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TASTE RESPONSES IN THE NUCLEUS TRACTUS SOLITARIUS OF SODIUM-DEPRIVED RATS

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SUMMARY

1. Maintenance of sodium balance is crucial to mammals and is expressed in the innate salt appetite. With depletion, sodium preference is exaggerated, hypertonic solutions accepted and salt balance restored. This compensatory behaviour is thought to result from a centrally induced change in taste responsiveness. This proposal was tested by recording taste activity from ninety-four single neurones in the nucleus tractus solitarius of sodium-replete ($N = 44$) and of deprived ($N = 50$) rats. Twelve Wistar rats were given a nominally sodium-free diet for 10–13 days, and the resulting sodium depletion confirmed by flame photometry of their urine. Nine rats provided control data. Taste stimuli included five concentrations of NaCl (0·003–0·3 M) plus eight other salts, acids, sugars and alkaloids.

2. Taste responsiveness was generally reduced in sodium-depleted rats. Spontaneous activity was 33% lower while responses to sodium salts lagged by a mean of 30%, to acids by 25% and to bitter salts and quinine by 17%. Mean activity to sugars was 60% higher in the deprived group.

3. Activity in sugar- and salt-profile neurones was most affected. In deprived animals responses to sodium salts were lower by 80% among salt-profile cells while among sugar-profile neurones activity to these stimuli was nearly 10 times greater than in controls. These changes in activity resulted in a dramatic shift in the participation of sodium- and sugar-profile cells in the afferent signal for NaCl. In replete animals 60% of sodium-induced activity was transmitted through salt-profile cells while only 1% occurred in sugar-profile neurones. In deprived subjects this situation was nearly reversed as 7% of the total NaCl response was conveyed through salt-profile cells while the contribution of neurones with sugar-profiles rose to 46%.

4. Multidimensional stimulus spaces based on average activity in each of four identifiable neurone subgroups demonstrated a shift in the affiliation of sodium salts away from bitter and acid stimuli and towards sugars.

5. These results confirm earlier findings from the chorda tympani that sodium deprivation suppresses activity evoked by sodium salts. However the application of

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more recent analytical procedures permits quite a different interpretation of this finding. The overall decrease is merely the net effect of a shift in the major responsibility for encoding sodium from salt-profile neurones to those whose primary sensitivity is to sugars. The implication is that the mechanism for sodium repletion involves not only a change in perceived taste intensity as had been suggested, but, perhaps more importantly, an alteration in the perceived quality of sodium. This change may underlie the increased hedonic appeal for sodium that accompanies salt depletion, an appeal manifested in avid acceptance that is normally reserved for sweet stimuli.

INTRODUCTION

Mammals have evolved predominantly in sodium-poor environments. Thus most mammals seek out and consume sodium wherever it is found and, when plentiful, in excess of need. Both rats and humans select sodium salts in their diets, even when sodium replete (Richter, 1936; Bare, 1949; Denton 1976). This preference becomes exaggerated under conditions of sodium deficiency. Humans depleted by pathological states (Wilkins & Richter, 1940) or by experimental manipulation (McCance, 1936) show a pronounced craving for salt. Rodents subjected to adrenalectomy (Clark & Clausen, 1943; Carr, 1952; Epstein & Stellar, 1955; Fregley, 1958; Nachman, 1962; Harriman, 1967) or injections of formalin (Wolf & Steinbaum, 1965; Smith, Stricker & Morrison, 1969; Stricker & Wilson, 1970; Morrison & Young, 1972), cyclophosphamide (Mitchell, Parker & Woods, 1974), aldosterone (Wolf, 1964; Fregley & Waters, 1966; Wolf & Handel, 1966) or DOCA (Rice & Richter, 1943; Richter, 1956; Wolf & Quartermain, 1966) show sharp increases in salt consumption. This also occurs with dietary restriction of sodium (Fregley, Harper & Radford, 1965; Handel, 1965; Cullen & Harriman, 1973), a manipulation that more closely approximates the natural condition.

The capacity to adjust sodium intake to match need is apparently mediated through gustation. When taste input is interrupted by surgical intervention, adrenalectomized rats no longer compensate for sodium loss, even as the deficiency leads to their deaths (Richter, 1956). The putative involvement of gustation in maintaining sodium balance is reinforced by anatomical considerations. The area postrema, lesions of which result in exaggerated salt consumption (Contreras & Stetson, 1981), is thought to communicate by way of the reticular formation with gustatory nucleus tractus solitarius (Shapiro & Miselis, 1985), and parabrachial nuclei (Cedarbaum & Aghajanian, 1978; van der Kooy & Koda, 1983; Shapiro & Miselis, 1985). Thus a plausible circuit is available through which sodium imbalances could be detected and activity in the sensory system responsible for sodium restoration could be modified.

Electrophysiological studies, however, have not uniformly supported a dynamic role for gustation in sodium maintenance. Pfaffmann & Bare (1950) measured threshold responses in the whole chorda tympani (CT) nerve of the rat and found them to be unmodified by adrenalectomy. Nachman & Pfaffmann (1963) reported that sodium deprivation sufficient to cause a robust salt appetite had no effect on CT responses to a concentration series of NaCl solutions. Conversely, Contreras & Frank (1979) established sodium deficiency in rats through deprivation and discovered a significant reduction in whole CT responsiveness to NaCl. This decrement was

specific to NaCl among the four prototypical taste stimuli and existed only for concentrations at or above 0·03 M. Below this level, the difference between responses evoked from sodium-depleted and control animals diminished, to vanish at a common threshold of 0·001 M. When they took this analysis to the single-neurone level (Contreras, 1977; Contreras & Frank, 1979) Contreras and Frank found that most of the effect was attributable to reduced responsiveness among the 50% of neurones that responded best to NaCl. They interpreted these results to indicate that the gustatory appreciation of saltiness is conveyed exclusively through a neural channel 'staffed' by sodium-best taste cells; moreover, higher concentrations of sodium would be required to drive the now-insensitive channel to its normal activity levels, thus encouraging the rat towards sodium repletion.

While the effects of sodium depletion on taste activity in the peripheral nervous system remain at issue, those in the central nervous system have yet to be studied. The nucleus tractus solitarius (NTS), the first central relay for taste, receives not only gustatory information from the external environment, but also input regarding physiological condition by way of the vagus nerve. Its neurones communicate reciprocally with ventral forebrain regions concerned with hedonic appreciation and motivation, factors necessary for the maintenance of sodium balance. Taste cells in the NTS have already been shown to be sensitive to physiological condition (Glenn & Erickson, 1976; Giza & Scott, 1983; Chang & Scott, 1984*a*; Giza & Scott, 1987). Thus it is reasonable that an extraordinary need for sodium may be appreciated here. Therefore, we recorded the responses of single gustatory neurones in the NTS of sodium-deprived rats to a wide range of chemicals and compared them with corresponding records from non-deprived animals.

METHODS

Subjects and dietary conditions

Subjects were twenty-one Wistar rats of both sexes, weighing 310–420 g. Twelve animals, assigned to the sodium-deprived group, were individually housed and acclimated to a powdered sodium-deficient diet (6 mg sodium per 100 g; ICN Pharmaceuticals, Cleveland, OH, U.S.A.) which was supplemented by 1% (w/w) NaCl. After 5 days, sodium was withheld from the mixture. Subjects were maintained on the sodium-deficient diet alone for 10–13 days prior to recording. Nine other rats composed the sodium-replete group, and were given a diet of standard laboratory chow. Distilled water was available to all subjects *ad libitum*.

Prior to a recording session, a urine sample was collected and analysed for sodium content by flame photometry. The mean sodium loss for control rats was 2·78 mequiv in 24 h, while that for sodium-deprived animals was 0·07 ($t = 4{\cdot}29$; $P < 0{\cdot}01$). Salt-deprived rats maintained normal activity levels and gained weight at a rate typical of non-deprived animals. There was no indication that the physiological integrity of these rats was significantly compromised by the manipulation.

Surgery

Each subject was anaesthetized with an intraperitoneal injection of sodium pentobarbitone (50 mg/kg). A tracheotomy was performed and the oesophagus ligated to prevent suffocation and ingestion of stimuli. The head was fixed in a stereotax and a 5 × 10 mm rectangular section of skull was removed over the cerebellum to afford access to the NTS bilaterally. Body temperature was maintained between 36 and 38 °C. Heart rate was continuously monitored through subcutaneous electrodes. Expired CO_2 levels were analysed periodically and maintained between 3 and 4%.

Stimuli and stimulus delivery

Taste stimuli were five concentrations of NaCl (0·003–0·3 M in half-log molar steps), and single concentrations of KCl and Na_2SO_4 (0·1 M), $CaCl_2$ (0·3 M), fructose and sucrose (0·5 M), hydrochloric

and citric acids (0·01 M) and quinine hydrochloride (0·01 M). All chemicals were mixed in distilled water except for fructose and sucrose to which 5% tap water was added to increase conductivity and so ensure activation of a stimulus onset marker.

Five millilitres of each stimulus were sprayed into the mouth at a rate of 2 ml/s according to the method of Chang & Scott (1984*b*). Evoked activity was recorded for 10 s, the tongue was rinsed with 50–75 ml distilled water over a period of 30 s, and 45 s of rest was permitted to avoid adaptation effects. The moment of stimulus contact with the oral tissues was marked by a TTL logic device (Chang & Scott, 1984*b*).

TABLE 1. Breadth of sensitivity comparisons between neurones in deprived and replete rats

Number of neurones responding to all or part of the stimulus array

	Neurones (%)	
Stimuli	Replete	Deprived
10/10	17 (39)	22 (44)
9/10	9 (20)	18 (36)
8/10	8 (18)	4 (8)
7/10	2 (5)	3 (6)
6/10	3 (7)	0 (0)
4/10	5 (11)	3 (6)

Number of neurones responding to each stimulus

	Neurones (%)	
Stimulus	Replete	Deprived
0·30 M-NaCl	42 (95)	49 (98)
0·10 M-NaCl	40 (91)	48 (96)
0·10 M-Na_2SO_4	39 (89)	49 (98)
0·03 M-NaCl	38 (86)	47 (95)
0·01 M-HCl	38 (86)	44 (88)
0·10 M-KCl	37 (84)	46 (82)
0·50 M-sucrose	34 (77)	42 (84)
0·01 M-NaCl	34 (77)	45 (90)
0·01 M-quinine	33 (75)	42 (84)
0·003 M-NaCl	30 (68)	36 (72)

Breadth-of-tuning metric (±S.D.)

Replete	Deprived
0·70±0·17	0·76±0·16

Recording and histology

With the stereotaxic bite bar positioned 5·5 mm below the interaural line, typical electrode co-ordinates were 11·0–11·6 mm posterior to bregma, 1·5–1·7 mm lateral to the mid-line and 6·2–6·9 mm below the surface of the cerebellum. Single-unit potentials were isolated using pipettes filled with potassium citrate ($Z = 3$–8 MΩ). Signals were amplified, filtered and displayed by conventional techniques and recorded on magnetic tape for off-line analysis.

Electrolytic lesions were made at the final recording sites in three representative animals (20 μA for 20 s, electrode negative). Subjects were given a lethal dose of barbiturate and perfused with 10% formalin–saline. Brains were removed, blocked and frozen. Sections were made each 40 μm and stained with Cresyl Violet so that lesion sites could be identified.

Analyses

A PDP 11/03 computer was used to count action potentials for 3 s prior to (spontaneous) and 5 s following (evoked) stimulus presentation. The 5 s net (evoked minus spontaneous) post-stimulus response was divided into fifty 100 ms intervals. Figures from each interval were transferred to an IBM 3081D computer for calculation of response magnitude summaries and the following derived

analyses: hierarchical cluster analyses, interstimulus and interneuronal correlation matrices and multidimensional scaling.

Cluster analyses were used to identify groups of cells with similar functional properties, as measured by correlations between neurone response profiles across the stimulus array. Clusters were labelled according to which of the four prototypical taste stimuli (NaCl, HCl, sucrose or quinine) dominated the response profiles of their constituent neurones. Mean spontaneous and evoked rates were determined for each of the neural clusters as well as for the entire sample of cells.

RESULTS

Overview

Across all cells, significantly less activity was elicited by salts in sodium-deprived rats while responses to sugars were greater. More specifically, the response to sodium salts underwent a dramatic shift: in control rats 60% of the sodium-induced activity was conveyed through salt-profile neurones while only 1% occurred in sugar-profile cells; in salt-deprived rats, sodium neurones nearly fell silent, reducing their contribution to the code for NaCl to just 7% of the total activity, while the proportion carried by sugar-profile cells rose to 46%. The proportions carried by acid- and quinine-profile neurones remained rather constant. Thus the afferent signal for sodium salts shifted from a salt channel towards a sugar channel, possibly implicating a change in the taste quality for sodium in salt-deprived rats.

Spontaneous activity and response criterion

The criterion for an evoked response was a change in neural activity of 1·28 S.D. ($P < 0{\cdot}10$) from the mean spontaneous rate for each cell, sustained for 3 s.

In sodium-replete animals, mean spontaneous activity was $6{\cdot}1 \pm 6{\cdot}9$ spikes/s, a value typical of spontaneous discharge rates in anaesthetized rats (Doetsch & Erickson, 1970; Ganchrow & Erickson, 1970). In the sodium-deprived group, mean spontaneous rate was $4{\cdot}1 \pm 4{\cdot}2$ spikes/s. This moderate reduction in the overall rate, however, resulted from an interplay among larger changes in the spontaneous activities of neural subtypes. The spontaneous rate of cells that were most responsive to sodium was lower by 39% in sodium-deprived rats; among acid-profile neurones the difference was 60%. Conversely, spontaneous activity among cells whose response profiles were oriented towards sugars was 48% higher in the sodium-depleted group. While large variations in individual rates that are typical of taste cells render these differences non-significant, the pattern that they establish among identifiable subsets of neurones foretells the more impressive differences in evoked activity patterns described below.

Evoked activity

Breadth of responsiveness. The characteristic broad sensitivity of NTS taste cells was preserved in sodium-deprived rats. Of 563 total stimulus applications in replete animals, 81% fulfilled the criterion for excitation, 3% for inhibition and 16% evoked no response. The corresponding figures for 605 applications in deprived rats were 88% excitation, 1% inhibition and 11% no response. Three standard measures of breadth were applied: (1) the proportion of stimuli to which each neurone responded, (2) the proportion of neurones responding to each stimulus and (3) the breadth-of-tuning metric (Smith & Travers, 1979). By each of these, there were no

significant differences between deprived and replete animals. These figures are summarized in Table 1.

Evoked response rates. Across all stimuli and neurones, evoked responses were 22% lower in sodium-deprived rats than in controls ($t = 2{\cdot}61$; $P < 0{\cdot}01$). The effects, however, were differential by taste quality. Responses to NaCl showed the largest difference, 30% lower in sodium-deprived rats ($t = 2{\cdot}16$; $P < 0{\cdot}05$). Activity evoked by acids was 25% lower while responses to bitter salts and quinine were off by 17% (both non-significant). Conversely, responses to sugars were higher by 63% ($t = 2{\cdot}35$; $P < 0{\cdot}05$) in deprived subjects. These relationships are shown for each stimulus in Fig. 1.

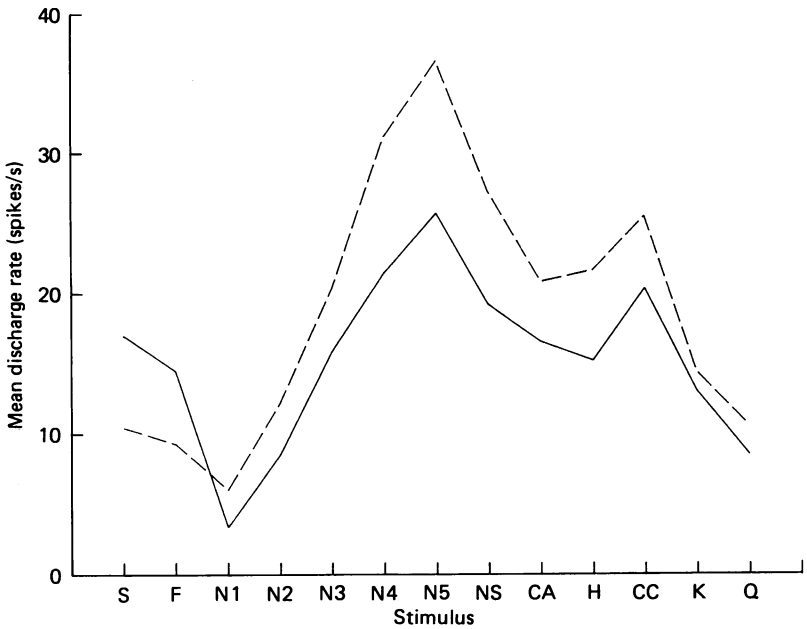

Fig. 1. Mean net (evoked minus spontaneous) discharge rates to each stimulus in sodium-deprived (continuous line) and replete rats (dashed line). Responses to all non-sugar stimuli were attenuated in deprived animals: activity to sodium salts (N1–N5, NS) was reduced by an average of 30%; to acids (CA, H) by 25% and to bitter salts and quinine (CC, K, Q), 17%. Conversely, responses to sugars (S, F) were increased by 63%. Abbreviations: S, 0·5 M-sucrose; F, 0·5 M-fructose; N1, 0·003 M-NaCl; N2, 0·01 M-NaCl; N3, 0·03 M-NaCl; N4, 0·1 M-NaCl; N5, 0·3 M-NaCl; NS, 0·1 M-Na_2SO_4; CA, 0·01 M-citric acid; H, 0·01 M-HCl; CC, 0·3 M-$CaCl_2$; K, 0·1 M-KCl; Q, 0·01 M-quinine hydrochloride.

The effects of sodium deprivation were perhaps more meaningful when the responses from different subtypes of cells were considered independently. While the response to NaCl was 30% lower across all cells in deprived rats, the more striking change was in the relative contributions of the various neural subgroups to that evoked activity. Salt-profile cells responded to 0·1 M-NaCl at a rate 80% lower than the corresponding neurones in sodium-replete rats. In absolute terms, the rate was 48·2 spikes/s in replete animals (a typical value in NTS) *vs.* 9·6 spikes/s in those that were deprived ($t = 2{\cdot}41$; $P < 0{\cdot}05$). This difference was not specific to NaCl, for salt-profile neurones showed an 82% reduction in responsiveness across the entire

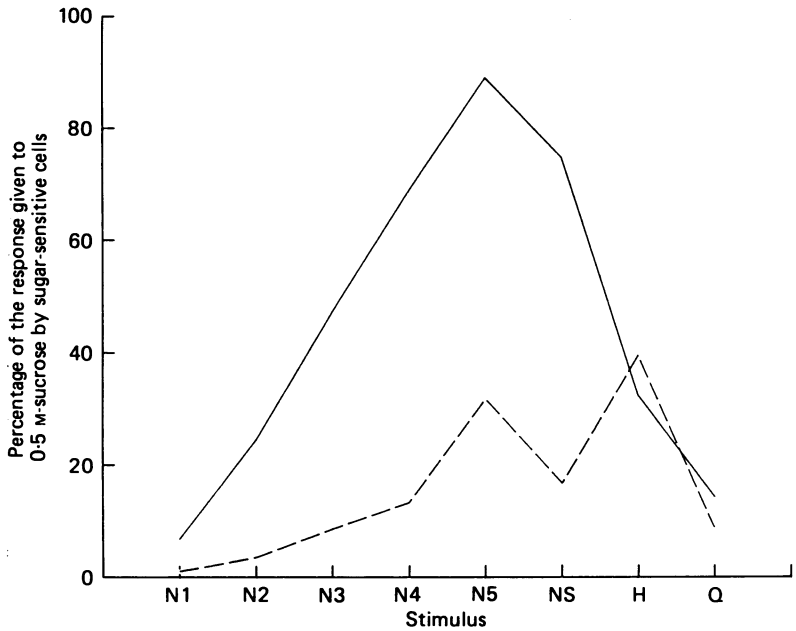

Fig. 2. Responses to sodium salts (N1–N5, NS), HCl and quinine plotted as percentages of the response given to sucrose by sweet-profile cells in sodium-deprived (continuous line) and replete rats (dashed line). Sodium salts consistently activated sweet-profile cells to a greater degree in deprived subjects whereas the effectiveness of HCl and quinine was nearly equal in both groups. Abbreviations are listed in Fig. 1.

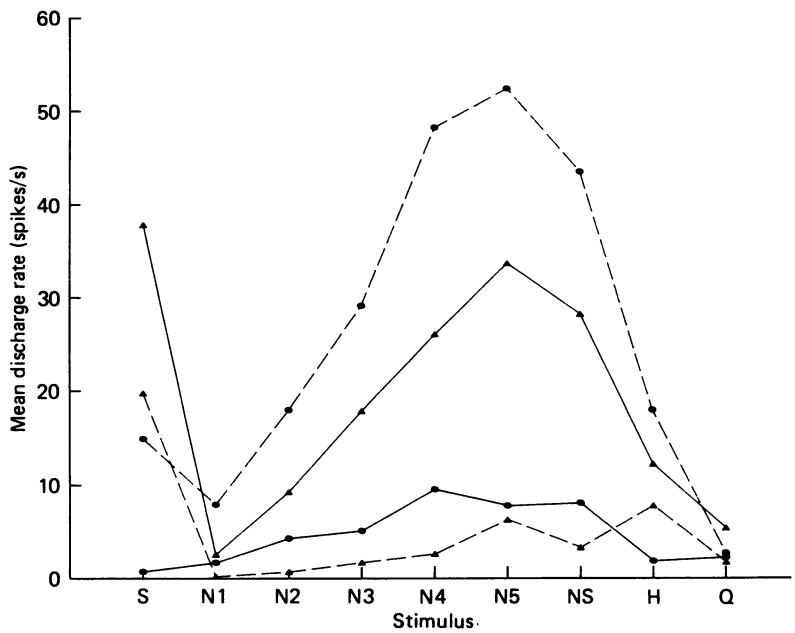

Fig. 3. Mean net discharge rates averaged across cells with salt and sugar profiles in sodium-deprived (continuous lines) and replete rats (dashed lines). Responses in salt-profile neurones (●) were reduced to all stimuli in deprived rats while activity in sugar-profile cells (▲) was increased. These shifts in activity were most pronounced for sodium salts resulting in response functions for sugar-profile neurones in deprived subjects which resembled those of salt-profile cells in replete animals. Abbreviations are listed in Fig. 1.

stimulus array ($t = 6{\cdot}25$; $P < 0{\cdot}001$). Cells with acid and quinine profiles showed mostly non-significant trends in the same direction: responses of acid cells were 39% lower overall and 40% lower to NaCl in deprived animals; those of quinine cells were 43% lower overall and 35% lower to NaCl.

In sharp contrast, activity in neurones with sugar profiles was 155% higher in deprived subjects ($t = 6{\cdot}88$; $P < 0{\cdot}001$) and the effect was greatest for NaCl. While responses evoked by sucrose were 91% greater and those to HCl and quinine were 96% higher (both non-significant), the mean activity elicited by 0·1 M-NaCl from sugar-profile cells was elevated by 891%, from 2·6 to 26·1 spikes/s in deprived rats

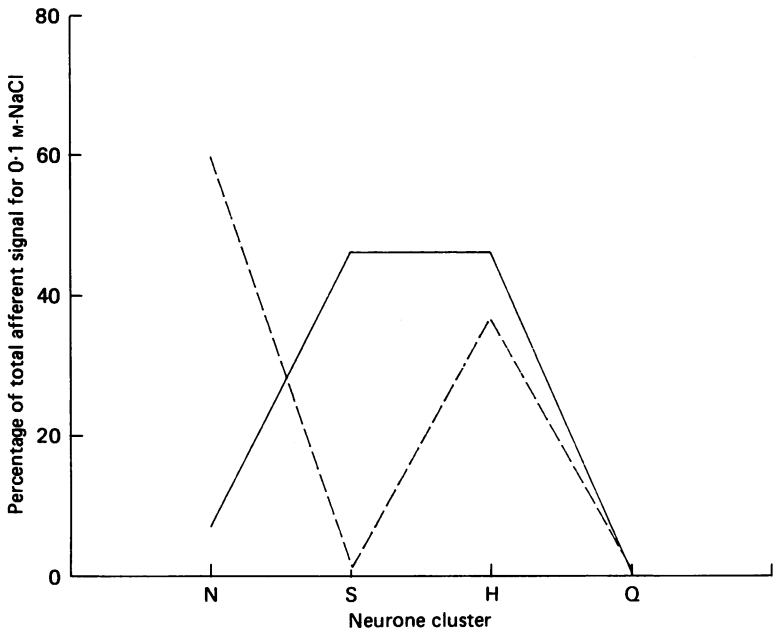

Fig. 4. Percentages of the total number of impulses elicited by 0·1 M-NaCl across neurone samples (i.e. the total afferent signal for salt) which occurred in each of four identifiable neurone clusters. In sodium-replete rats (dashed line) 60% of the NaCl signal was carried through salt-profile neurones (N) while only 1% occurred in sugar-profile cells (S). In deprived rats (continuous line) salt-profile cells nearly fell silent, contributing only 7% of the signal. The participation of neurones with sugar profiles, however, rose to 46%. The percentages of activity conveyed through acid- (H) and quinine-profile (Q) cells remained rather constant.

($t = 4{\cdot}26$; $P < 0{\cdot}001$). This strong, selective shift carried through Na_2SO_4 and all five concentrations of NaCl. Its magnitude is made evident in Fig. 2 where the response to each concentration of NaCl as well as to HCl and quinine is plotted as a function of the response to sucrose, thus factoring out the general increase in activity among this subgroup of neurones. While responses to HCl and quinine remain stable in these relative terms, those to sodium salts are markedly higher in deprived rats. Thus, set against the background of a change in general activity from cells with non-sugar profiles to those most responsive to sugars, there was a particularly large shift in responsibility for signalling NaCl from neurones with salt profiles to those with sugar profiles. In fact, in sodium-deprived rats, the mean responses of salt-profile and

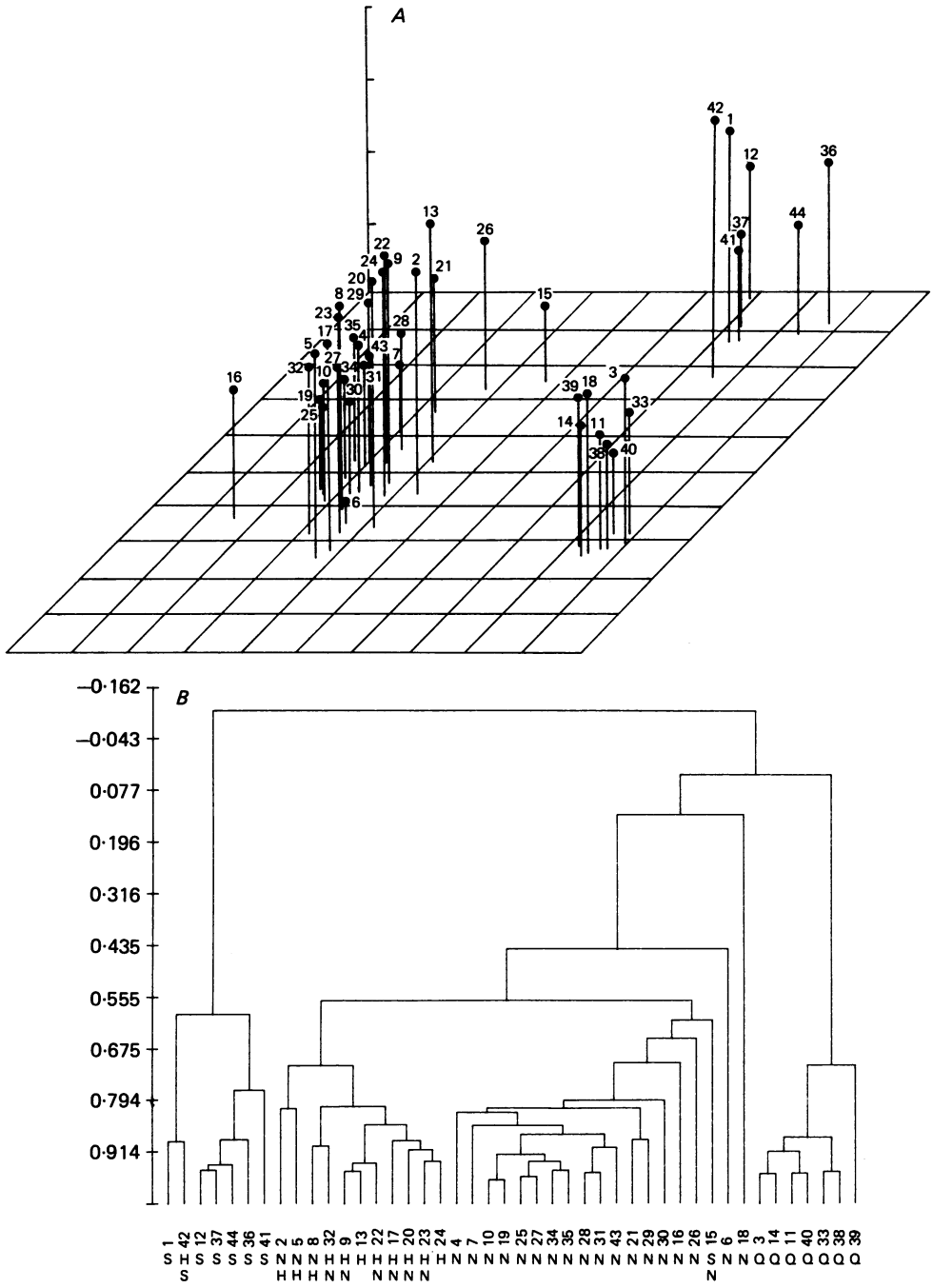

Fig. 5. *A*, three-dimensional 'neurone space' indicating relative similarities between response profiles evoked from cells in replete rats. Three distinct groups of neurones are evident each of which can be labelled according to their dominant sensitivity: sugar profile (upper right), quinine profile (lower right) and salt–acid profile (left). *B*, dendrogram resulting from a cluster analysis of the same neurone profiles used in *A*. Pairs or groups of cells are connected with horizontal lines at heights corresponding to correlations between profiles. Below each cell is marked the stimulus quality which evoked its greatest response followed by any other quality which was more than 85% as effective. In both *A* and *B* neurones are numbered in the order in which they were isolated. Neurones: N, salt profile; S, sugar profile; H, acid profile; Q, quinine profile.

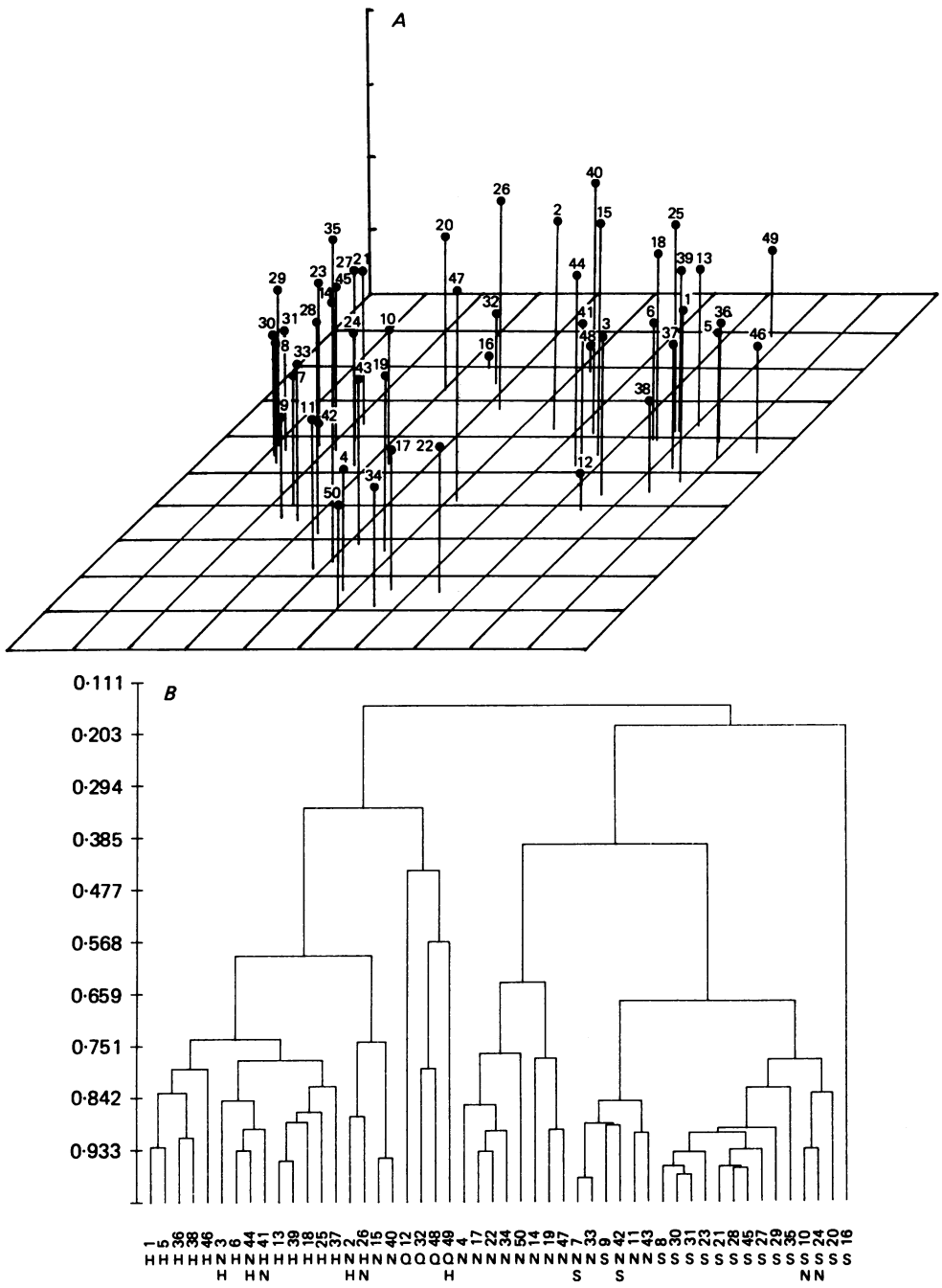

Fig. 6. For legend see facing page.

sugar-profile neurones to NaCl and sucrose were nearly reversed from the normal patterns they showed in replete subjects (Fig. 3).

The full impact of this shift was apparent when the mean increase of 891 % to NaCl among sugar-profile cells was combined with the finding that the proportion of these cells in the neural sample increased from 16% in replete rats to 38% in sodium-deprived animals, while the mean decrease of 80% to NaCl in salt-profile neurones was accentuated by a reduction in the proportion of these cells from 39% in replete animals to 16% in those that were deprived. Taken together, these data indicate that the proportion of the total afferent signal for 0·1 M-NaCl that was carried by salt-profile neurones declined from 60% in replete subjects to 7% in deprived rats while the proportion carried by sugar-profile cells rose from 1 to 46% ($\chi^2 = 90{\cdot}48$; $P < 0{\cdot}001$; Fig. 4). Most of the remaining signal for NaCl was carried by acid-profile neurones that also responded well to sodium.

Neurone types. The identity of the neurone types referred to in the analyses above were determined by the functional properties of the taste cells we studied. Despite their broad sensitivity, taste cells frequently have predictable response profiles across a stimulus array. If correlation coefficients are calculated between the response profiles of each possible pair of cells in the neural sample, there will tend to be clusters of high, medium and low coefficients signalling the possible presence of a discrete number of identifiable types of taste cells. The full matrix of correlations may be spatially represented through a multidimensional scaling routine (Guttman, 1968), the product of which is a 'neurone space' such as appears in Fig. 5*A*. Neurones are numbered in the order in which they were isolated and are placed in this three-dimensional space according to the relative similarities of their response profiles across this stimulus array. The neurone space of Fig. 5*A* is derived from the responses of sodium-replete rats. There appear to be three distinct groups which can be labelled according to their dominant sensitivities: sugar profile (upper right), quinine profile (lower right) and salt–acid profile (left). Within the last group, neurones with salt-oriented profiles are towards the top of the figure and those that are acid oriented are towards the bottom. A cluster analysis of this multidimensional space (Wishart, 1978) is shown in Fig. 5*B* in the form of a dendrogram. Cells have the same numbers as in the neurone space and beneath each is marked the stimulus that evoked its largest response followed by any other chemical that was more than 85% as effective. The dendrogram permits a more thorough statistical analysis of the groupings in the neurone space. The quinine-profile cluster is on the right of the

Fig. 6. Three-dimensional 'neurone space' indicating similarities between neurone profiles in sodium-deprived rats. Clusters of cell profiles are less distinct than for replete subjects primarily due to the disintegration of the large acid–salt-profile group in the centre of the figure. *B*, dendrogram resulting from a cluster analysis of neurone profiles in deprived rats. Two major clusters of neurones are identifiable: to the left of the figure are acid-profile cells 1–40 ($N = 18$) and quinine-profile cells (12–49, $N = 4$). The right side of the dendrogram is occupied by salt-profile (cells 4–47, $N = 8$) and sugar-profile (cells 7–20, $N = 19$) groups. It is perhaps the most meaningful finding of this analysis that, in deprived rats, salt-profile cells reduce their relationship with acid cells and become more functionally similar to sugar-profile neurones. In both *A* and *B*, neurones are numbered in the order in which they were recorded.

dendrogram and is composed of cells whose response profiles are, with the exception of neurone 39, significantly intercorrelated. The sugar-profile cluster is on the left, markedly separate from all other cells. The centre of the dendrogram is occupied by the salt–acid-oriented cells, the constituents of which are significantly intercorrelated (save for neurone 18). It is composed of two subgroups, each containing cells whose profiles are significantly intercorrelated: acid–salt towards the left (neurones 2–24, $N = 11$) and salt alone towards the right (neurones 4–15, $N = 17$). The neurone space of Fig. 5*A* and the cluster analysis of 5*B* are both typical of the gustatory neuronal arrangements reported by others, though the group of quinine-profile cells is rather larger and more distinct than is commonly found (Woolston & Erickson, 1979; Smith, van Buskirk, Travers & Bieber, 1983; Chang & Scott, 1984*a*; Scott & Chang, 1984; Scott, Yaxley, Sienkiewicz & Rolls, 1986*a*).

The corresponding analyses based on responses from sodium-deprived rats are presented in Fig. 6. From the neurone space of Fig. 6*A*, it is clear that clusters are less distinct. The blurring of group boundaries results from a loss of coherence of the large acid–salt-oriented cluster. However, the disintegration of this group is orderly: neurones with acid orientation are positioned nearer the quinine-profile cluster while those that respond more specifically to sodium salts are affiliated with sugar-profile cells. This modified arrangement is shown more clearly in Fig. 6*B*. Disregarding outlying cell 16, two major clusters are identifiable in the dendrogram. To the left is a large federation of acid-profile (cells 1–40, $N = 18$) and quinine-profile (cells 12–49, $N = 4$) neurones, in which all the constituents of the acid-profile group are significantly intercorrelated. To the right are salt-profile (cells 4–47, $N = 8$) and sugar-profile (cells 7–20, $N = 19$) groups, the profiles of which are significantly intercorrelated. It is perhaps the most important finding of this analysis of neurone profiles that, in sodium-deprived rats, salt-profile cells reduce their relationship with acid cells and become more functionally similar to sugar-profile neurones. Whereas the sharpest distinction in sodium-replete subjects is between sugar and non-sugar profiles, the clearest separation becomes salt–sugar *vs.* acid–quinine in deprived animals. This modification presumably results from the increased burden that sugar-profile neurones assume for coding sodium, as detailed above.

Classifications among neurones are based on similarities of response profiles which, in turn, are derived from spike rates. Since we are unable to record the activity of these cells continuously throughout the 10- to 13-day deprivation period, we cannot demonstrate conclusively that sugar-profile cells increased their responsiveness to salt in sodium-deprived rats as opposed to the converse, i.e. salt-profile cells may have become more responsive to sugars, causing their inclusion in a sugar-oriented cluster. The former interpretation is preferred for two reasons: (1) the profiles of these cells, while indicating considerable responsiveness to sodium, are still dominated by activity evoked by sugars, and (2) the experimental manipulation was of sodium; increased responsiveness to sugars among salt-profile cells is not an appropriate strategy to address a salt deficiency, whereas increased responsiveness to salt among cells associated with highly appetitive stimuli – the sugar cluster – is. Thus this conclusion is consistent with and explains the rat's behaviour.

Stimulus quality. The preceding section implies that the profiles which characterized salt-profile and sugar-profile cells became more similar following deprivation. Moreover, a massive increase in response to sodium among sugar-profile neurones

was suggested as the basis for this increased similarity. If, as a consequence of salt deprivation, neural activity representing sodium and sugars is being carried by the same or more functionally similar neurones, then the patterns of activity they evoke – and, by implication, their taste qualities – should be more closely related. This appears to be the case.

If the evoked activity of taste neurones within each neurone type is averaged to establish the mean response in that channel, then the taste quality of each chemical

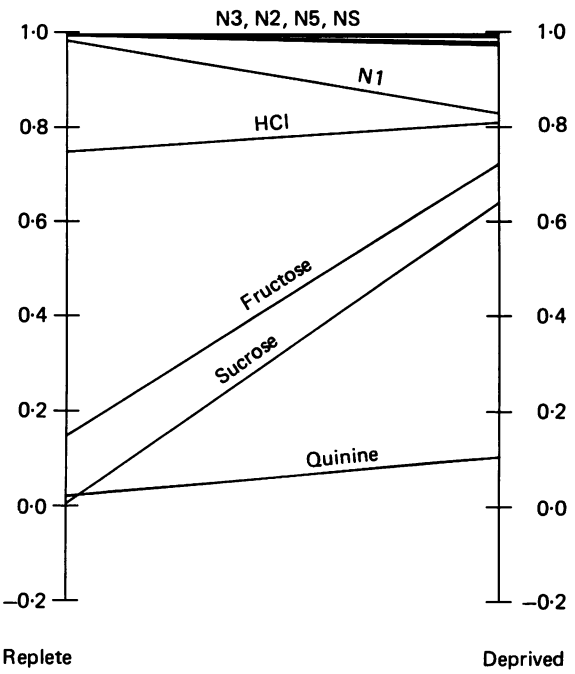

Fig. 7. A comparison of mean interstimulus correlation coefficients between 0·1 M-NaCl and other stimuli in replete and deprived subjects. While correlations between 0·1 M-NaCl and other sodium salts (N1–N3, N5, NS), HCl and quinine remained rather constant, those between NaCl and sugars rose dramatically in deprived rats. Abbreviations are given in Fig. 1.

will be represented by a 4-point profile (this 'stimulus profile' is analogous to the 'neurone profiles' that were used earlier to define neurone types). Correlation coefficients may be calculated between all possible pairs of profiles to determine relative similarities among taste qualities. Such an analysis, performed on the responses from both groups of rats, indicates that the relationship of sodium salts to acids and quinine, and to one another across concentrations, was rather stable (Fig. 7). In contrast to this relative stability, the relationship between sodium salts and sugars was much closer in deprived subjects. The correlation between 0·1 M-NaCl and 0·5 M-sucrose increased from +0·01 in replete rats to +0·67. The corresponding coefficients for fructose were +0·16 to +0·73 (Fig. 7). The mean shift across all twelve sodium–sugar pairings (six sodium salts × two sugars) was from +0·07 to +0·64.

A representation of these relationships is shown in the multidimensional spaces of Fig. 8. The space derived from the responses of replete animals (Fig. 8*A*) is

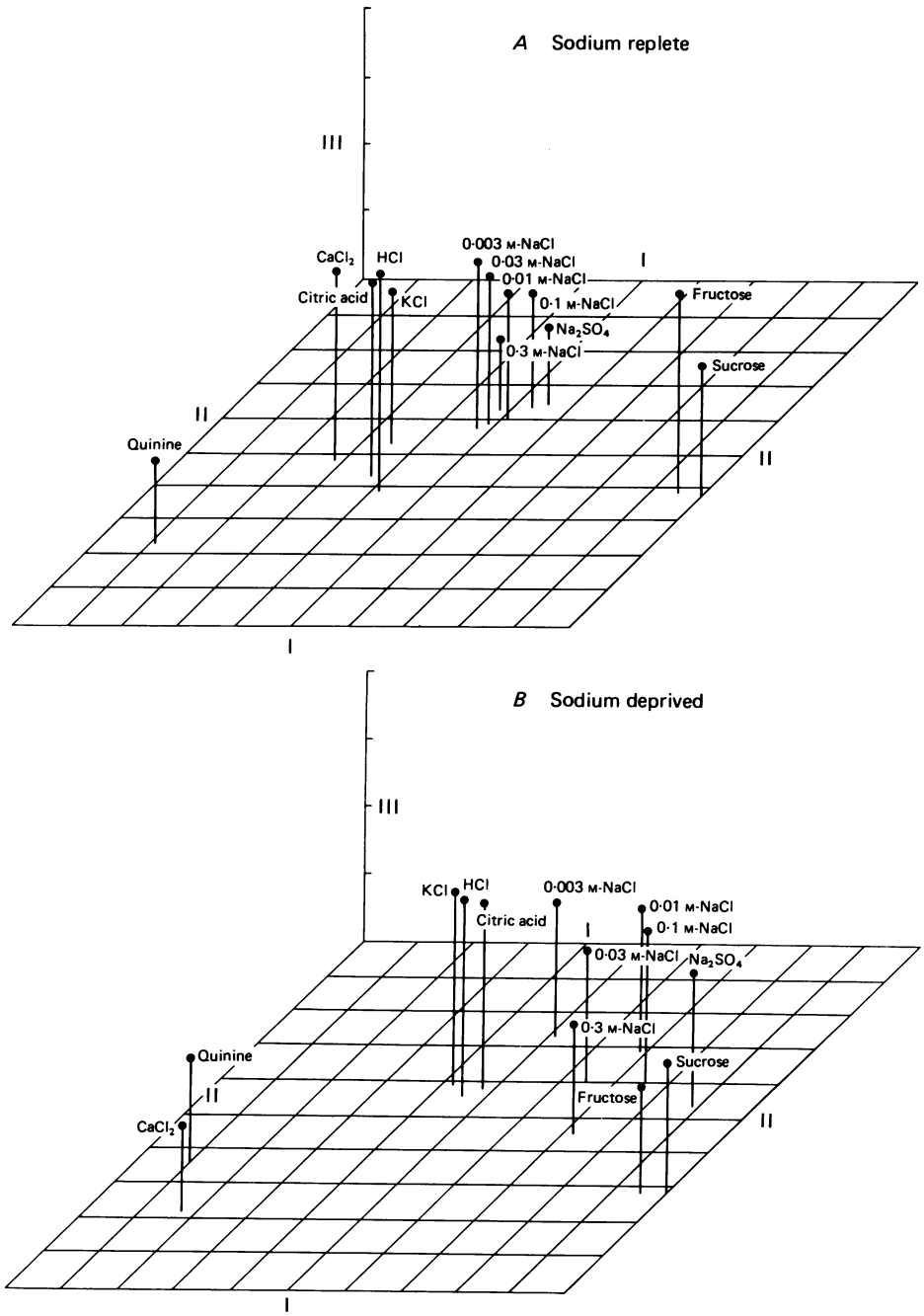

Fig. 8. Three-dimensional 'stimulus spaces' derived from interstimulus correlation coefficients calculated across 4-point profiles for each stimulus. Each point was generated by averaging evoked activity in four identifiable neurone types. The space derived from replete rats (*A*) is typical of a normally functioning taste system. A major division is between sweet and non-sweet stimuli. Within non-sweet compounds, distinctions are possible among salts, acids and quinine. The space representing stimulus quality in deprived subjects (*B*) differs in that sodium salts make a closer approach to sugars.

exemplary of a normally functioning taste system. There is a clear division between sweet and non-sweet stimuli; within non-sweets, distinctions are possible among sodium salts, acids and quinine representing salty, sour and bitter stimuli. The space representing taste quality in sodium-deprived rats (Fig. 8*B*) differs in that the sodium salts make a closer approach to the sugars, obscuring what is normally a clear distinction. The inference is that, with sodium depletion, the taste quality of sodium salts was more like that of the sugars used in this study.

DISCUSSION

The major results of this study are similar in some ways to those reported by Contreras (1977) and Contreras & Frank (1979) in recordings from the chorda tympani nerves of salt-deprived rats. Responsiveness to sodium decreased and activity evoked by simple sugars was greater in accord with those earlier findings. However the application of more comprehensive analytical procedures permits these changes to be interpreted according to the wider coding strategies employed by the rat's taste system.

The original interpretation from CT data held that taste quality was unaffected by salt deprivation, but that reduced responsiveness to sodium caused a shift in the intensity–hedonic curve such that higher concentrations fell on its appetitive limb and so were consumed, promoting repletion. Our interpretation of the NTS data emphasizes a change in quality rather than intensity. We suggest that sodium deprivation leads to a selective shift in responsiveness among taste cells such that those responsible for coding the most appetitive of taste qualities – sweetness – assume a greater load in signalling the presence of a much-needed chemical. This may imply that sodium tastes sweeter to rats when they are salt deprived. Alternatively, the concepts sweet, salty, sour and bitter may be human constructs while the rat may deal only with a dimension of appetite-aversion, bounded by sugars and by alkaloid toxins. Such a dimension has been implied by neural recordings (Scott & Mark, 1987). We suggest that the taste system is sufficiently compliant to the rat's physiological state that the chemical code for a desperately needed substance may be shifted to become more appetitive (as in these data), or, if a chemical proves toxic, to become more aversive (as in Chang & Scott, 1984*a*). By this scheme, sodium would not taste 'sweeter' to salt-deprived rats, only 'better'. A behavioural test of perceived qualitative similarity among the basic tastes would be instructive in verifying the increased similarity between sodium and sucrose in salt-deprived rats, but would not distinguish between these two theoretical alternatives: does sodium assume a sweet component, or is it simply more appetitive and so, on a dimension of appetite-aversion, closer to the sugars that define the appetitive extreme?

The interpretation of a shift in taste quality with sodium deprivation may better account for the consummatory behaviour that accompanies this condition. The behaviour of salt-deprived subjects presented with sodium is that of avid consumption normally associated with extremely palatable substances (Harriman, 1955; Berridge, Flynn, Schulkin & Grill, 1984). The same degree of avidity is not elicited by lower sodium concentrations in replete rats, suggesting more than a simple shift in perceived intensity.

Despite the finding of these effects that may offer a mechanism for repletion in the NTS of intact rats, decerebrate rats, with NTS intact, do not manifest a sodium appetite with depletion (Grill, Schulkin & Flynn, 1986). Thus the loss of communication with forebrain areas presumably interferes with the expression of these neural changes. This situation is analogous to that observed with conditioned taste aversions, where a neural effect is seen in the NTS, but decerebrate rats do not form the aversion.

Gustatory neurone types

In previous manuscripts we have adopted a neutral stance on the proposed existence of gustatory neurone types, saying that by loose definitions they may be identified, but that by strict definitions the notion largely fails (Scott & Chang, 1984; Scott, Yaxley, Sienkiewicz & Rolls, 1986*b*). This, however, relates to a system operating under normal conditions. An alternative approach to this issue is to alter physiological need – a factor that has been shown to affect taste responsiveness – and to determine whether the system changes as a whole or whether only subsets respond to the challenge, perhaps identifying functional neurone types. In the present experiment, an intense physiological need was created for sodium. When one group of neurones, identifiable *a priori* according to their response profiles, react in a selective and distinct manner in direct opposition to the behaviour of all cells that do not belong to the group, and when that action reveals a plausible hypothesis about the mechanism by which a phenomenon (avidity for sodium) may operate, we take that to be substantial support for the notion of gustatory neurone types.

The suggestion of types, however, does not imply that each is labelled for a specific basic chemical, such as sugar. Rather, salt deprivation caused formerly sugar-sensitive neurones to increase their involvement in signalling sodium. Thus, these are not labelled lines for the intrinsic substance sugar, though they may be for the normal intrinsic sequelae of sugar: 'sweetness' or 'goodness'. While it is not entirely clear how the apparent interplay of taste qualities in the CNS may be reconciled with the receptor mechanisms responsible for the transduction of sweeteners and sodium salts, there is evidence to suggest that the taste perceptions of these chemicals share a common sodium transport mechanism (Schiffman, Lockhead & Maes, 1983).

Several manipulations that influence food intake have been shown to affect the responsiveness of the taste system. These include gastric distension (Glenn & Erickson, 1976), hyperglycaemia (Giza & Scott, 1983), moderate hyperinsulinaemia (Giza & Scott, 1987) and the development of a conditioned taste aversion (Chang & Scott, 1984*a*). In the present study we report that sodium deprivation is associated with a shift in taste responsiveness to sodium salts that may lead to an afferent code with increased hedonic appeal. This extends the concept that the selection of appropriate foods under diverse conditions may be at least partially attributable to modifications in the very system that is largely responsible for those selections.

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