J. Physiol. (1965), **177**, *pp.* 377–397
With 10 *text-figures*
Printed in Great Britain

COMPONENTS OF RECEPTOR ADAPTATION IN A PACINIAN CORPUSCLE

BY W. R. LOEWENSTEIN AND M. MENDELSON*
From the Department of Physiology, Columbia University, College of Physicians and Surgeons, New York, U.S.A.

(*Received* 12 *August* 1964)

The sensory nerve ending in the Pacinian corpuscle is surrounded by a lamellated fluid-filled capsule. The capsule, as such, does not partake in the receptor process of mechano-electric conversion; this appears to take place entirely within the nerve ending, the sensor proper (Loewenstein & Rathkamp, 1958). But the capsule is clearly the coupling element between the external stimulus and the sensor, and the character of the sensor response must depend on the mechanical properties of this coupling. The present study concerns this coupling, particularly its role in receptor adaptation.

At the level of the earliest detectable sensor response, the generator potential, adaptation manifests itself as a decline in potential in the face of a sustained mechanical stimulus. In the Pacinian corpuscle, the decline is complete within a few milliseconds, leaving time for only one or, at best, two nerve impulses to be discharged by a single stimulus. In mechanoreceptors, in general, the rate of generator-potential decline seems closely related to the rate of decline of impulse frequency (Katz, 1950; Álvarez-Buylla & de Arellano, 1953; Gray & Sato, 1953; Eyzaguirre & Kuffler, 1955). Apparently, the receptor adaptation process, or at least a significant component of it, resides at the level of generator potential production. The mechanisms of the adaptation process are still largely unknown. A hypothesis was proposed by one of us some time ago (Loewenstein, 1956). In this hypothesis (hereafter referred to as 'mechanical hypothesis'), the adaptation rate is considered to depend on mechanical properties, namely visco-elastic properties, of the coupling between receptor element and external stimulus. Support for the hypothesis was derived from work on stretch receptors of crustacean muscles (Eyzaguirre & Kuffler, 1955) and touch receptors of the frog skin (Loewenstein, 1956) in which differences in adaptation rate appeared to be due to mechanical properties of the adventitious tissues rather than to basic electro-chemical ones of the sensor proper.

* Present address: Department of Physiology, New York University, College of Medicine, New York.

In the case of the touch receptor, for instance, adaptation was found to depend on the elastic tension of the skin tissues surrounding the sensory ending. Support along similar lines came later from work on cat muscle spindles (Lippold, Nicholls & Redfearn, 1960) and crustacean stretch receptors (Krnjević & van Gelder, 1961; Wendler & Burkhardt, 1961) in which adaptation was found to be determined, at least in part, by mechanical factors. Further support for the mechanical hypothesis came from Hubbard's (1958) experiments measuring displacement of the peripheral lamellae of Pacinian corpuscles during compression. Hubbard showed that static components of lamella displacements resulting from compression attenuate markedly from periphery to centre of the corpuscle; while dynamic components attenuate less. The time course of the dynamic component of lamella displacement is of the same order of magnitude as that of the generator potential. But, unfortunately, displacements could be measured only at peripheral lamellae, and the relative spatial distributions of lamella displacements had to be extrapolated from the periphery with wide fluid-filled spaces to the much more solid core wherein the sensor is located.

In the present work, we have taken another approach. The laminar capsule was to a large extent by-passed as a mechanical coupling: the capsule was eliminated by dissection, and the external stimulus applied more directly to the sensor. After a substantial reduction in intermediary mechanical coupling as represented by the capsule, and in absence of other mechanical filters, one expects in the light of the mechanical hypothesis that the time courses of external stimulus and generator response be more closely related than in the normally encapsulated receptor. This, in fact, turned out to be the case.

A preliminary report of the work has already appeared (Mendelson & Loewenstein, 1964).

METHODS

Isolated Pacinian corpuscles from the mesentery of the cat were used in the experiments. The corpuscle and a length of its nerve were freed by dissection from mesentery and fat tissues. This preparation will be referred to as *intact* corpuscle. For resection of the capsule, lamella layers were stripped off progressively from periphery to centre of the capsule. (Details of technique are given by Loewenstein & Rathkamp, 1958.) In this way, preparations of sensory endings were made available for stimulation which had lamella sheaths varying from about 800 μ in diameter, nearly the diameter of the intact corpuscle, to about 50 μ, the diameter of the inner core. These preparations will be referred to as *decapsulated* corpuscles. (For a recent description of the structure of Pacinian corpuscles, see Pease & Quilliam, 1957.)

The preparation was set up in a small drop of Krebs's solution surrounded by mineral oil (Fig. 1). The nerve fibre of the capsule was lifted into the oil so that the Krebs's solution–oil interface came to lie at about the place where the remaining outermost lamellae joined the axon. This was one lead-off point of the receptor's electrical activity. The other one was a more central region of the nerve on to which a platinum electrode was placed. The electrical

activity of the receptor was fed into an oscilloscope through a condenser-coupled amplifier of 18 μsec rise time and 0·9 sec decay time constants.

The receptor was stimulated by rapid compression. The preparation was compressed between two smooth, parallel glass surfaces. One was the surface of a fixed glass plate on which the preparation lay with its long axis parallel to the surface; and the other was the flattened surface of a glass stylus of about 1·2 mm in diameter attached to a piezo-electric crystal (Fig. 1). The crystal was driven with electrical pulses of varying wave forms, so that pulses of displacement of varying rates of rise and durations were available for compression.

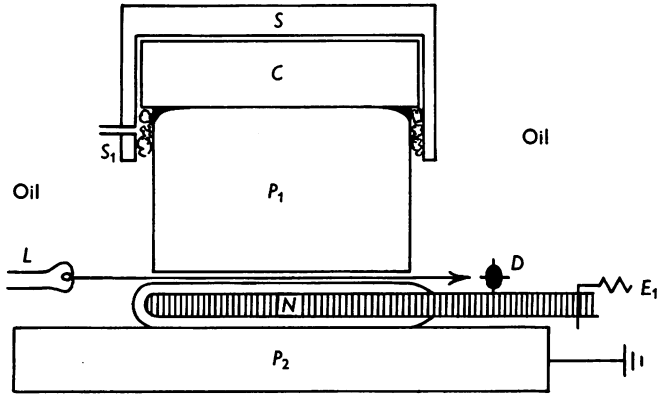

Fig. 1. Diagram of apparatus. *N*, decapsulated nerve ending; P_2, fixed plate; P_1, movable plate driven by Rochelle-salt crystal *C*; *L*, miniature light source with focused beam; *D*, photo-transistor; *S*, metal housing of crystal serving as electromagnetic shield. Paraffin is injected through S_1 into the space between P_1 and *S* for viscous damping. The core lamellae of the ending are covered by a film of Krebs's solution not shown on the diagram. Diagram not drawn to scale.

The displacements of the stylus were monitored with a photo-electric system consisting of a silicon photodiode (Photo-Duo Diode NPN, Texas Instruments, Inc.) and a miniature light source (Pinlite L 14–45, Kay Electric Co.) affixed to the housing of the crystal stimulator. The photo-electric system occupied very little space (1·5 × 1·5 × 4 mm) and the entire stimulator-monitor assembly could be manoeuvred with ease. The photocurrent-displacement relation of the system was linear over the range used, and stylus displacements could be monitored with an accuracy of 0·09 μ. Receptor response and external stimulus were displayed simultaneously on separate beams of an oscilloscope.

With the stimulator system in air, considerable ringing occurred when the crystal was driven with single electrical pulses of fast rates of rise. Some degree of ringing persisted also in the usual working position of the stimulator, namely with the stylus partly in oil and in contact with the preparation. For the experiments in which relations between receptor response and (single) stimulus durations were examined, it was essential, however, that there be excitation strictly by one stimulus. The stimulator system was therefore damped. For this purpose a petroleum jelly was injected into the sleeve around the stylus until stimulus after-oscillations were no longer detectable with the photo-electric system. This ensured that there were no oscillations greater than 900 Å. Stimuli at least one order of magnitude greater than this were required to produce detectable generator potentials under the most favourable conditions.

In the remainder of the experiments, in which some oscillation could be tolerated, damping was achieved by mixing electrical pulses of appropriate polarity, amplitude, duration

and delay with the primary pulse. This provided stimuli of faster rates of rise than those obtained by the above method of damping. The two kinds of damping will be referred to in the results section as 'viscous' and 'electrical' damping. Examples of stimuli obtained by viscous damping are given in Fig. 2, and by electrical damping, in Fig. 3.

In the experiments in which it was desirable to record generator potentials uncomplicated by action potentials, the latter were blocked by applying procaine (0·5 g in 10^2 ml.) (Katz, 1950), or tetradotoxin (1 g in 10^5 ml.) (Loewenstein, Terzuolo & Washizu, 1963), to the preparation, or by stretching or compressing intracorpuscular regions of the myelinated axon. Blocking by tetradotoxin was the preferred procedure, because it does not affect the generator potential. However, no basic differences were noted in the results obtained with the three procedures.

All experiments were done at room temperature ranging between 20–25° C. The Krebs's solution had the following composition: NaCl, 115 mM; KCl, 4·60 mM; $CaCl_2$, 2·46 mM; $MgSO_4$, 1·15 mM; $NaHCO_3$, 24·1 mM; KH_2PO_4, 1·15 mM; glucose, 10 mM.

RESULTS

Mechanical component of adaptation

Special properties of the receptor response after decapsulation

Response duration. The generator potential of an intact corpuscle in response to a mechanical stimulus is continuously variable in its amplitude with that of the stimulus, but in its time course it bears little relation to the stimulus. Typically, the potential, as recorded in the present experiments, lasts for about 6 msec, and this duration is the same whether the potential is elicited by a stimulus of long or short duration (Gray & Sato, 1953). Figure 2 shows an example. After removal of the capsule, the response acquires a different aspect. The generator potential is now related in its time course to the mechanical stimulus. Figure 2 shows this for the case of a corpuscle decapsulated to the core. Here the generator potential follows the stimulus over a wide range of durations. The potential still decays, as is evident with long stimuli, but the rate of the decay is of a different order of magnitude from that of the intact corpuscle; and the phases of compression ('on') and decompression ('off') of the stimulus have clearly discernible counterparts in the response over a range of durations of 60 msec. In general the extent of this range depended on the amount of lamella tissue left around the nerve ending. In our cleanest decapsulated preparation, which still had some lamellae left, the generator potential could be prolonged up to about twelvefold with respect to that of the intact corpuscle.

We have made no systematic study of the relation between maximal response duration and number of lamellae left after decapsulation. This would require a systematic study in one and the same capsule which is technically difficult. But from information obtained in different corpuscles stripped down to varying numbers of lamellae, we have gained the impression that removal of lamellae in the region just outside the core where the lamella spacing begins to be close has a greater effect on the duration of the generator potential than within the core itself or within the outermost lamellae separated by wide fluid-filled spaces.

When stimuli of fast rates of rise were used, and particularly in incompletely decapsulated preparations, the response had an initially fast falling phase followed by a more slowly decaying one. But even then, the total duration of the response was markedly prolonged. An example is illustrated in Fig. 3.

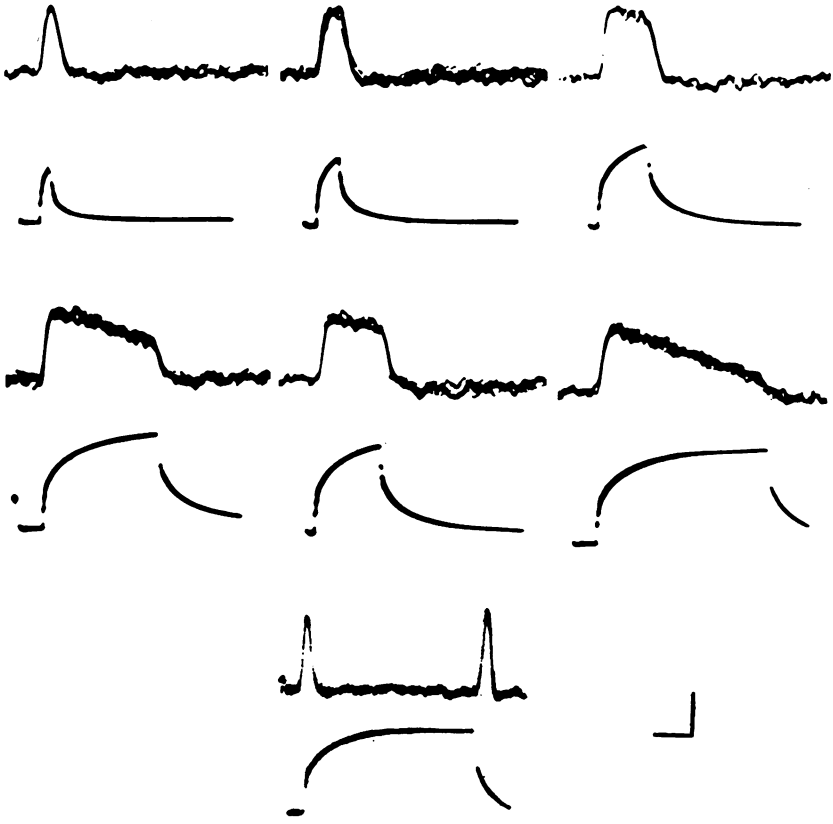

Fig. 2. Prolongation of receptor response after removal of capsule. Generator potentials of an intact corpuscle (lower row) and after decapsulation (upper two rows). Mechanical stimuli are of relatively slow rates of rise (viscous damping). Impulse production blocked by procaine. Upper beam, receptor response; lower beam, photo-electric record of mechanical stimulus in this and subsequent figures. Each record contains two to seven responses superposed. Note absence of off-generator potentials in the decapsulated preparation. Calibration for all records: 10 msec; 50 μV upper beam.

Response velocity. The response of the decapsulated corpuscle follows the stimulus closely also in its rate of rise. Figure 4 illustrates this for an experiment in which stimuli of equal intensity and varying rates of rise were applied to the receptor.

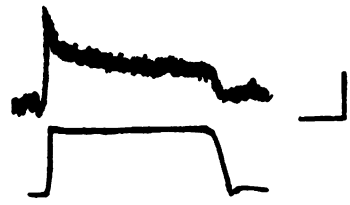

Fig. 3. Generator potential of decapsulated corpuscle in response to a mechanical stimulus of fast rate of rise (electrical damping). Superposed responses. Action potential blocked by tetradotoxin. Calibration: 10 msec; 50 μV.

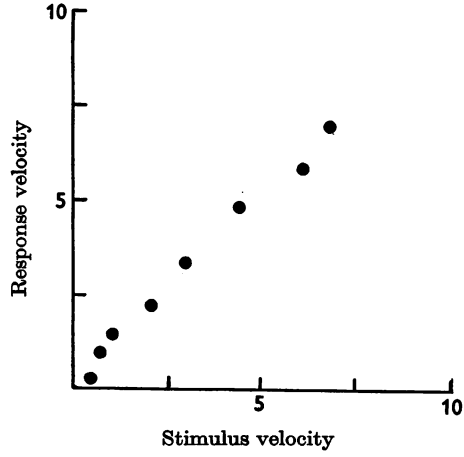

Fig. 4. Relation between velocity of generator potential and stimulus velocity. Mechanical pulses rising linearly to a constant plateau (total pulse duration 30 msec) are applied to a decapsulated corpuscle. Abscissae: rate of rise of mechanical pulse (relative units); ordinates: maximal rate of rise of resulting generator potential (relative units).

'On' and 'off'-responses. Both compression and decompression, if sufficiently fast, elicit a generator potential in the intact corpuscle. The characteristic pattern in the intact corpuscle is thus an 'on' and an 'off'-response to a mechanical pulse which are similar in time course and of like sign (Álvarez-Buylla & de Arellano, 1953; Gray & Sato, 1953). (Under the present stimulating conditions, the off-generator potential was, in fact, generally somewhat greater than the on-generator potential.) The decapsulated capsule is a better discriminator of direction of the stimulus force. Upon removal of capsule lamellae, the 'off'-responsiveness diminishes and is completely absent when only a few lamellae are left covering the sensory nerve ending (Fig. 2). Absence of 'off'-responses was the constant feature of decapsulated receptors, regardless of whether the 'on'-

phase of the stimulus outlasted the response, and regardless of the stimulus velocity (maximal velocity used on the order of 0·1 m/sec).

Tests. Tests were made to ascertain that the responses recorded after decapsulation were, in fact, generator potentials and not electrical artifacts. Artifacts, such as microphonic potentials, could arise from changes in resistance in nerve and other tissues due to mechanical deformation. A possible current source for microphonic potentials was the grid current of the amplifier. We have tried to produce such artifacts experimentally, but to be detectable they required grid currents several orders of magnitude greater than those of our amplifiers. Furthermore, unlike the generator response, the microphonics reversed sign with change in the direction of the mechanical force. Another possible current source was injury current. This was a more serious possibility of error, since appreciable microphonics can be elicited with strong stimuli from injured tissue. In fact, in a few cases in which decapsulation was carried on within the core in the immediate vicinity of the nerve ending, artifacts of this sort were observed which may have been due to injury of the nerve ending. But, again, these artifacts reversed sign with the direction of the stimulus and were, therefore, easily distinguishable from generator potentials. Such artifacts were, however, sufficiently large to interfere with analysis. We used, for this reason, preparations decapsulated down only to the outer core lamellae.

Tests for sign reversal of the response were made in all experiments. Besides, at the end of some experiments, the sensory axon was crushed at its junction with the outermost lamellae so as to eliminate the axon as a core conductor; or the nerve ending itself was crushed. This abolished all true receptor potentials.

Generator characteristics of the receptor response

In its special features, as described in the preceding section, the response of the decapsulated corpuscle to a sustained stimulus resembles that of the more slowly adapting type of mechano-receptors (see, for examples, Katz, 1950; Eyzaguirre & Kuffler, 1955; Wolbarsht, 1960; Thurm, 1964). This change in response character is merely what one would expect if the only essential difference in generator potential responsiveness in fast and slowly adapting receptors resides at the level of the mechanical coupling between external stimulus and sensor element, as supposed by the mechanical hypothesis. In its general features, the response has all the salient points of a sensory generator potential: the energy requirements for its production are low; it is continuously variable with the intensity of the stimulus; it reaches a maximum with strong stimuli; and nerve impulses are elicited when it attains a critical magnitude (Figs. 5 and 9*a*). Moreover, the response retains also the property of a depressive state following excitation,

as presented by the intact corpuscle: when the decapsulated preparation is driven with two successive stimuli, the second response is, within certain limits, depressed in amplitude, and, as in the intact corpuscle (Álvarez-Buylla & de Arellano, 1953; Gray & Sato, 1953; Loewenstein & Altamirano-Orrego, 1958), this depression increases with the amplitude of the first response and decreases with increasing response interval (Loewenstein & Rathkamp, 1958). For all these reasons it seems safe to regard the response of the decapsulated preparation as a generator potential. A further argument to this effect is given by the experiment described in the next section (Fig. 8).

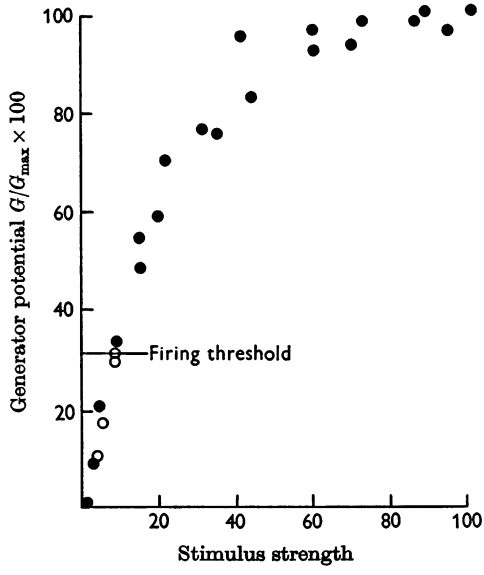

Fig. 5. Peak amplitude of prolonged generator potential of the kind illustrated in Fig. 2 as a function of stimulus strength (viscous damping). Peak amplitude of generator potential (G) is given as percentage of maximal generator potential (G_{max}). Decapsulated preparation. Durations of generator potentials and stimuli are approximately 27 msec. Open circles, responses before and filled circles, after blocking of nerve impulse production with tetradotoxin. Horizontal bar on ordinate marks threshold for impulse firing before application of tetradotoxin which in this preparation was conveniently high. In most other preparations, threshold was lower, but generator potential–stimulus strength curves were in all respects similar.

There are some quantitative differences. To produce a generator potential of a given magnitude, generally a greater stimulus strength is required in the decapsulated corpuscle than in the intact one. But this difference is minimized if large areas of the sensory ending are stimulated; in any event, the order of magnitude of the sensitivity remains unchanged

after decapsulation. Another difference concerns the depression following a generator potential. When short stimuli are used, so as to produce responses of durations comparable to those of an intact corpuscle, the depression is similar in duration and intensity to that of the intact corpuscle (see Fig. 10 by Loewenstein & Rathkamp, 1958). With responses of longer duration, the depression lasts longer (Figs. 6 and 7). In some cases responsiveness was still subnormal 15 msec after the end of the potential, i.e. 15 msec after the release of the conditioning stimulus.

This type of depression of the generator process appears to take place after release of the stimulus only, and not during the actual phase of excitation. It is difficult to make precise tests of responsiveness during excitation, i.e. in the course of a prolonged generator potential, because of non-linearity of the generator potential–stimulus strength relation. However, from rough tests done in the quasi-linear range of the relation, it is clear that depression during excitation, if present at all, must be small by comparison with that developing upon stimulus release. The generator process seems thus to lack the kind of refractoriness characteristic of processes of impulse production. This brings the Pacinian corpuscle in line with the behaviour of synaptic generator processes (see, for reviews, del Castillo & Katz, 1956; Grundfest, 1957, 1964; Eccles, 1957).

In the intact corpuscle, depression occurs with so short a delay from stimulus application that it gives the impression of a refractory state. This depression may now be interpreted in the light of the present results as a release phenomenon and the shortness of its delay explained by the shortness of the 'active' phase of excitation: owing to mechanical filter action of the capsule in the intact corpuscle, stimulus release at the level of the nerve ending occurs within a millisecond of stimulus application, regardless of stimulus duration at the periphery of the capsule (see Discussion), and depression sets in before the externally recorded generator potential has dissipated itself. On this basis, the depression in the Pacinian corpuscle may be compared with the depression that follows stimulus release in a wide variety of mechano-sensitive receptors, such as muscle spindles (Matthews, 1931; Katz, 1950), semicircular canal organs (Löwenstein & Sand, 1940), stretch receptor nerve cells (Eyzaguirre & Kuffler, 1955; Florey, 1956), and skin receptors (Loewenstein, 1956; see also Hensel & Zotterman, 1951; Landgren, 1952; Wiersma, Furshpan & Florey, 1953, for a comparison with other receptors).

Artificial 'capsule'

In the following experiments an attempt is made at changing the prolonged response of the decapsulated preparation to a response of the fast pulse-like kind by surrounding the preparation with an artificial capsule.

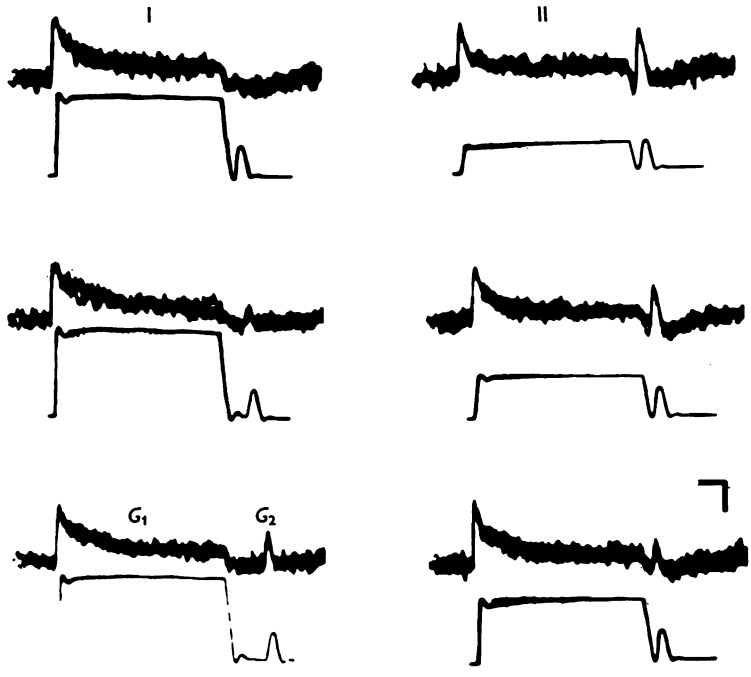

Fig. 6. Depression of receptor responsiveness following a prolonged generator potential. Two successive stimuli are applied to a decapsulated preparation, one to produce a long conditioning generator potential G_1, and the other a brief test generator potential G_2 (electrical damping). Column I, depression at constant conditioning stimulus strength and varying stimulus interval. Column II, depression at constant interval and varying conditioning stimulus strength. Impulse production blocked by pressure. Calibration: 5 msec; 50 μV.

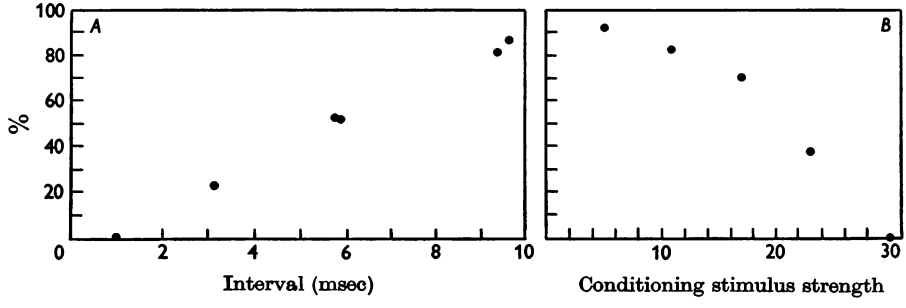

Fig. 7. Depression following a prolonged generator potential. *A*, Depression as function of stimulus interval (measured from end of conditioning mechanical stimulus to start of test stimulus). *B*, Depression as function of conditioning stimulus strength. Stimulus interval 32 msec. Ordinates for *A* and *B*: peak amplitude of test generator potential as percentage of unconditioned generator potential.

We experimented with a number of different materials and procedures to make such a capsule. The most satisfactory results were obtained with mesothelia from the mesentery of frogs (*Xenopus laevis*). These offer fine layers whose thickness is of the same order as that of Pacinian corpuscle lamellae. Single mesothelial layers were dissected and, after soaking in oil, covered with Krebs's solution. Ten to thirty such layers were then stacked

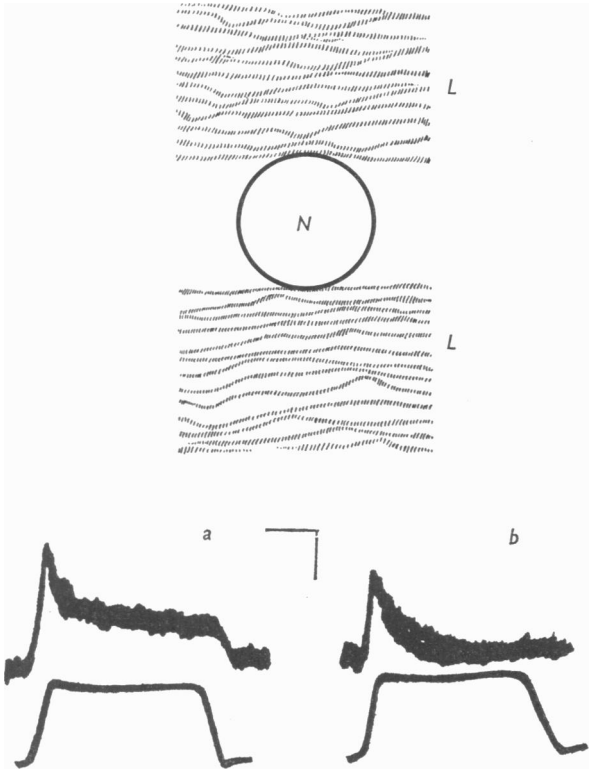

Fig. 8. *Top.* Diagram of artificial capsule. *N*, nerve ending and its core; *L*, artificial lamellae made of mesenteric mesothelium, separated by fluid-filled spaces over most of their surface and in close contact at a few spots. *Bottom.* Generator potential of decapsulated preparation in response to a stimulus of long duration. *a*, applied directly to the core; *b*, applied through the multi-layered artificial capsule. Several responses superposed. Impulse production blocked by pressure. Calibration: 5 msec; 50 μV.

one on top of the other and the decapsulated preparation placed in the centre of the stack (Fig. 8, top). Under the microscope, the layers were seen in close contact in some regions and in others separated by a film of Krebs's solution. This multi-layered system was placed between the plates of our crystal stimulator.

Figure 8 illustrates the results of an experiment in which the receptor is

stimulated with long pulses of compression. After the receptor is enclosed in the artificial 'capsule', the response becomes markedly shorter, although not quite as short as in the receptor surrounded by its natural capsule.

We varied several factors, such as number of lamellae, lamella spacing and lamella thickness, which all may be expected to influence the filter action of the capsule. To obtain good mechanical filtering, i.e. adequate shortening of the response, the following conditions had to be met: (i) the system had to be multi-layered; (ii) the layers had to be thin and elastic; and (iii) there had to be fluid between the layers.

Electrical component of adaptation

Adaptation of impulse production to generator currents. As discussed further on, the capsule seems to shorten the 'active' phase of a steady stimulus so severely that the generator response of the intact corpuscle lasts only a few milliseconds, most of which correspond almost certainly to passive decay of the potential over the membrane resistance and capacitance of the nerve ending and adjacent portions of myelinated axon. The generator potential decays rapidly and for 2 or 3 msec, at most, is it sufficient to trigger nerve impulses. This alone would seem sufficient to limit the number of impulses discharged by the receptor to one or two impulses, and to account for adaptation on the basis of mechanical filter action alone. But is this the only factor in adaptation?

Generator potentials can be conveniently prolonged by stimulating the decapsulated corpuscle with single mechanical pulses of long duration, as in the preceding experiments, or by stimulating the intact corpuscle with mechanical pulses of high frequency, as shown previously (Loewenstein, 1958). By these procedures, generator currents can be produced which remain above threshold for impulse firing (single) over durations many times longer than a refractory period of the impulse. None the less, the impulse discharge is as brief as that following generator currents of short duration. With the stimulus rates of rise used, the response consists typically of one action potential, in spite of the generator current being several times threshold for single impulse production (Fig. 9). The number of nerve impulses discharged in response to a steady stimulus by the receptor seems thus to be limited also at the level of the nerve impulse. The situation is similar in this respect to that at the fast-adapting crustacean stretch receptor in which impulse adaptation cannot be accounted for by decay in generator potential alone (Krnjević & van Gelder, 1961; Wendler & Burkhardt, 1961), and in Limulus photoreceptors in which there is adaptation of impulse production to externally applied steady currents, as well as generator potential adaptation (Hartline, Coulter & Wagner, 1952; MacNichol, 1956; Fuortes & Poggio, 1962).

Adaptation of impulse production to externally applied currents. In the following experiments, we have by-passed the mechano-electric generator and stimulated the sites of impulse initiation directly with an electric current. In these experiments, the corpuscle was decapsulated until the first node of Ranvier became exposed. The preparation was placed across two paraffin bridges, so that the first node and a central portion of non-myelinated ending (compartment of E_2, Fig. 10 top) were electrically insulated from the terminal portion of the ending (compartment of E_1). Square pulses of current were passed between electrodes E_1 and E_2 to stimulate the normal site of impulse origin, and action potentials were led off between E_2 and a central region of the sensory axon (E_3).

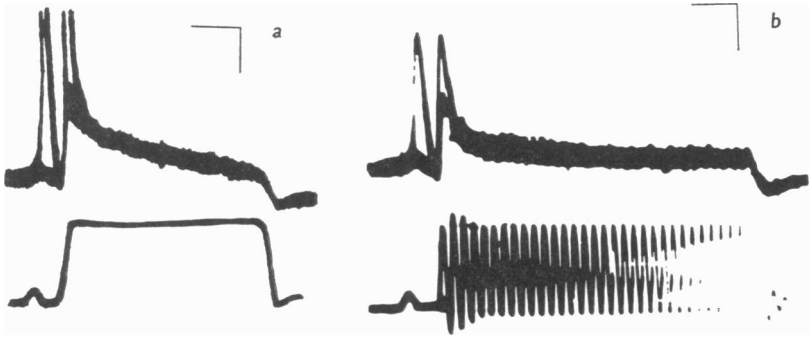

Fig. 9. Adaptation of impulse production to generator currents. *a*, adaptation to prolonged generator potentials produced by stimulation of a decapsulated corpuscle with a long mechanical test stimulus. *b*, adaptation to prolonged generator potentials produced by repetitive stimulation of an intact corpuscle. The test stimulus in *a* and *b* is preceded by an auxiliary stimulus of short duration critically at threshold for production of action potentials. The auxiliary pulse serves to raise the threshold of the test responses, which start during the relative refractory period of the action potential, so as to display the test generator response. The test generator response is just threshold for impulse firing at its beginning, but, thereafter, for at least 30 msec in *a* and throughout the entire period of stimulation in *b*, the generator response is of an amplitude greater than the minimum required for triggering of impulses at the beginning of a single stimulus. This minimum is displayed by the auxiliary response. Calibration for *a* and *b*: 5 msec; 50 μV.

A typical result is illustrated in Fig. 10. Only one impulse is discharged in response to a suprathreshold current lasting 50 msec. Out of six preparations tested, three gave one impulse, two gave two impulses and one gave three impulses to currents up to 12 times threshold strength regardless of their duration.

It is thus clear that a second factor enters here in receptor adaptation. This factor resides in the process of impulse production and, as in squid giant axon (Hodgkin & Huxley, 1952), could possibly be sodium inactivation caused by constant depolarization.

The fast accommodating character of the sensory axon of the Pacinian corpuscles was already known from Gray & Matthews' work (1951) (see also Gray & Malcolm, 1950). Gray & Matthews found that only a few impulses could be elicited from extracorpuscular regions of the axon by electrical stimulation. Our experiments differ from theirs in that we are stimulating the region of the impulse initiation inside the corpuscle, instead of more central regions of axon. In any event there appear to be no basic differences in accommodation between the two regions. It is interesting in this connexion that a recent study on accommodation of the site of impulse

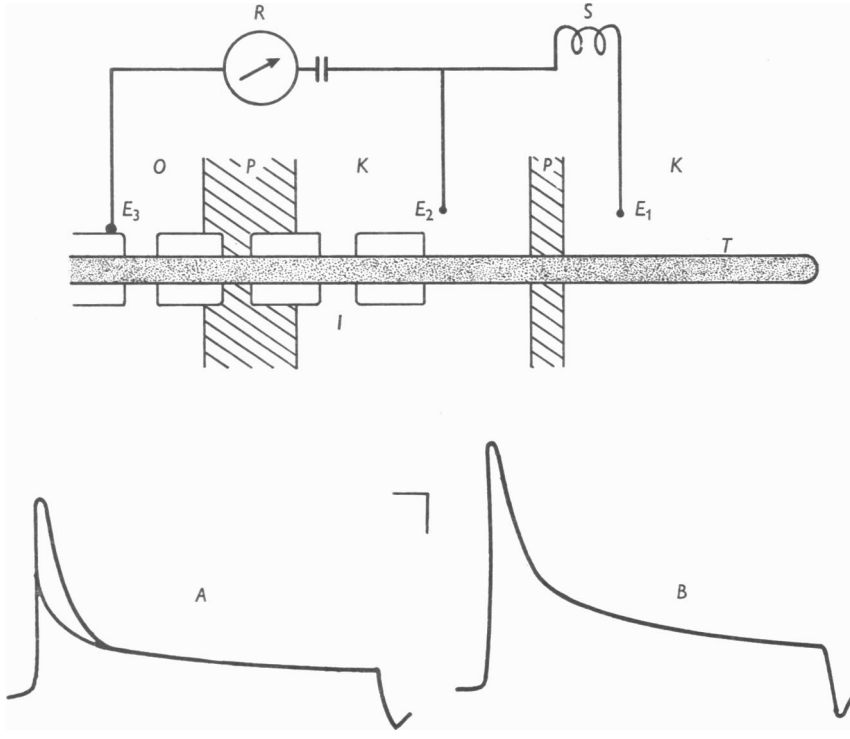

Fig. 10. Accommodation of impulse production to constant currents. *Top.* Diagram of electrode arrangement. *T*, nerve ending and its core; *P*, paraffin insulation; *K*, pools of Krebs's solution; *O*, mineral oil; *I*, first node (intracorpuscular) of Ranvier; E_1, stimulating electrode connected through an isolation unit (*S*) to a square-pulse generator; E_3, recording electrode in direct contact with axon connected through a cathode follower to a capacity-coupled amplifier of an oscilloscope. E_2, electrode in common with stimulating and recording circuits. *Bottom.* Tracings of electrical response to square pulses of current ($1{\cdot}5 \times 10^{-8}$ A) flowing outward through membrane regions located in the pool of E_2. *A*, current of just threshold strength for impulse firing; *B*, about two times threshold. Lower trace in *A* is the resistive and capacitative coupled potential spread; its beginning and end indicate the 'on' and 'off' of currents. Calibration: 5 msec; 50 μV.

initiation in the fast-adapting crustacean stretch receptor revealed a similar factor in receptor adaptation (S. Nakajima, personal communication).

DISCUSSION

The filter action of the capsule. The following steps can be distinguished in the chain of conversion of mechanical into electrical energy at mechanoreceptors: external mechanical stimulus (1) → internal mechanical transmission of stimulus energy (2) → generator current (3) → nerve impulse (4). In the Pacinian corpuscle, the structural elements that can be identified with steps 2 and 3 are the capsule and the non-myelinated sensory ending, respectively. The present results show that when step 2 is by-passed significantly by elimination of the capsule, the sensory ending produces generator currents which follow the stimulus in time course with relatively little distortion. The sensory ending appears to be capable of converting mechanical energy rather faithfully. It is the capsule that introduces the main distortion in stimulus energy on its way to the ending. The capsule appears to be a good transmitter of fast mechanical transients, but a poor transmitter of slow ones. In fact, slow velocities do not seem to propagate to any significant extent to the centre of the capsule where the ending is located.

What are the mechanics of this filter action? A hydro-mechanical model of a capsule has been developed by Skalak & Loewenstein (1965). The mechanics of this model fit the present results of decapsulation rather well; they also fit closely Hubbard's measurements (1958) of lamella displacements. A detailed quantitative analysis of the mechanics of the model is given by Skalak & Loewenstein (1965); here only the aspects immediately relevant to generator potential adaptation will be considered qualitatively. The model is essentially a multi-layered squeeze-bulb. Its principal elements are concentric lamellae similar in number, thickness and arrangement to those of the Pacinian corpuscle. It incorporates the elastic character of the lamellae and the viscous character of the inter-lamellar fluid. Adjacent lamellae are loosely coupled. The system behaves essentially like a dashpot with pistons (the lamellae) in series. To mechanical stimuli of slow rates of rise, that is, compressions of the outer surface, such a system offers relatively little viscous resistance. Thus, little viscous force is developed in it. Elastic force is virtually the only force produced, and this is small and falls steeply from periphery to centre. With fast rising stimuli, on the other hand, viscous resistance is high. Hence, a high viscous force is developed in addition to the elastic one. The viscous force is transmitted with comparatively little spatial decrement through the system. In contrast to a slowly rising stimulus, a fast rising stimulus, therefore, sets up

a pressure field which is of significantly high amplitude at the centre of the system (Skalak & Loewenstein, 1965). A mechanical stimulus with an initially high velocity, slowing down to zero, such as used in the present experiments (see, for example, Fig. 2) will thus give rise in the intact corpuscle to a brief pulse of pressure at the centre where the sensor is located, lasting only as long as the fast rising phase of the stimulus. During the remainder of the stimulus duration and particularly during the static phase, no significant pressure exists at the centre, because (i) the pressure, then only chiefly of elastic origin, is low to start with and (ii) falls off steeply from periphery to centre.

Some of these features, those concerning the velocity dependence of pressure, are already intuitively seen (the familiar behaviour of a hydraulic door damper pushed at high and low speeds provides an analogy); the spatial distribution of pressure and the quantitative aspects are demonstrated analytically (Skalak & Loewenstein, 1965). Analysis shows, in particular, that pressure at the centre of the capsule system has, in fact, durations of the same order as that of the generator potential in the intact corpuscle. *The rate-limiting factor in the decay of the generator potential in response to a sustained stimulus, that is, the rate-limiting factor in generator potential adaptation, appears thus to be the capsule operating as a mechanical filter.*

The primary elements of this mechanical filter are the lamellae, their inter-connexions and the fluid. The former two provide the structural stiffness and the elasticity, and the latter, the viscosity. The system has thus the elements of capacitative reactance (elasticity) and resistance (viscosity). The relation between the energy of the external stimulus and that transmitted through the capsule presents the following characteristics when a step compression is applied at the surface: the initial force transmitted increases with the velocity of the stimulus as determined by the resistive element. An additional force is developed at later times, as determined by the reactive elements. But this is small by comparison and may be neglected at first.

It is important to note that the filter characteristics of the lamellar system, and hence the mechanics of receptor adaptation, are strongly dependent on the viscous element and cannot be explained by a consideration of lamella displacements (as those given, for example, in Hubbard's, 1958, excellent data) and elastic forces alone.

'On' and 'off'-response. During the 'off'-phase of the stimulus, stimulus energy stored in the elastic elements of the system is released. If this energy is sufficiently large and if the release is fast, viscous force is again developed. The sensory ending at the centre receives then during the 'off'-phase a pressure pulse similar to that of the 'on'-phase. The lamella dis-

placements and the corresponding force vectors are now rotated by 90°; what was a direction of maximal compression during the 'on'-phase becomes a direction of maximal decompression in the 'off'-phase, and vice versa. The quantitative analysis of the dynamics of the 'off'-phase shows that the pressures developed at the centre of the capsule are, in fact, of the same order as those during the 'on'-phase for similar stimulus velocities (Skalak & Loewenstein, 1965). This provides a simple explanation for the production of the 'off'-response in the intact corpuscle, and for its absence in the more rigid decapsulated core preparation.

On the physiological role of the capsule. It seems worth while at this point to consider some of the possible functions of the capsule of the Pacinian corpuscle. The capsule is clearly directly involved not in the generator process (Loewenstein & Rathkamp, 1958), but only in the transmission of mechanical energy to the receptor proper. One possibility is that the capsule serves to protect the nerve ending from excessive stress. This may be so perhaps in corpuscles located in joints or on bones, but it can hardly be of importance in corpuscles in skin, muscle and viscera, where there are naked nerve endings of many kinds under similar stress.

Another possibility is that the capsule serves as a distributor of force of the external stimulus, as suggested by the finding that the generator current is built up by spatial summation of small local currents at the ending (Loewenstein, 1959, 1961). This is an interesting possibility. But a judgement of its validity depends on the physiological role that is to be ascribed to the Pacinian corpuscle. If, for example, the sensory ending were a detector for generalized vibration (cf. Hunt, 1961), it seems likely that the soft and relatively thick tissues in which corpuscles are generally located, would themselves produce significant dispersion of the stimulus force. On the other hand, the sensory ending functioning as a detector for local vibrations, as for instance a naked ending in partial contact with the wall of a pulsating blood-vessel, may require a larger effective stimulus than an encapsulated one.

Another possibility is a role of the capsule in isolating chemically the sensory ending. The capsule is, indeed, a formidable barrier for diffusion (Gray & Sato, 1955). But even one lamella is a strong diffusion barrier, and one cannot help feeling that the complex arrangement of hundreds of such layers is superfluous, when in sensory endings of many sorts one layer suffices.

Another possible role is that of changing the direction of the stimulus force, i.e. of changing compression perpendicular to the length axis of the corpuscle into a stretch along the axis. Such a possibility was already considered by Adrian & Umrath (1929) (see also Gray & Ritchie, 1954; and Granit, 1955) at the time of the first electrophysiological study of the

Pacinian corpuscle and would undoubtedly be of importance if changes in length, that is, longitudinal strain, were the immediate causes of the generator process. However, since viscous force in the capsule is produced predominantly in the direction of compression, it is not likely that changes in length are important. So far no changes in length of the ending have been resolved, even with strong stimuli (Hubbard, 1958).

From the present results, a role in receptor adaptation of the capsule emerges clearly as a possibility. Yet it is also clear that there is a second adaptive mechanism at the level of impulse initiation which alone would be sufficient to confer on the receptor its fast-adapting character. Finally, another possibility that comes forth from the present work is a role in the receptor's 'off'-responsiveness. Indeed, one of the striking changes following decapsulation is the disappearance of 'off'-responses. However, it should be kept in mind that this ensues under conditions in which stimulus after-oscillations have been eliminated. In the receptor organ *in situ*, the mechanical stimulus field following the natural stimulus may be oscillatory and, hence, 'off'-responsiveness be inherent in the system quite apart from that provided by the capsule. An evaluation of the physiological significance of the capsule in 'off'-responsiveness must thus await information on the mechanical damping provided by the tissues surrounding the receptor.

SUMMARY

1. Mechanical stimuli, namely, pulses of compression, were applied rather directly on to the nerve ending of a Pacinian corpuscle, from which most of the lamellated capsule had been removed by dissection. The capsule was thereby effectively by-passed as a coupling in the transmission of mechanical energy to the sensory ending, and transducer properties of this ending could be examined relatively undistorted by non-nervous mechanical factors. Under these conditions, the electrical response of the ending behaves as that of more slowly adapting sensory endings:

(*a*) The duration of the generator potential is continuously variable with stimulus duration. The potential in response to a sustained mechanical stimulus eventually decays. But, while the response of the encapsulated ending decays to zero within about 6 msec (Gray & Sato, 1953), that of the decapsulated ending may last more than 70 msec and, over considerable part of this time, the response follows roughly the time course of the stimulus.

(*b*) Generator potentials are produced in response to compression (on), but not to decompression (off). By contrast, the encapsulated ending does not discriminate direction of stimulus force and, typically, responds to both the on- and off-phase of the stimulus.

(*c*) The off-phase of the stimulus is followed by a period of depression of generator responsiveness.

2. When the decapsulated ending is enclosed in an artificial capsule made of several layers of tissue separated by fluid, the generator potential in response to a sustained stimulus applied to the surface of this capsule acquires a short pulse-like character resembling that of the ending in its natural capsule. This mechanical filter effect requires (i) that the structural elements of the artificial capsule be multi-layered, (ii) that the layers be thin and elastic, and (iii) that there be fluid in between them. The tissue layers of the artificial and natural capsules appear to behave like in-series pistons moving in a system with viscosity (as given by the fluid) and elasticity (as given by the tissue layers and their interconnexions) such that only viscous force, but no significant elastic force, is transmitted from the periphery to the centre of the system. Thus mechanical transmission in the system is highly velocity-dependent; stimuli of slow velocities applied to the capsule surface produce no significant force at the centre where the sensory ending is located.

3. The number of nerve impulses is independent of the duration of the generator potential. Typically, only one impulse is produced regardless of whether the generator potential is brief, as in the encapsulated ending, or prolonged to maximum in the decapsulated ending. When the generator mechanisms are by-passed, and electrical currents from an external source are applied to the terminal region of the sensory axon, the number of resulting impulses is also quite independent of current duration.

4. There appear thus to be, at least, two components in adaptation in this receptor. One, a mechanical component, operates as a mechanical filter in the form of a laminated capsule preventing slow stimulus components from reaching the sensory ending; and, another, an electrochemical component, operates at the level of nerve impulse production preventing a steady outward current from producing repetitive impulses.

REFERENCES

ADRIAN, E. D. & UMRATH, K. (1929). The impulse discharge from the Pacinian corpuscle. *J. Physiol.* **68**, 139–154.

ÁLVAREZ-BUYLLA, R. & DE ARELLANO, J. R. (1953). Local responses in Pacinian corpuscles. *Amer. J. Physiol.* **172**, 237–250.

ÁLVAREZ-BUYLLA, R. & REMOLINA, J. (1959). The initiation of action potentials at Pacinian corpuscles. *Acta physiol. latinoamer.* **9**, 178–187.

CASTILLO, J. DEL & KATZ, B. (1956). Biophysical aspects of neuromuscular transmission. In *Progress in Biophysics*, vol. 6, ed. BUTLER, J. A. V. London: Pergamon Press.

ECCLES, J. C. (1957). *The Physiology of Nerve Cells.* Baltimore, Md.: Johns Hopkins.

EYZAGUIRRE, C. & KUFFLER, S. W. (1955). Processes of excitation in the dendrites and in the soma of single isolated sensory nerve cells of the lobster and crayfish. *J. gen. Physiol.* **39**, 87–119.

FLOREY, E. (1956). Adaptionserscheinungen in den sensiblen Neuronen des Streckrezeptors des Flusskrebses. *Z. Naturf.* **11***b*, 504–13.

FUORTES, M. G. F. & POGGIO, G. F. (1962). Transient responses to sudden illumination in the cells of the eye of Limulus. *J. gen. Physiol.* **46**, 435–452.

GRANIT, R. (1955). *Receptors and Sensory Perception.* New Haven, Conn.: Yale University.

GRAY, J. A. B. & MALCOLM, J. L. (1950). The initiation of nerve impulses by mesenteric Pacinian corpuscles. *Proc. Roy. Soc.* B, **137**, 96.

GRAY, J. A. B. & MATTHEWS, P. B. C. (1951). A comparison of the adaptation of the Pacinian corpuscle with the accommodation of its own axon. *J. Physiol.* **114**, 454–464.

GRAY, J. A. B. & RITCHIE, J. M. (1954). Effects of stretch on single myelinated nerve fibres. *J. Physiol.* **124**, 84–99.

GRAY, J. A. B. & SATO, M. (1953). Properties of the receptor potential in Pacinian corpuscles. *J. Physiol.* **122**, 610–636.

GRAY, J. A. B. & SATO, M. (1955). The movement of sodium and other ions in Pacinian corpuscles. *J. Physiol.* **129**, 594–607.

GRUNDFEST, H. (1957). Electrical inexcitability of synapses and some consequences in the central nervous system. *Physiol. Rev.* **37**, 337–361.

GRUNDFEST, H. (1964). Evolution of electrophysiological varieties among sensory receptor systems. In *Turpeyev: Essays on Physiological Evolution.* V.E.P. Leipzig: Leipziger Druckhaus.

HARTLINE, H. K., COULTER, JR., N. A. & WAGNER, H. G. (1952). Effects of electric current on responses of single photoreceptor units in the eye of Limulus. *Fed. Proc.* **11**, 65–66.

HENSEL, H. & ZOTTERMAN, Y. (1951). Quantitative Beziehungen zwischen der Entladung einzelner Kältefasern und der Temperatur. *Acta physiol. scand.* **23**, 291–319.

HODGKIN, A. L. & HUXLEY, A. F. (1952). The dual effect of membrane potential on sodium conductance in the giant axon of Loligo. *J. Physiol.* **116**, 497–506.

HUBBARD, S. J. (1958). A study of rapid mechanical events in a mechanoreceptor. *J. Physiol.* **141**, 198–218.

HUNT, G. C. (1961). On the nature of vibration receptors in the hind limb of the cat. *J. Physiol.* **155**, 175–186.

KATZ, B. (1950). Depolarization of sensory terminals and the initiation of impulses in the muscle spindle. *J. Physiol.* **111**, 261–282.

KRNJEVIĆ, K. & VAN GELDER, N. M. (1961). Tension changes in crayfish stretch receptors. *J. Physiol.* **159**, 310–325.

LANDGREN, S. (1952). On the excitation mechanism of the carotid baroceptors. *Acta physiol. scand.* **26**, 1–34.

LIPPOLD, O. C. J., NICHOLLS, J. G. & REDFEARN, J. W. T. (1960). Electrical and mechanical factors in the adaptation of a mammalian muscle spindle. *J. Physiol.* **153**, 209–217.

LOEWENSTEIN, W. R. (1956). Excitation and changes in adaptation by stretch of mechanoreceptors. *J. Physiol.* **133**, 588–602.

LOEWENSTEIN, W. R. (1958). Generator processes of repetitive activity in a Pacinian corpuscle. *J. gen. Physiol.* **41**, 825–845.

LOEWENSTEIN, W. R. (1959). The generation of electric activity in a nerve ending. *Ann. N.Y. Acad. Sci.* **81**, 367–387.

LOEWENSTEIN, W. R. (1961). Excitation and inactivation in a receptor membrane. *Ann. N.Y. Acad. Sci.* **94**, 510–534.

LOEWENSTEIN, W. R. & ALTAMIRANO-ORREGO, R. (1958). The refractory state of the generator and propagated potentials in a Pacinian corpuscle. *J. gen. Physiol.* **41**, 805–824.

LOEWENSTEIN, W. R. & RATHKAMP, R. (1958). The sites for mechano-electric conversion in a Pacinian corpuscle. *J. gen. Physiol.* **41**, 1245–1265.

LOEWENSTEIN, W. R., TERZUOLO, C. A. & WASHIZU, Y. (1963). Separation of transducer and impulse-generating processes in sensory receptors. *Science*, **142**, 1180–1181.

LÖWENSTEIN, O. & SAND, A. (1940). The individual and integrated activity of the semicircular canals of the elasmobranch labyrinth. *J. Physiol.* **99**, 89–101.

MACNICHOL, JR., E. F. (1956). Visual receptors as biological transducers. In *Molecular Structure and Functional Activity of Nerve Cells*, ed. GRENELL, R. G. & MULLINS, L. J. Washington, D.C.: Amer. Inst. Biol. Sci.

MATTHEWS, B. H. C. (1931). The response of a muscle spindle during active contraction of a muscle. *J. Physiol.* **72**, 153–162.

MENDELSON, M. & LOEWENSTEIN, W. R. (1964). Mechanisms of receptor adaptation. *Science*, **144**, 554–555.

PEASE, D. C. & QUILLIAM, T. A. (1957). Electron microscopy of the Pacinian corpuscle. *J. biophys. biochem. Cytol.* **3**, 331–342.

SKALAK, R. & LOEWENSTEIN, W. R. (1965). Mechanical transmisson in a Pacinian corpuscle. An analysis and a theory. (To be published.)

THURM, U. (1964). Das Rezeptorpotential einzelner mechanorezeptorischer Zellen von Bienen. *Z. vergl. Physiol.* **48**, 31–156.

WENDLER, L. & BURKHARDT, D. (1961). Zeitlich abklingende Vorgänge in der Wirkungskette zwischen Reiz und Erregung (Versuche an abdominalen Streckrezeptoren dekapoder Krebse). *Z. Naturf.* **16***b*, 464–469.

WIERSMA, C. A. G., FURSHPAN, E. J. & FLOREY, E. (1953). Physiological and pharmacological observations on muscle receptor organs of crayfish, *Cambarus clarkii* Girard. *J. exp. Biol.* **30**, 136–150.

WOLBARSHT, M. L. (1960). Electrical characteristics of insect mechanoreceptors. *J. gen. Physiol.* **44**, 105–122.