, displayed on a dual-beam oscilloscope, and recorded both on film and on magnetic tape. Infra-red stimulus was obtained from a small incandescent lamp and camera shutter, according to methods described by Terashima *et al.* (1968). The lamp was lighted by a 6 V direct current, and delivered, at a distance of 20 cm, approximately 9.74 mW/cm<sup>2</sup> with an energy peak at a wavelength of 1.2  $\mu$ m (Terashima *et al.* 1968). This will be referred to below as the 'standard stimulus'. The lamp and shutter were mounted on a pivoting brass arm which could be swung 180° in either a horizontal or a vertical plane (Fig. 1). A phototransistor attached to

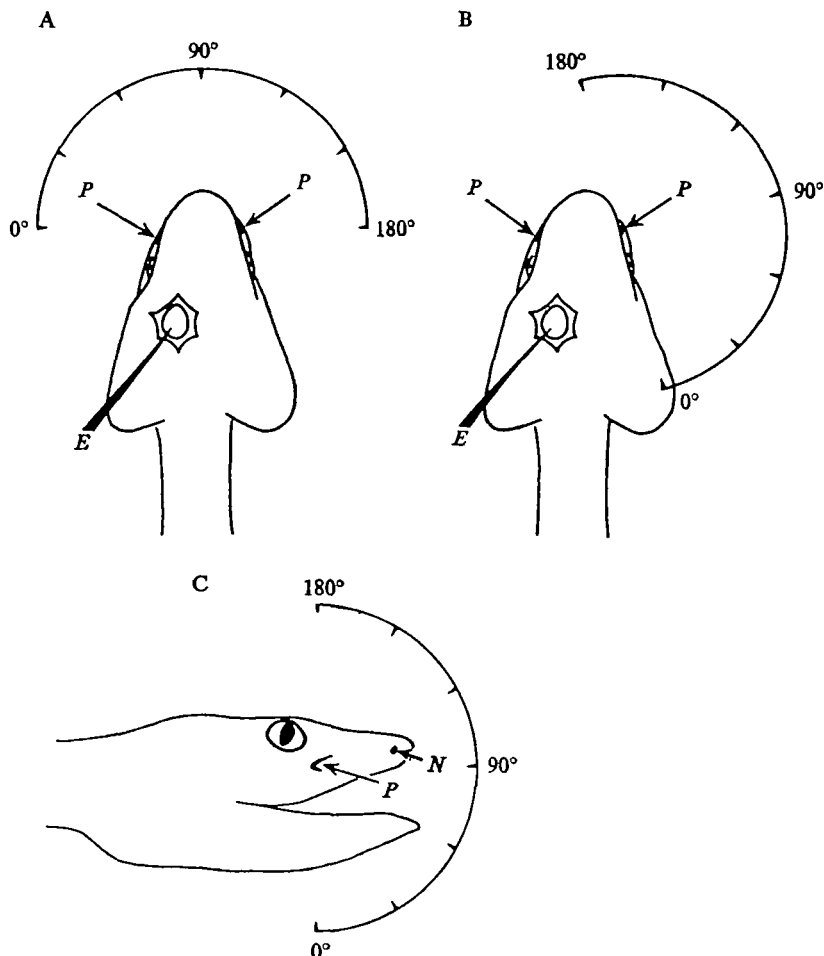


Fig. 1. Position of recording electrode and movement of stimulus course.

(A) Bilateral stimulation: arc indicates horizontal movement of stimulus. The 0–180° line bisects the pit openings. The 90° line is an extension of the midline body axis. *E*, electrode; *P*, *P*, pit openings.

(B) Contralateral stimulation: arc indicates movement of stimulus for recording horizontal receptive field. The 0–180° line is parallel to the plane of the pit opening. *E*, Electrode; *P*, *P*, pit openings.

(C) Arrangement for measurement of vertical response field: arc indicates vertical movement of stimulus. This arc represents a 90° vertical rotation of the arc in (A). *P*, Pit opening; *N*, nostril.

the front side of the shutter and connected to one beam of the oscilloscope indicated onset and duration of the stimulus. The head of the snake was mounted on a beam of balsa wood and centred directly above the pivot of the arm, and the lamp was adjusted on the arm so that the bulb was approximately 20 cm from this point, with the filament centred at the level of the pits.

Recordings were attempted from the trigeminal ganglion and the region of the tectum opticum. The trigeminal ganglion was approached dorsally, ventrally and laterally. This proved extremely difficult. The ganglion is well protected on all sides

by bony projections of the skull, and the removal of these was complicated by the relatively large blood vessels associated with the ganglion or passing through foramina in the bone. This difficulty was overcome by drilling very precise holes with fine dental burrs, but then these holes were too small to allow removal of the hard connective tissues surrounding the ganglion. This tissue was tough enough to break glass capillaries, and the tips of the tungsten electrodes usually bent instead of penetrating.

The trigeminal ganglion was also approached by entering from the occipital region. The neck muscles were cut with an electric scalpel at their point of attachment to the skull. A hole was opened in the occipital bones, and a slit was made in the dura. The cerebro-spinal fluid was then carefully blotted with absorbent cotton until the surface of the brain was clearly visible. Electrodes were inserted deeply at an angle of about  $15^\circ$  to the horizontal plane in hopes of penetrating to the neighbourhood of the trigeminal ganglion.

The most successful operation was exposure of the dorsal surface of the tectum opticum. The snake was fixed in a natural position on the board, with the snout protruding over the front edge as far as the region of the pits. The snake was held lightly to the board by a piece of tape at the nuchal region and a pin through the upper labials on each side. Since saliva often clogged the glottis, a plastic catheter was inserted to facilitate breathing. The skin of the head above the parietal bones was then excised. For a few moments after this operation the tectum opticum could be seen clearly through the bone, as well as the sinus venosus between the two lobes of the tectum. The bone became opaque as soon as it dried, but there was time to delineate precisely the area of bone to be removed. This area came extremely close to the sinus venosus, but the bone over the sinus had to be scrupulously avoided. The membranes over the sinus adhere to the bone, and if this part of the bone was drilled or moved the sinus would rupture and the snake would soon die. It was found impossible to stem the haemorrhage once this sinus had been ruptured. Using the finest available dental burr a series of cavities was drilled in a circle above one of the tectal lobes. The thickness of the bone in this area varies, so that considerable skill was necessary. The burr could not be permitted to penetrate to the dura, as this would tear the dura and rupture the blood vessels within, spoiling the preparation. Thus the holes had to be drilled down to, but not quite touching, the dura. All work was done under a dissecting microscope at a magnification of  $\times 15-20$ .

When the circle of holes was complete, the bone inside the circle was carefully and slowly chipped away with tweezers. Again great care had to be taken to avoid rupturing the dura. Once the dura was exposed, the course of the blood vessels through it could easily be seen. The dura was slit with fine scissors between the blood vessels and parallel to them. Thus, with a minimum of trauma and practically no haemorrhage an area of cortex 1-2 mm in diameter was exposed. Into this both tungsten and microcapillary electrodes could be easily inserted. With the passage of time the surface of the cortex became hardened, and it was increasingly difficult to insert the electrode. However, this did not cause any great problems, since good response could usually be obtained from the beginning of the experiment.

## RESULTS

*A. Site of recording*

No satisfactory results were obtained from the trigeminal ganglion directly. When electrodes were inserted into the neighbourhood of the ganglion from the occipital region, some response to infra-red stimulation was obtained. However, the location of the electrode tip was imprecise, and it was difficult to obtain consistent results. Therefore data from these recordings will not be considered in this paper.

Response to infra-red stimulation was obtained clearly and repeatedly from electrodes inserted into the tectum opticum, or in contact with its surface. Consistently good results were obtained from 17 snakes. A total of approximately 100 units were observed. Of these, 50 preparations lasted long enough to make satisfactory recordings. Recordings were made with both glass electrodes and tungsten electrodes. Excellent action potentials were obtained with the glass electrodes, while tungsten electrodes, being of a greater tip diameter, recorded mostly evoked potentials, together with a few multi-unit action potentials. Similar potentials were recorded from both right and left tectal lobes, but for convenience of manipulation most recordings were made from the left lobe.

*B. Potentials recorded*

Single-unit action potentials were recorded with glass microcapillary electrodes penetrating the tectum opticum. The precise depth of penetration was difficult to determine because the brain surface was depressed when the electrode penetrated. However, the best results were obtained when the electrode was advanced about 1 mm after touching the surface at the centre of the tectum. Recordings were also made at other depths, both shallower and deeper, and throughout the entire exposed region of the tectal lobe.

A few of the recorded units responded to optical stimulation, or to vibration, or to touch and vibration. Continuously firing units of unknown affiliation were also seen. However, the vast majority of single units encountered responded only to infra-red stimulation of the pits. This was confirmed by covering the pits, whereupon response immediately ceased. Covering the eyes, stroking the facial region with a brush, tapping the table, pinching the body, etc., did not affect the response in any way. Further confirmation was obtained by comparison with optical, vibration, and touch units, which manifested entirely different modes of discharge.

When unilateral stimulation was being used, care was taken to shield the pit not being stimulated from the stimulus source.

The following discharge patterns were observed:

*Background discharge* (Fig. 2A). By background discharge we mean discharge observed when no special stimulus was present in the receptive field of the pit. All the infra-red units exhibited background discharge to some degree. In some units it was barely 1–2 spikes or even less per minute; in others it reached a frequency of 5–7 impulses per second, which approximates the background discharge of primary fibres (Goris & Nomoto, 1967). No completely silent fibres were encountered. Some fibres were relatively silent, but resumed background discharge after receiving a stimulus. The background discharge frequency of any one unit also showed considerable fluctuation, increasing and decreasing from time to time.

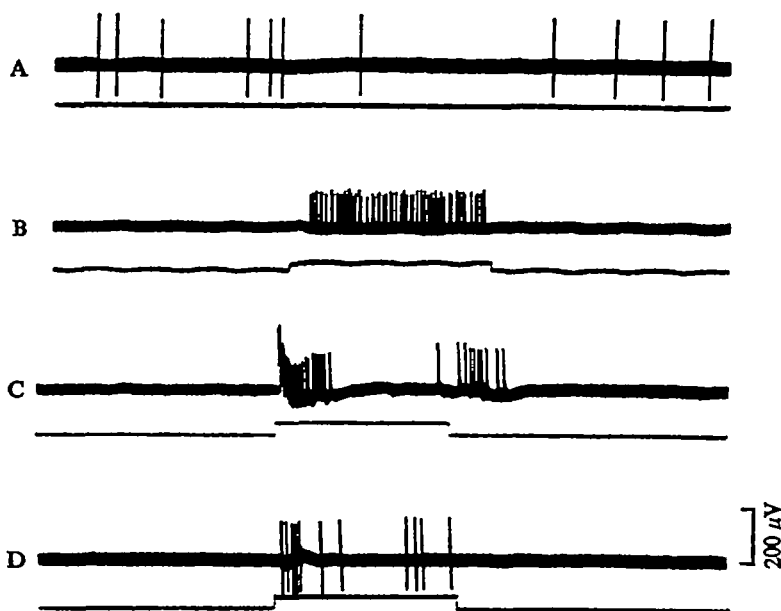


Fig. 2. Various types of central response: A, background discharge; B, tonic response; C, On-Off phasic response; D, phasic-tonic response. Lower line indicates stimulus. Stimulus duration 1 sec.

*Tonic discharge* (Fig. 2B). We recorded units which exhibited rapidly increasing firing after onset of stimulus up to a certain peak, then a gradual adaptation. For any given unit the frequency increased and decreased in proportion to the intensity of the stimulus. However, no two units showed the same frequency of discharge in response to the same stimulus. In response to the standard stimulus we recorded frequencies of from a few impulses per second to a maximum of 83 impulses/sec, each unit being different. Intervals between spikes were always highly irregular. There was no recognizable recurring pattern. There was also some fluctuation in frequency and intervals each time the stimulus was repeated. In some units the firing was more regular than in others; but all were characterized by a certain irregularity. When the stimulus was prolonged for several seconds, adaptation rapidly took place, eventually reaching the original background firing level. When the stimulus source was moved in a horizontal or vertical arc within the receptive field of the pit, frequency and firing pattern varied considerably with each new position of the stimulus (Figs. 3, 4).

*Phasic discharge* (Fig. 2C). A large number of neurones exhibited On-Off phasic discharge. In some of these both On and Off were represented by a single spike, regardless of the stimulus intensity or position. In these cases all possibilities of stimulus artifact were carefully excluded.

Other units showed an On burst and an Off burst. The number of spikes in the bursts and the duration of each burst differed from unit to unit. Changing the intensity or position of the stimulus also caused a change in the number of spikes per burst. As in tonic response, the number of spikes per burst was directly proportional to the stimulus intensity. On bursts generally contained more spikes than Off bursts. When

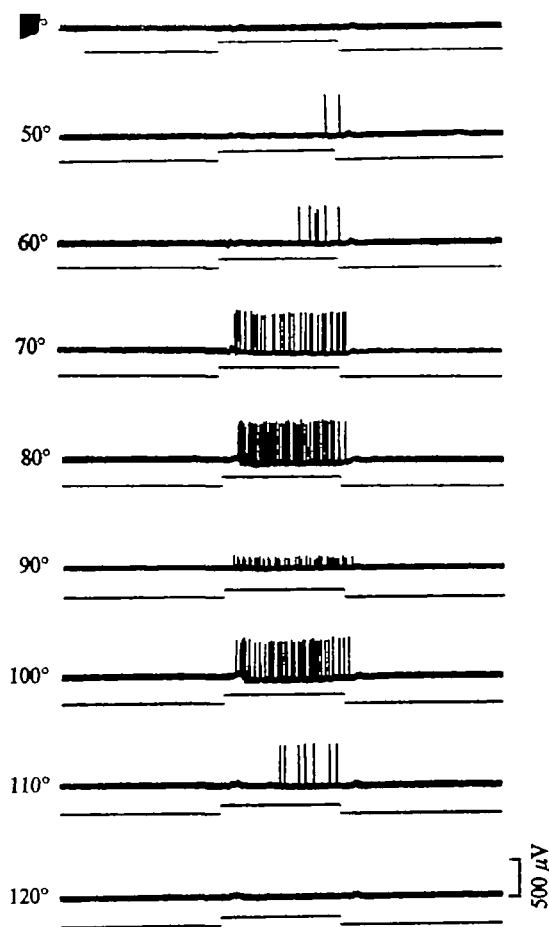


Fig. 3

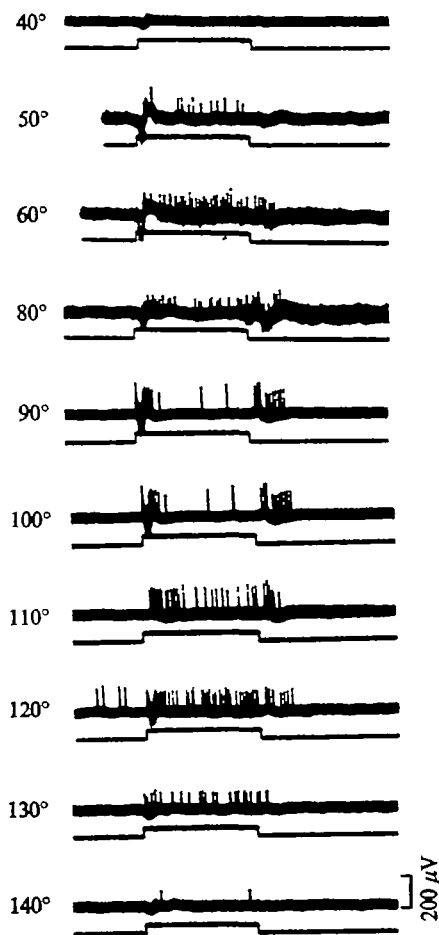


Fig. 4

Fig. 3. Typical example of horizontal response field. Degrees indicate positions of stimulus on arc of Fig. 1 B. Lower line indicates stimulus; standard intensity (see text); duration 1 sec. Lowered height of spikes at 90° is due to slight movement of electrode.

Fig. 4. Typical example of vertical response field. Degrees indicate positions of stimulus on arc of Fig. 1 C. Lower line indicates stimulus; standard intensity (see text); duration 1 sec. Record at 70° was lost.

stimulus intensity was reduced, the Off burst often disappeared, while the On burst remained.

*Phasic-tonic discharge* (Fig. 2 D). With the exception of single-spike On-Off units, no units were purely phasic. That is, although sometimes only On-Off bursts could be observed, varying the position of the stimulus would bring out tonic firing between On-Off bursts. This firing could be of high or low frequency; but for every phasic unit a stimulus position could be found where tonic firing also appeared.

In some units the phasic nature was predominant; in others the tonic nature was predominant. But even in these latter units the phasic nature of the initial burst was undeniable.

The vast majority of the units recorded showed this phasic-tonic mode of response.

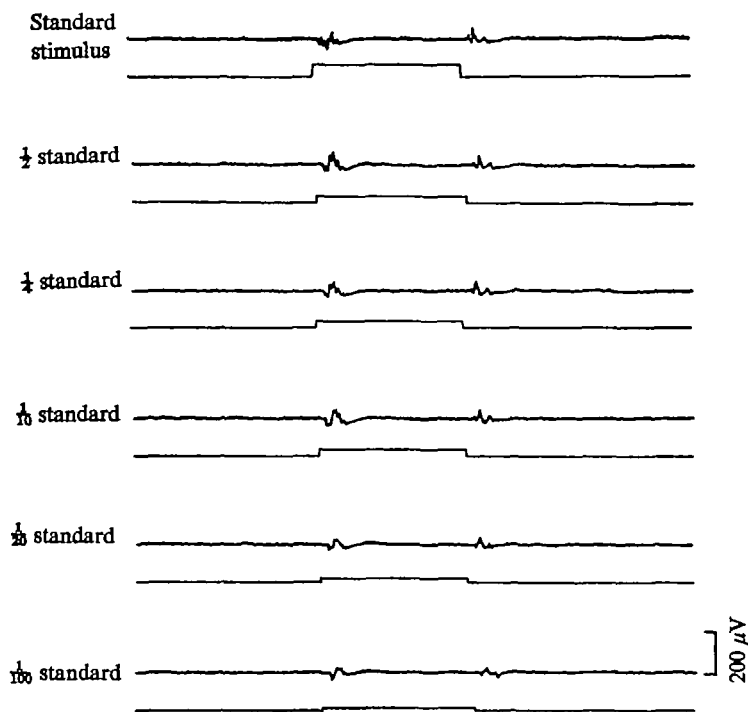


Fig. 5. Evoked response from the surface of the tectum. Tungsten electrode. Lower line indicates stimulus. Stimulus duration 1 sec.

Purely tonic units were relatively few; and it is possible that if the stimulus for these units had been moved sufficiently, phasic-tonic response would have been recorded. In fact, with some units where the stimulus was moved in a  $180^\circ$  arc, the response was tonic in some positions and phasic-tonic in other positions (Fig. 4). It was also possible for bilaterally responding units (see § E below, also Fig. 6A) to be, for example, tonic in response to ipsilateral stimulation and phasic-tonic in response to contralateral stimulation. In many phasic-tonic units there was no Off burst observable.

Phasic action potentials were usually accompanied by a clear evoked potential at the On position, presumably representing the On bursts of surrounding units.

When tungsten electrodes were lightly placed upon the surface of the tectum, very clear phasic evoked potentials (EP) were recorded in response to the standard stimulus (Fig. 5). There was always an On wave and an Off wave, the On wave being slightly larger than the Off wave. These EPs seem to correspond to the On-Off bursts of action potentials.

### C. Latency of response

Latency varied considerably from unit to unit. In response to the standard stimulus latency varied from less than 25 msec to several hundreds of milliseconds. For a given unit, if the stimulus was not moved, latency varied inversely with stimulus strength, although not linearly. Both action potentials and EPs showed this phenomenon (Table 1). When the stimulus source was moved in an arc, latency varied greatly from position to position, suggesting that changes in position caused changes in



Table 1. *Latency of response with decreasing stimulus strength; single preparation; wide-band heat filters used*

Stimulus	Action potential (msec)	Evoked potential (msec)
Standard	50	25
Standard reduced by		
50 %	65	40
75 %	75	50
90 %	112.5	50
95 %	No response	60
99 %	No response	65

stimulus intensity at the level of the receptor membrane. These changes were then manifested by changes in latency as well as in firing pattern.

#### D. *Sensitivity*

It was not the purpose of the present series of experiments to make sensitivity measurements. However, in contrast to the apparatus used when recording from peripheral nerves (Goris & Nomoto, 1967), recording from the brain left the receptive field of the pits relatively unencumbered. Hence some observations could be made on sensitivity, judged from the maximum distance at which a given stimulus would produce a response. The experiments were carried on inside a wire-mesh shield box to reduce external magnetic fields. The box was about 1 metre cube. Inside the box response was still obtained from the standard stimulus at the maximum distance possible within the box, about 90 cm from the snake's head. A hand also was detected at this distance. When the stimulus source was outside the shield box, response was severely attenuated or non-existent for most units recorded. This could be due to the interposed wire mesh, which was extremely fine and possibly acted as a heat sink. However, despite the wire mesh, several units were encountered which responded to the presence of a man standing outside the box at a maximum distance of 5 m. This was the maximum distance possible without leaving the room. Possibly response would have occurred at even greater distances if the room had been larger. With a man standing at 1 or 2 m response was tonic, with a few spikes per second, which gradually adapted. At greater distances adaptation was almost immediate and complete. However, any movement on the part of the man at these distances produced a response of one or two spikes. Waving the hands, moving the head from side to side, moving the body from the waist up all produced an initial response, which ceased immediately, even though the movement continued. When the type of movement changed or ceased, another response took place.

To exclude the possibility that the units thus responding were optical units, the experiment was repeated in total darkness. Response was unchanged. The eyes were covered and the experiment was again repeated; and again response was unchanged, leaving little doubt that a highly sensitive infra-red response was being observed.

#### E. *Ipsilateral-contralateral response*

The ipsilateral and contralateral pits were shielded from stimuli given to one or the other pit, and the responses to ipsilateral and contralateral stimuli were observed. It

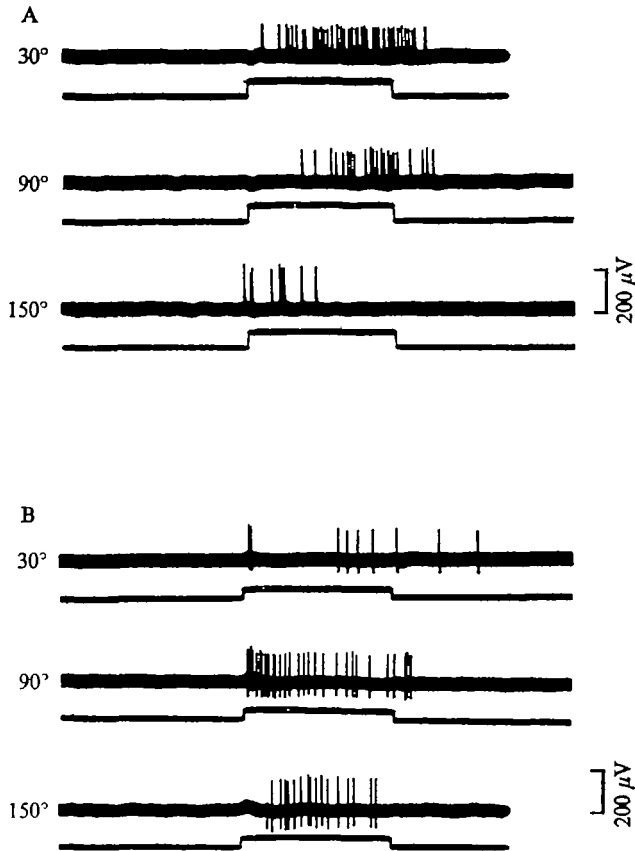


Fig. 6. Two types of bilaterally responding units; A, ipsilateral type; B, contralateral type. 30°, 90° and 150° indicate positions of stimulus on arc of Fig. 1 (A). Lower line indicates stimulus. Stimulus intensity: standard (see text); duration 1 sec.

was immediately noted (1) that most units responded to contralateral stimulus only, and (2) that some units responded to both ipsilateral and contralateral stimulation (Fig. 6). No purely ipsilateral units were encountered. When a bilateral unit was encountered, the shield was removed and the stimulus was adjusted so that the diameter of its arc was horizontal and perpendicular to the long axis of the snake's body, with the pivot centred between the pits, as in Fig. 1 A. The stimulus was then moved in an arc of 180° in steps of 5° or 10°, and the response was recorded.

With respect to the stimulus position at which maximum firing occurred, two types of bilateral units were encountered: (1) ipsilateral type – maximum firing occurred when the stimulus was in the field of the ipsilateral pit (Fig. 6 A), and (2) contralateral type – maximum firing occurred when the stimulus was in the receptive field of the contralateral pit (Fig. 6 B).

In most, but not all, of the bilateral units encountered, when the stimulus was in position to illuminate both pits equally, firing was reinforced to a frequency superior to that of either pit alone.

Table 2. *Horizontal width of receptive field; contralateral stimulation*

Unit no.	Width of field (deg. of arc)	Position of field	Maximum firing point
1	50	From 60° to 110°	80°
2	40	From 30° to 70°	40°
3	50	From 40° to 90°	70°
4	80	From 50° to 130°	90°
Av. 55			

Table 3. *Vertical width of receptive field; contralateral stimulation*

Unit no.	Width of field (deg. of arc)	Position of field	Maximum firing point
5	40	From 40° to 80°	70°
6	70	From 40° to 110°	70°
7	25	From 30° to 55°	55°
8	80	From 40° to 120°	70°
9	70	From 20° to 90°	50-60°
10	40	From 70° to 110°	100°
11	80	From 50° to 130°	110°
12	60	From 40° to 100°	60-70°
Av. 58			

#### F. *Effects of a cold stimulus*

While observing the background firing of a unit, ice was introduced into the receptive field. This caused immediate suppression of all discharge. When the ice was withdrawn, an On burst was produced which was similar to the On burst produced by a 'hot' stimulus. If a warm metal plate was introduced into the receptive field, it at first caused a phasic-tonic response. If the metal plate was allowed to remain in position, the unit rapidly adapted and firing returned to background levels. If at this point a hand was brought between the plate and the pit, the same effect as ice was produced, i.e. suppression of background firing, and an On burst when the hand was removed.

#### G. *Receptive field*

The receptive field was measured both horizontally and vertically. However, our apparatus was movable in only one plane, so that horizontal and vertical measurements had to be made on separate units.

Moving the stimulus source as shown in Fig. 1B and C, the measurements of Tables 2 and 3 were obtained.

In brief, horizontal field width averaged 55° (extremes 40-80°), and vertical width averaged 58° (extremes 25-80°) of arc.

#### DISCUSSION

The histology of the tectum opticum of the reptile brain has been studied in detail by Senn (1968) (*Lacerta sicula*), and by Leghissa (1962) (all classes of reptiles). According to these authors the surface of the tectum is formed by bundles of optic fibres, while the layers beneath the surface contain vast amounts of other fibres, many

of which derive directly from, or communicate with, the trigeminal nerve. Many of these fibres communicate with the contralateral side across the intertectal commissure on the dorsal side of the tectum. There is another commissure on the ventral side of the tectum, the tegumental commissure, but it is not clear whether fibres of the trigeminal system communicate across this commissure. In addition, at about the level of the 'stratum griseum et fibrosum periventriculare' of Leghissa, there are a number of large nuclei. These are the nuclei of the mesencephalic tract of the trigeminal nerve.

According to Leghissa (1962) 'the optic tectum represents the centre of arrival and departure of impulses that control the general nervous activity and the behaviour of non-mammiferous animals, which do not have a differentiated hemispherical neo-cortex... The movement of an animal towards its prey, the swallowing of acceptable food and the rejection of unacceptable food, the reactions of attack and defence, and finally the presence or absence, in an animal, of an associative memory all find in the optic lobe a most favourable morphological basis for realization. In the most highly evolved tetrapods (reptiles and especially birds) the tectum opticum acquires also an integrative function, analogous to that possessed by the mammals in the cortex of the hemispheres' (pp. 344-5 *passim*; translation by Goris).

Masai & Sato (1965, 1971), Sato (1971), Wells, Smith & Spaur (1971) and Masai (in preparation) have demonstrated how the relative degree of development of a given part of the brain (e.g. the olfactory lobes) in related species of lower vertebrates is directly related to the degree or extent that each species makes use, in its daily life, of the senses directly connected with that part of the brain. Thus, to give an example, a snake which depends primarily on the sense of smell to detect food will show a high degree of development of the olfactory lobes. In contrast, there will be a lesser degree of development of these lobes in a snake which feeds primarily by sight (Wells, Smith & Spaur, 1971).

Masai (1972) has examined the tectum opticum of a number of typical snakes, mostly colubrids and natricids, and has shown that the degree of development of the tectum is directly related to the diurnal or nocturnal habits of the species. In *Elaphe climacophora*, an almost entirely diurnal species, the tectal lobes are very highly developed (Fig. 7A) and present a distinct swelling in a dorsal view. In frontal section the fibrous layers of these lobes are seen to be very highly developed. In contrast to this diurnal snake, *Dinodon rufozonatus*, of strictly nocturnal habits, has very poorly developed tectal lobes (Fig. 7B), and in section the fibrous layers are less developed than *E. climacophora*.

*Trimeresurus flavoviridis*, the subject of the present experiments, is active primarily at night, with a few periods of activity in early morning and evening during cooler weather. However, its tectal lobes show just as high a degree of development as the diurnal *E. climacophora* (Fig. 7C). Compared with *D. rufozonatus*, the eyes of *T. flavoviridis* are larger and probably used to a greater extent. One would expect, therefore, a degree of development of the tectum opticum intermediate between *D. rufozonatus* and *E. climacophora*. However, this is not the case. The tectal lobes of this species show a degree of development nearly equal to that of *E. climacophora*. Now, the only other difference between *T. flavoviridis* and *D. rufozonatus* is the possession of the infra-red sensory pits, supplied by highly developed branches of the trigeminal nerve.

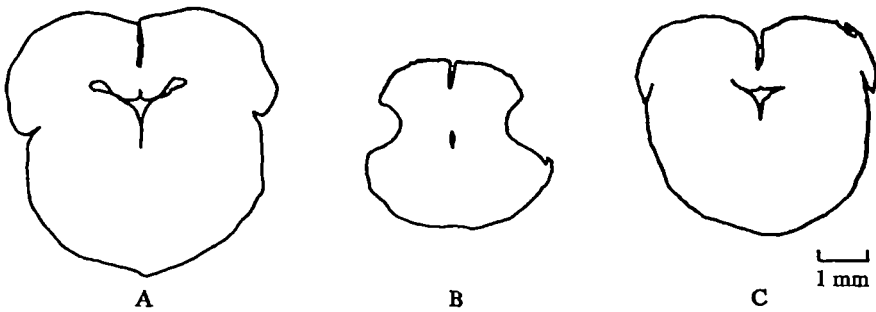


Fig. 7. Transverse sections through the tecta of three snakes; A, *Elaphe climacophora*, B, *Dinodon rufozonatus*, C, *Trimeresurus flavoviridis*.

This species relies on the pits to a high degree for appreciation of its surroundings and for the detection and capture of prey. Since the trigeminal system, which supplies the pits, enters and is integrated at the level of the tectum opticum, it is reasonable to suppose that the hypertrophy of the optic lobes in this species is a reflexion of the existence of the pit organs and the high degree of their use.

The results of our experiments confirm this theory. Below the immediate surface of the tectum known to be made up of retinal fibres, more than 90% of the response obtained, both with tungsten and glass electrodes, was infra-red response, presumably from the trigeminal system or in direct communication with it.

Further determination of the precise pathways of impulses from the pit to the tectum and inside the tectum must await the results of nerve degeneration experiments now in progress. However, the following conclusions can be stated with reasonable certainty.

#### A. Nature of the response

The response obtained from the brain is post-synaptic, and not primary, for the reasons that impulse patterns are totally different from those obtained by Terashima *et al.* (1968) and by Goris & Nomoto (1967) from receptor endings and primary fibres, respectively. According to these workers, primary firing patterns are either tonic or phasic. They are normally tonic in response to stimuli of the type arising in nature – for example, a bird in the snake's receptive field. The frequency of this firing increases in direct proportion to the intensity of the stimulus. If the stimulus is prolonged and steady, the firing frequency also remains steady and the intervals between spikes are quite regular (Fig. 8A). When the stimulus passes a certain threshold of intensity, peripheral response suddenly ceases to be tonic, and instead assumes a phasic i.e., On-Off, firing pattern (Fig. 8B). However, although the firing frequency is greatly accelerated (80–100/sec), the intervals between spikes remain as regular as in tonic response.

Both phasic and tonic responses were also recorded centrally (cf. Fig. 2). However, several differences from peripheral response were immediately apparent.

First, in both phasic and tonic firing, intervals between spikes were highly irregular. Very long and very short intervals succeeded each other almost at random. Highly accelerated bursts of 5–10 spikes could be followed by less rapid bursts, or by single spikes at varying intervals.

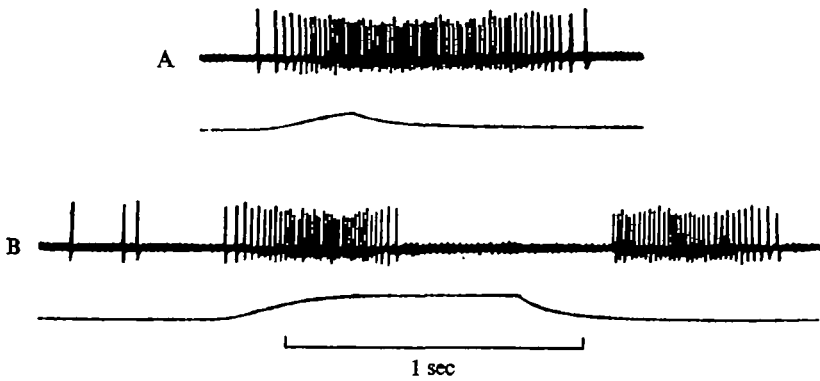


Fig. 8. Typical peripheral response: A, tonic; B, phasic.  
Lower line indicates stimulus.

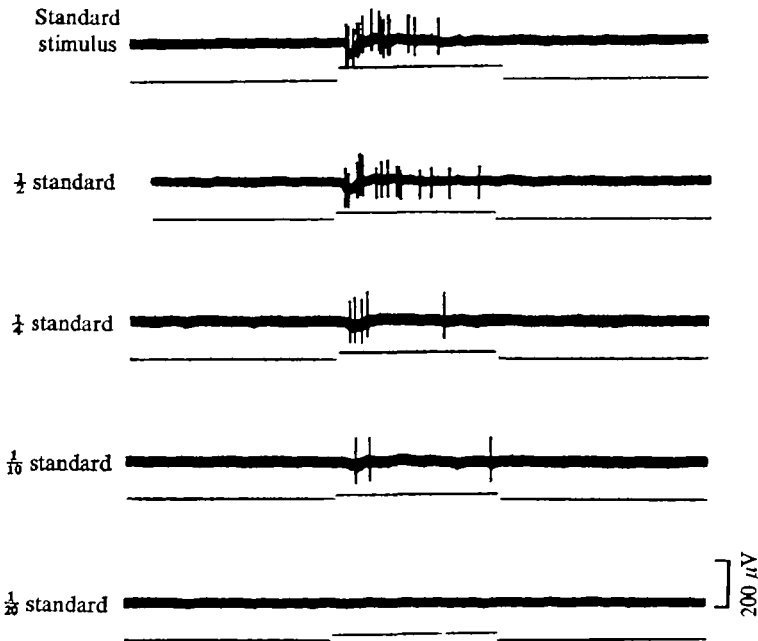


Fig. 9. Effect of reduction in stimulus intensity. Lower line indicates stimulus.  
Stimulus duration 1 sec.

Secondly, in peripheral response phasic response occurred only when a certain intensity of stimulus had been reached. It disappeared again when the stimulus intensity was reduced. Centrally, phasic response had no direct relation to the intensity of the stimulus. Units which showed phasic response retained that pattern no matter how much the stimulus was reduced. The spike frequency in each phasic burst varied with the intensity of the stimulus, just as in tonic response. However, the phasic nature of the response remained, as long as the stimulus source remained unmoved (Fig. 9).

A third difference was the appearance of phasic-tonic units. In these units firing

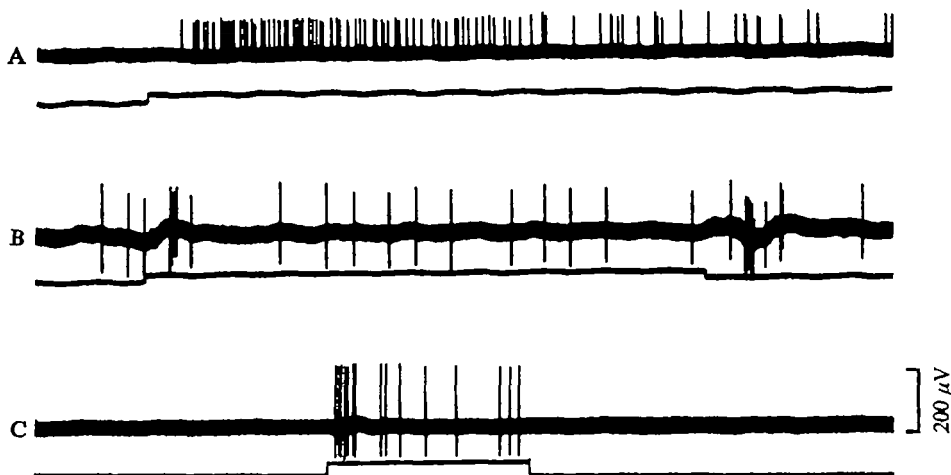


Fig. 10. Example of differing responses to the same stimulus by three different units. Stimulus was a hand at 20 cm. Lower line indicates onset and duration of stimulus. Stimulus at C is 1 sec. A is tonic response with fast adaptation. B and C are phasic-tonic responses. An Off burst occurs in B but not in C.

began with a phasic burst followed by a tonic train of firing. An Off burst was also often encountered in this type of phasic-tonic combination. In peripheral response such a combination was never encountered at any time.

A fourth difference was the fact that an identical stimulus, such as a hand at 20 cm, produced different types of response with different units (Fig. 10). The response could be either phasic or tonic or phasic-tonic. Such a difference suggests the possibility that recording was being made from several different types of units. However, peripherally all units were uniform (Terashima *et al.* 1968, 1970), and no evidence was found for the possible presence of different types of receptors.

Finally, the existence of bilaterally responding units with firing patterns similar to those of the unilateral units supports the supposition that all these units are not primary, for such bilateral units could only be multiple-input, post-synaptic units.

#### B. Background discharge

The term 'spontaneous discharge' was used by Goris & Nomoto (1967) to describe firing in the absence of any special stimulus in the receptive field of the pit. In the light of the present studies we believe that the term 'background discharge' is more appropriate, for the reasons set out below.

Bullock & Cowles (1952) and Bullock & Diecke (1956) showed that the pit functions by signalling changes in the radiation flux impinging on the receptor membrane. The work of Goris & Nomoto (1967) supported this observation. All of these workers showed how an object with a higher radiation flux than the background caused an increase in firing frequency, while an object of lower temperature, and hence lower radiation flux than the background, diminished or suppressed completely the background firing present in the nerve fibre. Thus ice suppressed the normal background discharge; and when a metal plate heated to 60 °C was set in the field of the pit and the pit was allowed to adapt, a hand produced the same effect as the ice – that is, complete suppression of the discharge (Goris & Nomoto, 1967).

We repeated the experiments of Goris & Nomoto (1967) while recording from the tectum, with identical results. Background discharge was present, in varying degrees, in every unit that we recorded. The firing pattern was essentially identical with that of peripheral units – that is, random firing, without any recognizable recurring pattern. It differed from peripheral background firing in that firing frequency was often considerably lower at central levels, being sometimes only 1–2 spikes/min; some units, however, approached the peripheral frequency of 5–6 impulses/sec. Adaptation – that is, a return, during stimulation, to pre-stimulus levels of discharge – was also much faster at central levels. Several minutes were required for adaptation at the peripheral level when the stimulus was a hand at 20 cm. At central levels tonically firing units adapted to the same stimulus in a matter of seconds (Fig. 10A).

We propose the theory that background firing represents a level of adaptation to the flux of background radiation. Any change in the flux causes a change in the firing pattern. Thus central units adapted quickly to a human body which entered the receptive field, but any movement of the limbs or trunk produced a noticeable, though transient, response.

We propose that there would be no background discharge if there were no background radiation, a condition which normally cannot exist. A similar condition is created temporarily when ice blocks the receptive field and acts as a heat sink, absorbing the background radiation. In this case background discharge disappears. When the ice is removed, there is a sudden On burst of firing similar to the On burst in response to a sudden stimulus. This is because the membrane receptors, now no longer adapted to the background radiation because of the ice, suddenly are confronted with this background radiation and react accordingly.

### *C. Field of response*

The configuration of the field response seems to be determined not so much by the nature of the receptors themselves but by the physical configurations of the pit – that is, the shape and width of the opening, the distance from opening to sensory membrane, the orientation of the plane of the opening, etc. These factors vary widely from species to species, and slightly even from individual to individual. Thus no absolute figures can be given.

In our experiments, as long as the stimulus was not moved, repeated stimuli produced repetitions of the same firing pattern. However, when the stimulus was moved, the firing pattern was different for each position (Figs. 3, 4). Assuming that all the receptors in the receptor membrane are identical and produce a response proportional to the amount of radiation received, the variation in firing pattern with a moving stimulus can be explained by a combination of the following two factors:

(1) The unit being recorded from is a multiple-input unit in the integration centre of the brain, possibly several synapses removed from the peripheral nerves.

(2) The amount of radiation striking the various receptors in the membrane is not the same for each receptor. This is caused by the physical configurations of the pit. In the first place, the membrane is not taut and flat but hangs loosely inside the pit in a more or less concave fashion, so that no two points on the membrane are exactly equidistant from the plane of the opening. In addition to this, the opening itself is quite irregular in shape, with considerable overhang of the scales lining the edge of



the pit. Thus with the stimulus at a given position, a considerable pattern of light, shadow and reflexion is formed on the surface of the membrane, with each individual receptor receiving different amounts of radiation. When the stimulus is moved, the pattern of light and shadow shifts across the membrane, so that the amount of radiation received by each individual receptor, and consequently the firing pattern which reaches the central nervous system also shifts accordingly. The effect is analogous to that of a pattern moving across the compound eye of an insect, although the pit receptors seem far too simple to even remotely approach the image-resolving power of an insect eye. Not only does the pit lack a lens, but the receptor population is completely homogeneous, as has been noted by Terashima *et al.* (1970), without any known inter-reactions such as facilitation or inhibition. However, it is apparent from our results that the general direction of a stimulus, as well as its movement and direction of movement, can be easily perceived even by a single pit.

Our results also support the supposition that the crotaline snakes have stereoscopic perception of stimulus objects. The existence in the tectum of bilaterally responding units is cogent proof of this. All of the bilateral units recorded showed a change of response pattern when the stimulus was moved from ipsilateral to contralateral pit. Some units showed maximum firing with ipsilateral stimulation, others with contralateral stimulation. Most showed reinforcement (increase) of firing when the stimulus was in a position to irradiate both pits at once (Fig. 6). Thus it is evident that the snake not only perceives an object as being simply to its right or to its left, but is also able to determine the position of an object within the overlapping receptive field of the two pits by comparing the discharge between right and left pits. The scanning behaviour noted by Goris & Nomoto (1967) is evidence that precisely this type of comparison is being carried out. The accuracy of this orientation was shown long ago by the behavioural experiments of Noble & Schmidt (1937).

#### SUMMARY

1. Both action potentials and evoked potentials were recorded from the tectum opticum of a crotaline snake, *Trimeresurus flavoviridis*, in response to infra-red stimulation of the facial pit organs. Action potentials from single units were recorded throughout the tectum.
2. Most units responded to contralateral stimulation, while some responded to both ipsi- and contralateral stimulation.
3. Firing patterns were tonic, phasic, or phasic-tonic, depending on the position of the stimulus and the type of unit being recorded.
4. Sensitivity to stimulus movement was observed.
5. All potentials differed from peripheral potentials in firing patterns.
6. Firing frequency was directly proportional to stimulus intensity.
7. Measurements were made of the vertical and horizontal response fields of single units.
8. Background discharge was noted in all units and its nature discussed.
9. The integrative function of the tectum in regard to infra-red perception was also discussed, as well as the possibility of stereoscopic perception.

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