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Speculations on Smell

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INTRODUCTION

This paper complements the detailed account of our experiments that will have appeared in *Journal of Physiology* by the time this volume is published (Gesteland, Lettvin, and Pitts, 1965). It presents a cloud of opinion, prejudice, and hunch that we try not to show when offering data for, our concrete results, however hard won, have not led us to any epistemological epiphany. Neither have they given us a molecular machine. When we had found that the responses of single nerve fibers in N_I were not simply related to the stimuli presented, we had two choices. First, we could have sought by other, yet uninvented methods, those more elementary events whose combinations yielded the functions we saw—i.e., we could have concentrated on the molecular basis for chemical transduction. Second, we could have accepted the combinational responses as given, and then made inductions on the nature of the olfactive code. Walter Pitts, with whom we began the experimental study, is more interested in the first problem, as are most scientists who think about smell. We, however, chose to study the second question in the hope of bringing off a coup like that involving the frog's eye. Well, we didn't for, astonishingly, the nose is more difficult to handle than the eye. In the end we developed some good methods for looking at olfactory tissue and getting data, but now we have no very clear ideas about how to handle the results. We felt that it would be most useful to our colleagues if we discussed matters informally.

PSYCHOLOGICAL QUESTIONS

How we smell is as hard to dissect as how we see. A scent may smell like lavender, a stink may smell like rotten eggs—but the figure of speech is as far as we can go. Just as a face is not perceived as a spatial array of light, so too a smell is not perceived as a mixture of components. Yet, as an artist can train himself to mark the chiaroscuro on a face, so a winetaster or perfumer can become expert in picking out the parts of a complex odor. Most odors are complex; we scent them as singular forms. There is the smell of home, the reek of fever, the stench of fear, the good odor in which saintly people die. All of us are expert on small differences in the exhalations of friends and family; given sufficient training, some of us can say if a vintage had seen the sea. But there is no way of telling these perceptions to others except by simile. Even when professional perfumers address each other, it is in a cant that evokes intuition rather than understanding. There are "floral top-notes" and "spicy overtones" modifying a "fragrance theme," and this is the language of journals, not of advertising men; and these are professionals trying to communicate, not art critics trying to confuse each other.

The classification of odors fares as badly as the description. Zwaardemaker's system has not been replaced with a more definite one, however much it is needed by industry if not science. Even the ambitious attempts of the Slawkenbergians at A. D. Little, who walk about with noses swaddled against the fuming turmoil of the world, to be unveiled, as with the flick of a yashmak, for the faintest flirt with an unknown wafted from the feeble wave of a stopper held at arm's length; even the soul searching of these introverted osphresiologists to whom the distant violet is as ashes and savors of the flesh, and who despise the chromatograph as a crude toy; even their practical programs, that have broken the awful complex of artificial root beer and brought chicken soup to its ultimate simplicity, have led to no coherent account of how to tell one smell from another.

If, instead of classifying differences we look to similarities, and search out metamers, or stimulus equivalents, we have many, but can make no sense out of them. Some war gases smell like new-mown hay, others like geraniums, etc.; the Air Force has an alarm compound that smells like dirty socks but could not be the active principle of them; certain arsenical compounds smell like garlic. The list is bizarre. It is all very well to maintain with Pauling that whatever looks like camphor (i.e., has the same molecular shape) ought to smell like camphor. But in the cases given above it is not that we are replacing one compound with another like it, but rather a complex of compounds with a single one. One might argue that in the case of the dirty socks there was a kind of butyrate that was mainly responsible for the smell, and it is to this that the alarm chemical was similar. But if one went about isolating chemicals from dirty socks, it would most often be the

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case that one or another seemed reminiscent of the full aroma, but none was really as good as that single compound that smells like the whole mixture got from dirty socks. This is the basis of the present traffic in cheats: compounds that smell like new-baked bread, others that smell like new cars, etc. Thus the problem of similarity is more difficult than we are led to believe by work that supports Pauling's comment, such as that excellent study of Amoore's that appears later in this volume. We must bear in mind that metamerism is possible almost in the same way as in color space. But smell space is really quite different from color space, for let us consider another instance: Suppose we have two flasks, one containing the essence of a shoe polish, and the other lavender water. When we mix air from the two flasks we smell a mixture of shoe polish and lavender, and say that it smells like a mixture. Often, by turning our minds to it, especially if asked beforehand, "Is there lavender in this air?" we can say "I think I can smell lavender in this mixture." As we alter the ratio of shoe polish to lavender we go from thinking that the shoe polish is perfumed by lavender to supposing that the lavender is corrupted by shoe polish. Such a mixture is quite different in one sense, from a mythical, but conceivable single compound that has the smell of lavender shoe polish. Professional noses are adept at telling the compounds in a mixture that imitates a pure compound. What we are trying to say is that the mixing of two compounds does not necessarily yield the smells of other compounds which smell as if they were intermediate between those two, although often enough great similarities will exist. This is the quality that is reflected in the language of perfumers who talk of overtones, undertones, dissonances, etc., as if a mixture partook more of the property of a chord in music than of addition of colors.

It is this analogy with sound that, unspoken, underlies the quest for chemically specific receptors just as one searches for frequency specific elements in the ear. This specificity may be intrinsic, as in the sex-attractant receptors of Schneider's bees. And we become heartened in this notion of specific receptors by the existence of those congenital partial anosmias now being collected by Amoore (1964). There are some people who cannot smell cyanides, others who cannot smell butyric acid or butyrates, others who cannot smell whatever the compound is that issues in the urine of one who has eaten asparagus. But these people seem to be able to distinguish other smells perfectly well, and their blindness is more similar to a notch defect in audition than to the absence of a pigment in vision.

There is another puzzling fact. It is possible to create organic compounds that probably never occurred in nature. These will have distinctive smells, different from all others one can remember. We should think it unlikely that there are definite receptors for them. But if they affect existing receptors, why should their smells be so distinctive? From this we would tend to suspect that if a receptive process could be called specific, it could only be so in the broadest sense, just as we speak of the "red" pigment that really has a wide action spectrum. Counter to this view, the congenital specific anosmias ought not be so clear if the various receptive processes were very broad-band and there were many different types (so that, e.g., some cyanides could affect the butyric acid receptive process, and vice versa).

The most mysterious quality of odors is that they frequently do not combine in an understandable way. The witchcraft of perfume making is a case in point. Musks do not, in pure essence, give the same impression of the quality they confer on musky perfumes. Monosodium glutamate does not smack of the quality it confers on a dish. Now we have other such salts, one of which, alone in solution, has little distinctive flavor, but makes any mess, even dishwater, taste like good chicken soup.

The definiteness of complex smells is also surprising. Ants of the same brood, separated into two colonies in the same area of land, adopt two different nest smells through their queens although their food has been, on the average, the same. An ant of one colony will be promptly destroyed if put in the nest of the other, unless he is washed carefully. Blinded frogs hop back to their home ponds when there are several ponds in the immediate neighborhood. Dogs, eels, termites and *fin de siècle* decadents like Huysmans, all attest to the highly individual properties of very complex smells, small variations in which cause large variations in quality. Witness the shipping of wines to sea for mellowing the bouquet, the precision in the compounding of perfumes, the secret of Coca-Cola. There is the Bruce and Parks effect on female mice, which if pregnant and exposed to a short sniff of a male of a different strain of the same species, promptly abort and become fertile again within two days. Finally, just as attack transients modify the quality of a maintained tone (that is, in part, how we tell a violin from a flute), so some olfactants modify others, and the result of the two given together is not decomposable perceptually. The slight acridity of a woman's sweat or other secretions added to perfume makes a far more exciting smell than the perfume alone—but it is almost impossible to detect the additives. All these phenomena suggest that one function of smell is not to tell pure chemicals apart but to distinguish clearly between very similar mixtures.

PHYSIOLOGICAL STUDIES

We had assumed at first that there might be many different kinds of olfactory receptors, and that all these combinatorial qualities would be conferred by the second-order and later olfactory systems in the brain. But the response of a single axon in the olfactory nerve is not odor-specific. That is, it does not discharge only when a particular chemical and its related compounds are wafted into the nose. While specific receptors of this sort exist elsewhere, as described in this volume in Schneider's account of sensors for sex attractants in insects, we have found no analogous elements in the frog's nose. Instead, almost every odor seems to affect almost every receptor one way or another. In this respect, the frog's elements are like Schneider's "generalized" receptors in moths (Schneider, Lacher, and Kaissling, 1964).

We measure the manner in which an olfactory receptor is affected by means of the chatter of impulses in the axon that issues from it. Such an axon is not silent when the olfactory mucosa is bathed in a stream of clean, moist air. Most axons have a low average rate of firing. This rate changes when odors are puffed into the stream of air. For any axon, some odors increase the chatter greatly and others decrease it greatly (Fig. 1). Most odors have a lesser effect in one direction or the other, and some have no effect. What changes from axon to axon is the ordering of odors from those that exalt it most, through those to which it is impassive, to those that depress it most. If we write the order by which a set of different pure compounds, given

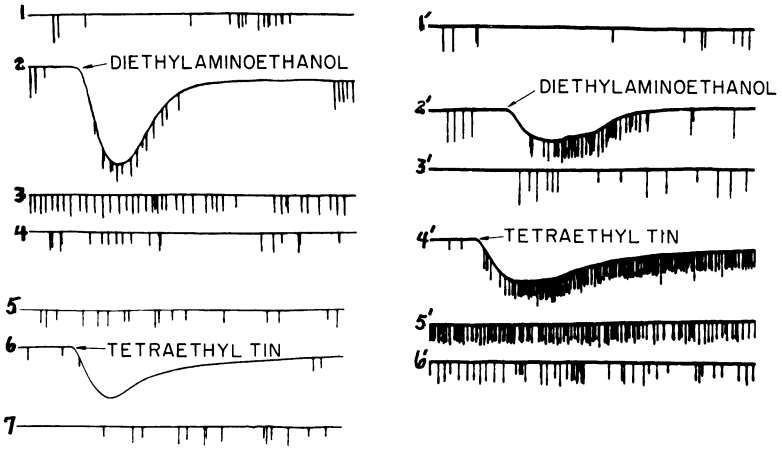

FIGURE 1. In each sweep we have combined the slow potential, recorded against an indifferent ground through a pipette filled with Ringer-gelatin just touching the mucus on the olfactory eminence, with the response of a few fibers of N_1 in a small bundle recorded by means of a Dowben-Rose electrode thrust into the basement membrane near the first electrode. The fiber activity is abstracted from the high noise level by means of a "window" filter gated by spike amplitude. This method is described in detail in J. Physiol. (Gesteland et al., 1965). The largest spikes are about 100–200 μv in amplitude, the slow potentials are, at maximum, about 2 mv. The two separate records, at their different gains, are added at the input to the oscilloscope. The sweep is 10 sec long.

Because the fiber activity is occurring on a very noisy baseline, the amplitude of the spikes due to a single unit fluctuates greatly. We have not troubled to separate the three or four units occurring in the recordings, since our arguments do not require a very good discrimination, but it is possible to do so using more complex techniques of data processing.

Records 1–7 show the responses at one position in the olfactory mucosa of a frog. Line 1 is a control sweep showing the low background activity of several units. Partway through 2 we delivered a short strong puff of diethylaminoethanol. At the very peak of the slow potential there is a moderate burst of activity and then, after an hiatus, at the end of the sweep, a moderately steady firing begins. This continues in sweep 3 taken a few sec later, and first begins to slow down in sweep 4 taken a few sec after 3. At another time we took sweep 5, showing a resting activity slightly larger than that of sweep 1. Ten sec after sweep 5 we began sweep 6, and partway through it delivered a short moderately strong puff of tetraethyl tin. There is an undeniable inhibition set up in all units by this puff. Sweep 7 was taken ten sec later.

Records 1′–6′ were taken at another point on the mucosa. Again, several units are being recorded. Sweep 1′ is representative of the resting activity. During sweep 2′ we gave a somewhat weaker and longer puff of diethylaminoethanol than for the first set of records. There is a definite excitatory response that does not involve, however, the largest spike seen at the beginning of sweep 2′. Ten sec later we took sweep 3′. Resting activity did not change much from this pattern thereafter. One min later we took sweep 4′ and during it gave a stimulus of the same strength of tetraethyl tin as we gave for the first series. The enormous and protracted response occurs primarily in a unit of intermediate height and clearly does not involve the large spike at all, for it can be seen to discharge three or four times during the sweep. 5′ was taken immediately after 4′ and shows the response of the intermediate-sized unit continuing. No large spikes appear during this sweep. Ten sec later we took 6′.

The important thing in this figure is that in the first electrode position, diethylaminoethanol excited some elements and tetraethyl tin inhibited all. In the second position, diethylaminoethanol excited some elements, but not all visible in the records, and tetraethyl tin excited one unit very violently while having no immediate effect on another unit except, possibly, a late inhibition.

separately, affect one axon, what we can say is that the probability that any two axons order the same set of compounds differently increases with the number of compounds in the set. Alternatively, when we choose two compounds that smell alike to us, or have the same form as in Amoore's hypothesis, or that belong to the same chemical group, e.g., aldehydes, then if we find both to have the same effect on one axon, a short search yields an axon that is affected differently by the two.

But it would seem unlikely that no two fibers, among the millions in the olfactory nerve, have the same ordering of odors. If we and the frogs have as related an olfactory sense as anatomy suggests, then our subjective feeling of similarities between different smells indicates that there are some general similarities in the affection of groups of the receptors by those smells. But our experiments are so slow-paced and the array of odors used in one experiment is so limited by time, that we have not been able to discover such groups. We are only sure that there are more than 10 kinds of ordering and trust that there are less than a million.

Because there are no synapses between receptor and axon, both being specialized parts of the same neuron, the rate of firing of an axon reflects how much current flows outward across the membrane where the impulses originate. It does not matter whether that place is cell body, axon hillock or olfactory rod. However long the chain of events that leads from absorption of a molecule to the representation by nervous signal, there is that penultimate link, the governing of current flow through an "electrically excitable" patch of membrane, whereby the rate of firing is governed. When a single fiber is predominantly exalted by one odor and predominantly depressed by another, we must infer that a single odor can either enhance outward current through that "electrically excitable" patch or suppress it even to the point of reversing its direction. These are two different effects. Because with a short search we can always find, for any odor, at least one axon that is depressed by its action and one that is exalted, the mixtures of these effects must be widespread across the mucosa. Thus we wanted to see if we could find signs of two different

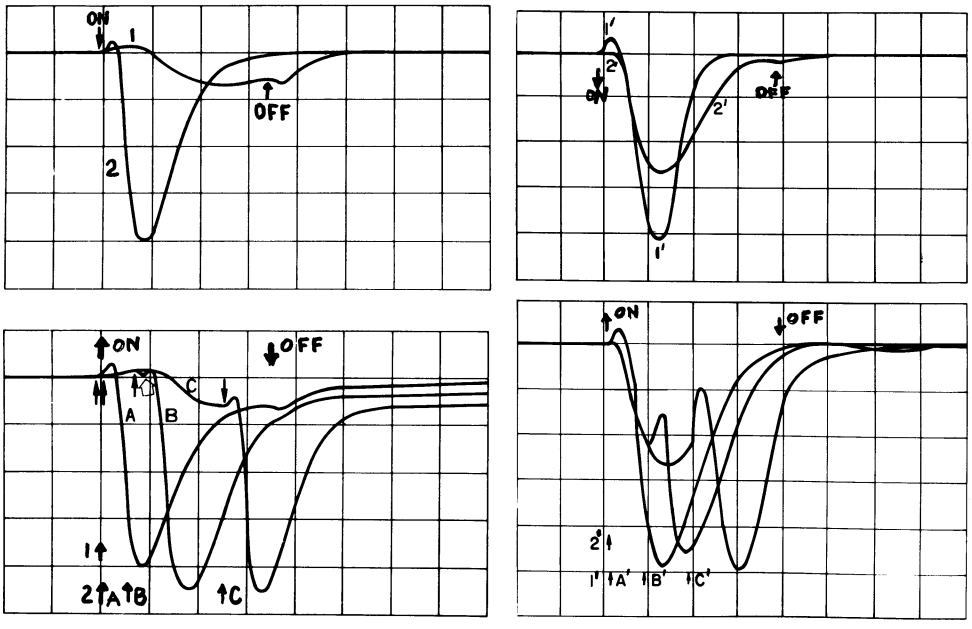

FIGURE 2. These are conventionally taken EOGs (Ottoson potentials). The horizontal scale is in sec, the vertical scale is in mv. We have one massive reversible electrode on the body of the frog, and the recording electrode is a pipette filled with Ringer-gelatin and just touching the mucus on the olfactory eminence. A laminar vortical flow of clean wet air is playing constantly on the mucosa, and into the stream we can inject our odors in such a way as to get sharp leading and trailing edges for the pulse of odorous air. This method is fully described in J. Physiol. (Gesteland et al., 1965.) At the top left we have superimposed two sweeps, one the response to a long weak puff of pyrrole, curve #1, and the other the response to a short stronger puff of n-butanol, #2. In the upper right we have superimposed the separate responses to a short strong puff of pyrrole, #1′, and to a long weak puff of n-butanol, #2′. In the bottom figures, the long weak puff is always turned on at the same point of the sweep for three different sweeps, and turned off at the same point. The short strong puff is turned on at different times for each of the three sweeps. Then the records are superimposed on each other. In the bottom left figure, curve B shows the inversion in sign of the initial positive transient to the short strong puff of n-butanol when it occurs during the initial positive transient due to the long weak puff of pyrrole. In the bottom right figure, curves B and C show the enhancement of the initial positive transient due to a short strong puff of pyrrole when the stimuli are given during the negative response to a long weak puff of n-butanol. See text.

effects with grossly recorded electrical measures on the mucosa. To this end, we have used the gross generator signals as well as gross "impedance" measures that treat mucosal elements as a transmission line.

The electro-osmogram (EOG) (also called *electro-olfactogram* and *Ottoson potential* [Ottoson, 1956]) is recorded between the surface of the mucus on the olfactory eminence and a distant ground on the animal. The changes in voltage that are measured are due to changes in flow of current between the outer parts of odor-sensitive elements and their inner parts, for both electrodes are in the external medium. These transients are not due to junction potentials, for the coupling electrodes are fluid bridges, and a significant change in current flow occurs between them when their leads are connected together and an odor is given. As shown in Fig. 2, there seem to be at least two processes at work in generating the osmogram. One tends to displace the voltage of the mucosal electrode in a negative direction, the other in a positive direction. Both processes are probably always evoked by an odor, and this is suggested by the response to pyrrole. When pyrrole is given alone, at moderate strength and for a few seconds, an off-response occurs (#4 in Fig. 3) when the flow of odor is stopped, as if another odor had been turned on, or as if one process set up by the stimulus outlasts another. If we revert back to Fig. 2 we can study some relations between the two processes. On the left side, at the top, we have the osmograms, superimposed, to a long weak puff of pyrrole given alone (1) and a short, stronger puff of *n*-butanol given alone (2). When we pair these stimuli, always giving (1) so that we turn it on at *ON* and off at *OFF*, and at different times with respect to *ON* we turn on (2) at the indicated points, we get interaction curves, three of which are shown superimposed at the bottom left of Fig. 2. Curve A occurs when both (1) and (2) are given simultaneously. Curve *B* is more interesting. The initial positive-going transient of *n*-butanol, seen when (2) occurs alone, is inverted in sign when it occurs during the initial positive transient due to (1). In curve *C* the transient becomes positive again when it occurs during the negative swing due to (1). On the right side of Fig. 2 we have at the top superimposed the responses to a short strong puff of pyrrole (1′) and a long weak puff of *n*-butanol (2′), each given alone. At the bottom figure we turn (2′) on at *ON* and off at *OFF* for each sweep. But now we stagger (1′) with respect to *ON* and superimpose three of the curves. Here you see that the initial positive transient of (1′) is much enhanced by occurring during the negative phase of the response to (2′). This initial positive transient acts as if it were due

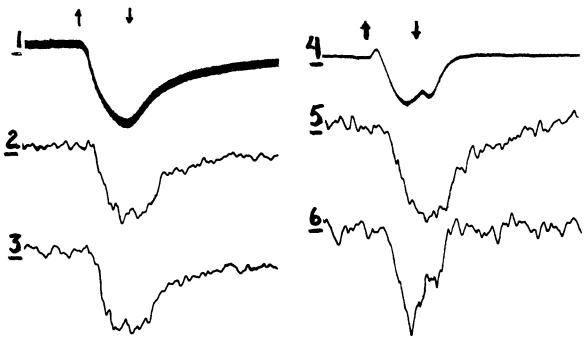

FIGURE 3. The top left record is the EOG response due to a medium-strong, moderate-duration puff of tetraethyl tin. The response is on the same scale as that in Fig. 2. The top right record is the EOG to a medium-strong, moderate-duration puff of pyrrole on the same scale. A 40 cycle/sec sine wave of current is being introduced a short distance away through a Ringer-gelatin pipette touching the mucus. This a-c signal is of about 2 mv amplitude where the signal injection electrode just touches the mucus. It barely thickens the base-line of the EOGs. This a-c signal at the recording electrode is filtered out through a phase-locked amplifier (Princeton Instruments), and balanced out to a small residue. This residue is studied for change in magnitude (the second line on both left and right sets of records) and phase angle (the third line in both sets) during the EOG. The measurements are really not quite of pure magnitude and phase angle, but these terms will do for the crudeness of the measure. The two imbalances are filtered and smoothed to yield the records shown. The sweep is 10 sec long.

It is apparent that the two different measures track each other for the most part to the stimulus with tetraethyl tin, but are markedly different for the stimulus with pyrrole. Indeed it would seem that the "phase angle" change stops sharply with the trailing edge of the pyrrole stimulus, while the "amplitude" change falls slowly, suggesting that the off-effect, shown in the EOG may be due to the release of one process from an interaction with another. For further details see Gesteland et al. (1965).

to a restorative force tending to bring the osmogram to some fixed level of voltage. Such behavior of transients is seen elsewhere in nervous tissue, and is usually explained as being due to the opening of channels through which an ionic species flows whose chemical potential is almost the same as the resting membrane potential. One sees this best in the classical study of invertebrate stretch receptors by Eyzaguirre and Kuffler (1955).

On such a basis, an excitatory (or exalting) event occurs with the opening of a membrane gate to Na^+ at a transductive site; and Na^+, flowing down its own gradient, constitutes a current inward at the site and hence a current outward everywhere else on the cell, including the electrically excitable patch where the axonal impulse begins. An inhibitory (or depressive) event occurs with the opening of a membrane gate to Cl^- or K^+ at a transductive site. But, because the chemical potential of these species is almost the same as the membrane potential of the resting cell, such an event, occurring alone, is not accompanied by any appreciable flow

of current at the site. If, however, there is in the vicinity a current being generated across the membrane, this increased conductivity to either Cl^- or K^+ shunts a good part of that current. The inhibitory event tends to hold the membrane to a constant voltage close to that measured as the resting potential. From the point of view of an electrically excitable patch elsewhere on the cell, the gating of Na^+ at a transductive site is seen as a disturbance that tends to drive current outward across the patch; while the gating of Cl^- or K^+ in the region where the Na^+ is being gated, diminishes the apparent strength of that excitatory event with respect to that patch. These two kinds of processes, current-generating and shunting, are the same that are used to explain excitatory and inhibitory synaptic events. We are not interested in making any quantitative guesses, but only mean to show that the same sorts of processes can be adduced for chemical receptors as for synaptic systems where chemical transmission is suspected.

While the nonlinearities exhibited in Fig. 2 support the notion of a dual ionic control to account for the responses in individual axons, we would have an even stronger case if we could find evidence from still another method. Because the receptors are arrayed in a sheet underlaid with a basement membrane of high resistivity and overlaid with air, we can treat the sheet as a complex delay line formed by the imbedding of many small a-c filters (the cells) in a resistive medium (the intercellular space and mucus). If we apply an a-c current somewhere between the overlying mucus and a ground elsewhere on the animal, some of the current will pass through the cells into the axons that issue through the basement membrane, some will flow through the basement membrane, some will flow through the mucus, some laterally between cells, etc. It would be folly to set up equations for so complex a situation. Nevertheless, we can say that if the electrical conductances in cell membranes change, the distribution of flow of the applied a-c current will also change. Thus, if we have a pick-up electrode elsewhere on the olfactory mucosa, and balance out the recorded a-c signal, we ought to record a bridge imbalance during the giving of an odor. Now, if there are at least two sorts of changes of conductance, and each is governed by a different set of time dependencies (as in the Hodgkin-Huxley equations), then there might be a frequency at which one process will primarily appear as a change in amplitude in the balanced-out signal, and the other as a change in phase. If we found such a frequency, it is unlikely that it would have any analytical value in so complex a situation, but it would support a logical cut between receptors governed by variation of a single ionic parameter and those governed by variations of two or more such parameters. In Fig. 3 you can see what happens in the case of pyrrole. The fact that at 40 cycle/sec with the electrodes used, we found that the change in amplitude of the bridged-out a-c signal decayed slowly after the turning off of the stimulus, whereas the change in phase angle decayed promptly, heartens us in the interpretation that the osmogram results from the interplay of at least two processes. The off-effect to pyrrole seems to result from the quick decay of one with respect to the other.

The very complexity that prevents us from treating this kind of measurement in detail does not deter us from using empirically the trivariance of the three measures shown, the osmogram and the amplitude and phase angle changes of the bridged-out a-c signal; for there seems to be no unique relation between the three. Thus we sought to find out whether different odors could be told apart only by looking simultaneously at all three signals. In this we were surprisingly successful, for all three taken together served to discriminate many more odors than any two taken together. What clear distinctions we can make in such a three-dimensional space only attest to the trivariance of the measures (just as trichromacy allows more color distinction than dichromacy).

If we suspect that there are two processes that can be turned on in a receptor, and that any odor turns on one or the other or even both in varying amounts, where is this transduction done? As neurophysiologists we naturally look first to the cell membrane as the all-purpose device. The membrane is indeed continuous over the extraordinarily long thin cilia, first properly described by Reese (1965). But a cilium itself is so fine that great differences in strength of electrotonic signal would occur at the cell body with small differences in site of adsorption of a molecule if no electrical regeneration took place down the cilium, which is approximately 0.2 mm long at maximum. It may well be that the accident of place of absorption of a particular molecule for the individual receptor accounts for the ordering code. But the growth rate of cilia is said to be high, and so the code would be changing steadily —an intolerable notion.

There are alternatives. Reese himself suggests that the cilium sustains impulses, since it is of about the same diameter as the axon leaving the receptor. But another ciliary function is mechanical action. There is an impressive ignorance of sensory cilia wherever they occur, but we do know that in protozoa, motile cilia change the direction in which they are stiff and that in which they are flexible with changes in ionic composition of the medium; e.g., reversal of ciliary stroke usually accompanies

increased K^+. When protozoan cilia reverse stroke to nonionic agents, how is that reversal accomplished? We have considered a view which, though probably incorrect, ought to be presented clearly enough to offer one an image. Suppose that along the ciliary surface there are receptive sites that, instead of only depolarizing or changing conductance of the membrane (to divalent as to monovalent ions), set up a ciliary wave (of tension perhaps, rather than gross movement) that is transmitted back to the junction of cilium and olfactory rod. We know that a cilium is capable of altering its stiffness, as well as the direction in which it moves (see any recent work in protozoology), and that both bending and stiffening travel in a wave along it. Suppose the flagellum lies in a highly viscous medium such as mucus. Our image is that wave patterns of different sorts are possible in such a case and could be transmitted without much actual movement as a kind of joy-stick wiggling at the ciliary base with adjustable direction and amplitude and even frequency. After all, considering the signal corruption to be expected from electronus, are we really worse off with mechanical signaling? There is certainly no sense in speculating beyond this point such as attributing control of the different ions to angle of the joy-stick, etc.

CODING

Now let us return to the actual signals once more and simplify the coding problem a bit by ignoring the obvious differences in timing of responses and the occasional alternating exaltations and depression of firing evoked in a single axon. Instead, let us take it that all pure compounds can be ordered from greatest exaltation to greatest depression with respect to a single axon. Then we can lay out a table of stimulus equivalents with respect to that axon and so transform an input, requiring many parameters to describe it, into an output requiring but one, i.e., the probability of occurrence of an impulse as a function of time. (This is the sort of transformation that is made by all neurons; each is affected by many different sorts of influence, but the effects of the influence are only seen as variations in the frequency of firing. Handling this kind of transformation is not within the province of this discussion. Besides we admit we can't treat it at all.) It is now important to ask: Suppose that two compounds separately evoke the same response; if we make a stimulus composed of a mixture of the two, can the response be seen as equivalent to the result of changing the concentration of either one alone? Well, sometimes it can but sometimes it cannot. For example, we have seen a single axon in which little change was produced either by ethanol or musk given separately in any concentration. But when the two were given together in moderate concentration, a strong exaltation occurred. Similarly, the depressing influence of a single compound on the firing rate of an axon does not characterize the manner in which it will change the response to an exalting compound when the two substances are given together.

In the case of musk and ethanol it is perhaps possible that a chemical reaction takes place—and in the case of other pairs of substances it is possible that they might weakly cling to each other to present new steric conformations, but it is also possible that absorptive sites interact nonlinearly. Thus we must consider not only the ordering of any set of odors, vis-a-vis an axon, but also the ordering of all mixtures in the set, since, at present, their actions cannot be seen as simple combinations of the actions of the compounds given separately. The situation takes on further poignancy when we realize that many substances, when zone-purified, lose their characteristic odors only to readopt them slowly. So, the mephitic skatol turns "not unpleasant" after such overpurification. The smell of many a "pure" compound may in fact be due to some characteristic mixture of products of oxidation, polymers, etc., along with the substance itself.

There are several different ways to account for the unpredictability of response to mixtures of odors. One is chemical binding or loose coupling of the compounds among themselves to form differently configured particles, as has just been mentioned. But another is interdependence or contingent operation of olfactory sites on the same receptor. Probably all of us have had the same image of the unknown transduction. There, on the surface of the cilium or cell wall, is a molecular trap, an ophidian proteinaceous affair that, on receiving a molecule of the right shape, coils about it in allosteric embrace. In so changing, the trap opens an ionic gate by some mechanical or electrical or chemical action, and through this gate particular ions speed, carrying electrically the glad tidings that a molecule is captured. If we think of a trap as an enzyme-like thing, it may not be able to exhibit a shape for accepting A except as it had already accepted B and become formed (activated) by it. Or, the trap having accepted B, the act of accepting A affects the ionic gate otherwise than if there had been no B. However, this intramolecular view of the trap is relevant only for *high* concentrations of odor, whereas odors interact at low concentrations too. We are willing to suppose that contiguous traps may also affect each other so that the response to substance A is contingent on whether a substance B or even another molecule

of A has been clasped elsewhere on the same membrane. We know of such field effects with other chemoreceptors—e.g., the action of botulinus toxin on motor end plates—the action of colicine K on oxidative phosphorylation in *E. coli*—the action of denervation on muscle fibers, etc.

Such a picture, if even partly correct, makes a temporarily hopeless object for the biophysicist. The raw coding data that evoked this picture also baffles the decipherer. This sort of coding can be called holistic. It is as if every axon expresses a point of view with respect to all compounds and combinations of compounds, and each axon has a separate point of view. Considered in the limit, such a system was first described by Leibnitz in his *Monadology*. A restricted case is found in the now popular optical hologram in which every point in the representation of the scene expresses an integral function with respect to the whole scene. But integral functions need not be simple and linear like Fourier transforms: if only they are regular, holistic coding preserves information about relations between elements represented. So, for example, in studying the frog's eye one finds certain contextual matters around a point more significant in determining the firing of a ganglion cell than the light value at that point. One of the advantages of holistic codes is that certain relations between elements are encoded together with the elements themselves, so that, insofar as form inheres in relations between elements, resolution of forms is what increases as the number of points of view increases.

This type of coding, implied by the very idea of "receptive field" as first voiced by Kuffler, occurs everywhere in physiology. When it is possible to guess at the nature of the transformation, as we did with the frog's eye, then all is golden; one has invariants by the tail and one's colleagues murmur approval. True, the results cannot be handled analytically, but then it is only the last grey lackeys of positivism who still hope for the quick and dirty algorithm to plug into a computer. Real nervous systems are above such low cunning. Yet we had not bargained for complete anarchy such as we seem to have in olfactory receptors. If we hold to the consequences of our studies, that every receptor differs in its ordering of odors from every other, we have a result worthy of the Royal Academy of Laputa. But the lie is given us by our gross records. How is it possible that there should be such consistent changes in the electro-osmogram with pyrrole, ethanol, menthol, that we can pick out by crude signs one from the other, if there were no underlying regularities? That, on the level of the single fiber, we have missed these regularities is beyond doubt; and it is to them that we must go in order to characterize the kind of transformation that the whole receptor makes on the world of odors.

So, while we are pleased at having been able to record from single elements sufficiently long to study them, and are also now certain that each receptor has a "receptive field" in odor space, we are at the same time unhappy at not being able to give the varieties of receptive field which must be there, both by reasoning backward from the perceptions and by observing the grosser electrical signs of receptor activity. We reject, on superstitious grounds, the possibility that receptors are not to be grouped or categorized in terms of operations on odors, and can but say that the lack of form in our results reflects only that we have not yet intuited the proper forms that are certainly there.

We shall now restate our crude image of the nature of olfactory coding.

1. Almost certainly there are specific chemically sensitive molecules or molecular traps. This follows at least from the partial congenital anosmias. How many species of trap there are cannot be said. But there are more than the three that we mentioned—we suspect many, many more.

2. Either every receptor has many species of trap on it or, if there is only one species of trap to a fiber, that trap has a complicated action spectrum in odor space. This follows from the ordering of any arbitrary set of odors by any receptor. Each trap may have one or the other (or both) of two signaling actions on clasping a molecule. It may activate a current source, i.e., have an excitatory influence, or it may activate a current shunt, i.e., have an inhibitory influence. (These actions of the molecular traps may be due either to direct gating of ionic processes in the membrane where the traps are, or they may be indirect by virtue of transmitted mechanical signals to the base of the cilium.)

3. The actions of the molecular traps on a single receptor are not linearly combinational or else, if there is only one sort of trap for each receptor, then the action of odors on this trap is not simply combinational. This, of course, follows trivially from the fact that the effects of current sources and shunts do not combine linearly in a transmission-line network of them. But we mean this in a nontrivial way. Two species of odor may each alone activate current sources separately on the same receptor—but in combination they may not. We have seen enough such cases to suspect that this highly nonlinear interaction is not an artifact. It is one of the factors that inclines us to believe that the action of the molecular traps on ionic gating may not be direct but rather goes through an intermediate step. Our suggestion of a mechanical transmission down the cilium is not strong but reflects that belief.

4. The receptor sees pure odors and combinations of odors along a single dimension (if we exclude relative timing of response) from most exalting to most depressing. This constitutes a single point of view. If every fiber in N_I has a different point of view from every other fiber (i.e., a different ordering of the odors) then each fiber represents a different dimension in a coordinate system. In this case we probably have more axes in the space than discriminable positions along any axis. (A similar condition occurs in the optical holograph. Every point along the developed film has either a silver particle or none. Each point expresses, by this 1 or 0 mark, the result of combining phase and amplitude information from every point in the scene photographed holographically.)

5. Such a coding procedure preserves relations between odors in combination. We do not yet have any inkling of these rules of encoding.

6. The initial transductive process is not adduceable from receptor action and the psychological laws for smell cannot be synthesized from knowing what we have said about receptors, even if we suppose that all we have said can be confirmed. All we can say is that such a receptor language is of the same form as other language of the nervous system, has the same kind of provenance, and resists reading for the same reasons. That is not saying much.

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