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AN ELECTROPHYSIOLOGICAL INVESTIGATION OF SUCCESSIVE CONTRAST IN THE GUSTATORY SYSTEM

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by

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ABSTRACT

AN ELECTROPHYSIOLOGICAL INVESTIGATION OF SUCCESSIVE CONTRAST IN THE GUSTATORY SYSTEM

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ROBERT SEIDENSTADT

An electrophysiological investigation was conducted with rats and hamsters to study the interaction between taste stimuli. Stimuli were applied to the tongue following a successive contrast paradigm. Three experiments were conducted: (a) to study the reliability of any interaction effect across preparations; (b) to study the effects of stimulus concentration and intervening water rinse duration upon the interaction effect; and (c) to study the interaction effect with sugars. The results showed that the interaction effect is a reliable phenomenon across preparations, that stimulus concentration and duration of the intervening water rinse were orderly variables that influenced the degree of interaction and that the interaction effect does apply across salt, acid and sweet stimuli. The interaction between stimuli was explained with a stimulus-receptor site bind model; the degree of interaction is dependent upon a stimulus' ability to form a bind with a particular receptor site. Implications of the results are discussed in terms of specificity - non-specificity mechanisms in the taste process and the nature of the stimulus domain for taste.

CHAPTER I

INTRODUCTION

A major assumption of gustatory theory is that the taste of any substance can be expressed as a function of four taste qualities; salt, sweet, sour and bitter (Beidler, 1962; Ogawa, Sato, and Yamashita, 1968). An equation for expressing this assumption is

$$T = aW + bX + cY + dZ$$

where T = taste substance

W,X,Y,Z = basic taste qualities

and a,b,c,d = coefficients.

The coefficients represent the degree of influence a basic taste quality has upon the taste substance in question.

There are several presuppositions associated with the basic taste equation. There is the idea that there are only four taste qualities and that these qualities are salt, sweet, sour and bitter. To date there is no firm evidence that supports this notion. Second, the basic taste equation is additive and therefore assumes that there is no interaction between the taste qualities in producing the taste associated with a substance. An implication of this additive model is that the receptor processes and the neural coding mechanism for the taste are specific. That is, there are four types of receptors in gustation, each associated with one of the basic taste qualities and that neurons associated with these receptors are quality specific. According to the taste equation, the neural coding mechanism for taste would involve various levels of activity in the four types of fibers dependent upon the taste substance. The central nervous system would then "analyze" this activity and, thus, taste perception. The general goal of the present research is to investigate the issue of interaction between taste stimuli and, hopefully,

relate this interaction to problems of gustatory theory.

In recent years, electrophysiological evidence has accumulated that challenges the specificity presupposition of the basic taste equation. Pfaffmann (1955) determined that chorda tympani fibers respond to more than one of the basic taste qualities and, on this basis, proposed the spectrum or across fiber pattern theory of neural coding for taste. The discrimination of taste quality is determined by the differential sensitivity of each fiber to different stimuli, i.e., Fibers A and B both respond to salt and sweet stimuli but Fiber A responds more vigorously to salt than Fiber B and, similarly, Fiber B responds more vigorourly than Fiber A to the sweet stimulus. The multisensitivity of chorda tympani fibers may be attributed to their innervation of the taste cell. Each taste cell is innervated by many fibers and each fiber branches to innervate several taste cells. The locus of specificity may be the taste cell and the multisensitivity of chorda tympani fibers simply reflects multiple innervation. However, Kimura and Beidler (1961) conducted a microelectrode study of taste cells of rats and hamsters. In both species, the taste cells demonstrated the property of multisensitivity; it responded to more than one of the basic taste qualities.

The cross-regeneration experiment conducted by Oakley (1967) also indicates the crucial importance of the taste cell in neural coding. The chorda tympani nerve innervates the anterior two-thirds of the tongue and primarily mediates the sensations of salt, sweet, and sour. The glossopharyngeal nerve (IX) innervates the posterior portion of the tongue and mediates the sensation of bitter. Oakley (1967) sectioned these nerves and then cross-regenerated them, i.e., the IXth

was sutured to the chorda tympani and the chorda tympani was sutured to the IXth. Electrophysiological experiments conducted after the regeneration process was complete showed that the IXth now mediated the sensation of salt, sweet and sour and that the chorda tympani mediated the sensation of bitter. This indicates that the different sensations mediated by these nerves are not due to any functional properties of these nerves but rather due to their anatomical relationship with the taste cells. It is the taste cell that is responsible for discriminating between taste stimuli.

McBurney (1969; 1972; McBurney & Lucas, 1966; Smith & McBurney, 1969; McBurney and Shick, 1971; McBurney, Smith & Shick, 1972) conducted a series of psychophysical investigations on the adaptation and cross-adaptation of taste stimuli. These experiments challenge the additivity presupposition of the basic taste equation. The experimental paradigm in McBurney's investigations requires the subject to estimate the quality and intensity of a taste substance under two conditions: (a) after adaptation to distilled water and (b) after adaptation to a taste stimulus. McBurney has reported that the adaptation effect does exist, that is, after adaptation to NaCl, the saltiness of NaCl is judged to be reduced compared with the judged saltiness of NaCl after adaptation to distilled water. McBurney has also conducted experiments that investigated cross-adaptation between stimuli, that is, whether adaptation to NaCl changes the judged saltiness of other salts, e.g., NaBr, KCl. McBurney determined that cross-adaptation existed not only for salt compounds (McBurney & Lucas, 1966) but also for sweet compounds (McBurney, 1972) and sour compounds (McBurney, et. al., 1972). In summary, McBurney's investigation indicate that taste substances do influence the perception of other taste substances, i.e., an interaction

or mutual influence exists between taste substances.

The adaptation and cross-adaptation effects are explained in terms of receptor processes. The presence of the adapting stimulus prevents the test stimulus from activating a receptor mechanism and, consequently, there is a reduction in the perceived intensity of the test stimulus. This conclusion has import for the neural coding mechanism of taste quality. If cross-adaptation did not exist then a separate receptor and coding mechanism is logically required for each taste stimulus. However, since the cross-adaptation effect does exist, we may assume receptor communality associated with taste substances. The extent of this receptor communality has not been determined.

Another approach to studying the interaction between taste stimuli has been developed by Hellekant (1968; 1969) who conducted electrophysiological experiments with a successive contrast design. The present research should be considered an extension of these experiments.

Before discussing these experiments, their logic must be made clear. The taste mechanism consists of taste cells that are located within the taste buds. The taste buds are found in papillae which are the projections on the surface of the tongue. Although the exact sequence of the taste transduction process is not fully known, it is believed to involve activation of receptor sites on the taste cell membrane by a stimulus. Activation of the receptor sites will produce depolarization of the taste cell membrane (Beidler & Gross, 1972). The depolarizations spread from the taste cell to the chorda tympani nerve. Thus activation, activation of the chorda tympani represents or parallels the activation of the taste cell receptor sites.

In Hellekant's (1968) first experiment, a water rinse was introduced between successive applications of the same stimulus. The duration of the water rinse was manipulated to study the time course of the interaction effect. The purpose of the water rinse was to remove any residue remaining from the first stimulation. The subjects in this experiment were cats. The amplitude of the integrated chorda tympani response was plotted against the duration of the water rinse, and this was defined as the receptor recovery function. The value, τ, was defined as that time period between stimulations when the second response was 2/3 of the maximum (original) response. Various values of are listed in Table 1.

Stimulus	τ (seconds)
0.016M HCl	6
0.15M NaCl	13
0.6M NaCl	6
0.1M KCl	5
0.3M K_2SO_4	5

Table 1. Values of the time constant, , for various stimuli. From Hellekant, 1968.

The results of this experiment indicate that two stimuli separated by a water rinse do interact with each other. This interaction may be characterized as a depression effect, i.e., the first stimulus depresses the response to the second stimulus. Hellekant (1968) determined that the time course of the depression effect varied between stimuli (see Table 1) but the effect was not influenced by the duration of the first stimulation (3 seconds or 10 seconds) nor by rate of the water rinse (1.7 ml/sec or 5.0 ml/sec). The latter fact indicates that the water rinse was successful in removing any stimulus residue and

thereby preserving the successive contrast design. The concentration of the stimulus did influence the depression effect (see Table 1 for NaCl): the stronger the stimulus concentration, the shorter the depression effect. That is, 0.6M NaCl exerted a weaker depression effect upon 0.6M NaCl than 0.15M NaCl exerted upon 0.15M NaCl. However, a problem in interpreting these results is that there were no comparisons across concentrations, i.e., 0.6M NaCl was never presented with 0.15M NaCl, and thus the terms weak or strong concentration are not entirely meaningful. One purpose of the present research is clarification of the relationship between the depression effect and stimulus concentration by comparing stimuli of different concentrations with each other.

In another investigation, Hellekant (1969) studied the interaction of different salts and acids in cats. A stimulus was applied to the tongue for 3 seconds followed by a 5 second water rinse and then application of a different stimulus for 3 seconds. Chorda tympani activity was monitored throughout the experiment with an integrated circuit. The measure of depression, if any, was the ratio of the response amplitude of the second stimulus to the response amplitude of this stimulus when there was no immediate prior stimulation of the tongue. Since each stimulus was paired with every other stimulus, the results of this study may be considered in terms of "ability to depress" and "susceptibility to depression". The most effective depressing stimuli were 0.3M Li_2SO_4, 0.3M K_2SO_4, and 0.01M QHCl (quinine hydrochloride) while the least effective depressors were 0.3M NaCl and 0.05M HAc (acetic acid). The stimuli most readily depressed were 0.1M $MgCl_2$, 0.3M NaCl, and 0.3M LiCl while the stimuli least susceptible to depression were

0.05M HAc, 0.016M HCl, and 0.3M K_2SO_4. (In fact, of the 27 times K_2SO_4 was the second stimulus, depression occurred 10 times, no change occurred 3 times and an increase in response occurred 14 times.) It should be noted that the concentrations of the stimuli were fixed rather than sampled across a range of concentrations. Hellekant (1969) observed that there were three types of depression effects. Adopting his notation, we have 'a' and 'b' as stimuli and ↓ as depresses. The various effects observed by Hellekant (1969) with different combinations of stimuli can be expressed as follows:

1. $a\downarrow a > a\downarrow b$ (A stimulus depresses itself more than it depresses another stimulus.)
2. $a\downarrow b < b\downarrow a$
3. $a\downarrow a < b\downarrow a$

Hellekant (1969) attempts to explain these effects in terms of Beidler's (1954) theory of taste stimulation which states, in part, that taste stimulation is a function of a binding process between the stimulus and receptor sites. Hellekant (1969) interprets the first effect as a reflection of stimulus similarity. Assume that receptor sites correspond to a particular set or class of stimuli and that differences between receptor sites are organized, i.e., arranged in some sequential or dimensional order. Any stimulus that depresses the response to another stimulus must be able to compete for the receptor site of the depressed stimulus. The degree of depression is an indication of similarity between receptor sites and, therefore, an indication of similarity between stimuli. Hellekant (1969) offers support for his statements by noting the general agreement between his results and those of Erickson, Doetsch, and Marshall (1965). Erickson et. al. (1965) developed a method for measuring the

similarity between stimuli on the basis of the responsiveness of single chorda tympani fibers. The second effect can also be interpreted within the framework of receptor sites. For this effect, we assume that the receptive field for Stimulus A is smaller than the receptive field for Stimulus B. A receptive field is the total collection of receptor sites to which a taste stimulus may bind. Furthermore, it is assumed that A and B share some receptor sites; any sharing between A and B will have a greater effect upon A than upon B. The third effect, a a b a, states that a stimulus is more depressed by another stimulus than by itself. Hellekant (1969) suggests that this effect may be due to lateral inhibition; one stimulus acts upon the receptor sites of another stimulus via a nerve influence and thereby prevents a response to this second stimulus.

Hellekant (1968; 1969) did not extend the interpretation of his results beyond blocking of receptor sites; no implications are given for the basic taste qualities as the stimulus domain for taste nor the related problem of neural coding of taste quality. Aside from this lack of interpretation, there are two serious methodological errors in Hellekant's experiments. First, all stimulus combinations were not presented to all Ss. It is possible that the length of the testing procedure and the consequent condition of the chorda tympani nerve made a complete design prohibitive. In order to evaluate any differences between preparations, Hellekant (1969) performed a sign test. This result showed a significant ($p = 0.05$) difference between preparations for the same stimuli. This, of course, raises the question of the degree of contribution of individual differences between preparations to the depression effect. The six most effective depressing stimuli were tested

in only four of the seven preparations. A weak depression effect for all stimuli was obtained in the other three preparations. Another methodological flaw in Hellekant's (1968; 1969) work is an incorrect sequence of stimulus presentations. In the case, the effect of Stimulus A upon Stimulus B, the standard or control condition for Stimulus B was obtained after the A-B presentation. In this sequence the possibility that Stimulus A may have had an effect upon the Stimulus B control measurement exists. A sounder methodological procedure would be to obtain the control measurements of Stimulus B before and after the A-B presentation. In summary, the major contributions of Hellekant (1968; 1969) are a method for studying the interaction between taste stimuli and the demonstration that, under some circumstances this interaction exists as a depression effect.

The purpose of the present research are to establish the reliability of the depression effect, to establish the effect of stimulus concentration upon the depression effect, to examine the effect with another class of stimuli, the sugars, and to generalize the effect to another species, the rat. If the successive contrast design does prove to be a reliable technique, then another method is available for studying the gustatory system. Specifically, the import of this technique is that it is a method to study the nature and characteristics of receptor sites.

Experiment I: Reliability of the Depression Effect

The major inadequacy of Hellekant's work, that all stimulus combinations were not presented to all preparations, has been discussed. It is not possible to eliminate individual differences between preparations, but it is possible to reduce the confounding effects of individual differences by presenting all stimulus combinations to each S. This can be

accomplished by reducing the number of stimuli employed in any given experiment. The stimuli in this experiment were 0.3M K_2SO_4, 0.3M LiCl, 0.3M NaCl and 0.05M HAc. These stimuli may be arranged in the following classification system:

		Depression Ability	
		Strong	Weak
Depression Susceptibility	Yes	0.3M LiCl	0.3M NaCl
	No	0.3M K_2SO_4	0.05M HAc

This classification system is based upon data provided by Hellekant (1969). It should be noted that each stimulus was ranked at or near the extreme of its classificatory cell with the exception of 0.3M LiCl. This stimulus was ranked as the fifth most effective depressor and the stimulus third most susceptible to depression; thirteen stimuli were employed by Hellekant (1969). If it is assumed that the depression effect between stimuli is a reliable phenomenon and, further, that no species differences exist between the cat and rat with respect to gustation, then the following predictions can be made:

1. The strongest depression effect will be exerted by 0.3M K_2SO_4 and 0.3M LiCl.
2. The weakest depression effects will be exerted by 0.3M NaCl and 0.05M HAc.
3. The stimuli most likely to be depressed are 0.3M NaCl and 0.3M LiCl.
4. The stimuli least likely to be depressed are 0.3M K_2SO_4 and 0.05M HAc.

Experiment II: The Effect of Stimulus Concentration and Intervening Water Rinse Duration Upon the Depression Effect.

The effect of changing the concentrations of the test stimuli and the consequent effects have already been presented. It should be re-emphasized that Hellekant (1968) varied the concentrations of the test stimuli simultaneously. In the present experiment the concentrations of the test stimuli was varied successively, and this allowed development of a parameter that describes the relationship between stimulus concentration and the depression effect. Manipulation of the water rinse duration provides another variable that may possibly describe the depression effect. If the depression effect is a viable phenomenon, and if it can be related to binding of a stimulus to a receptor site, then the following predictions can be made:

1. As the water rinse interval increases between Stimulus A A and Stimulus B, the degree of depression between these stimuli will decrease. The obvious rationale for this prediction is that the longer water rinse will be more able to break the bind between the stimulus and the receptor site than the shorter water rinse. Once the receptor site is free, it will be able to accept another stimulus.
2. a) As the concentration of a stimulus producing an effect increases, the degree of the depression effect will increase. The rationale in this case is that a stimulus at concentration (X) will occupy more receptor sites than a stimulus at concentration (X - N) and, consequently, the stimulus at the stronger concentration will produce a greater depression effect.

b) As the concentration of a stimulus that is being effected upon increases, the degree of depression exerted by a stimulus at constant concentration will decrease. It is felt that the stronger stimulus will attempt to occupy more receptor sites than the weak stimulus and thereby be less susceptible to depression.

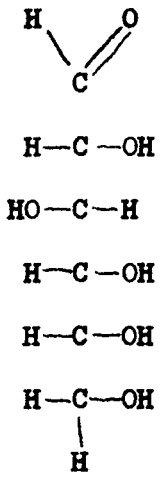
Experiment III: Sugars and the Depression Effect.

In order to have a comprehensive description of the depression effect it is essential that the effect be studied with as many different stimuli as possible. In Hellekant's (1968; 1969) experiment the sweet taste was not investigated. The reason for this omission is that the cat does not respond well to sugars. The purpose of the present experiment is to achieve a description of the interaction between taste stimuli when these stimuli are sugars. This goal was accomplished by studying the depression effect in rats and hamsters. Two species are used because the rat provides continuity with Experiment I and II, and hamsters were studied because they are more responsive to sugar than the rats.

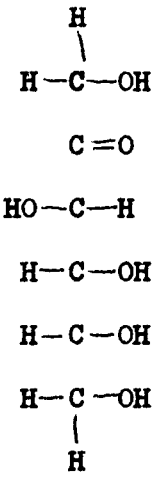
Previous investigations with sugar have shown that effective stimuli are sucrose, glucose and maltose for the rat (Hagstrom and Pfaffmann, 1959); 0.5M fructose, 0.5M sucrose and 0.5M glucose for the dog (Anderson, Funakoshi and Zotterman, 1963); and 0.5M sucrose for hamsters (Beidler, et. al., 1955). On the basis of these results and preliminary testing, 1.0M fructose and 1.0M sucrose were employed as sugar stimuli in the rat experiment. Since one purpose of the present series of experiments was to explore the interrelationships between

stimuli and classes of stimuli, it would be profitable to employ non-sugar taste stimuli. To this end, 0.05M NaCl and 0.05M HAc are also stimuli in this experiment. Glucose was not used with the rats because preliminary testing indicated that the chorda tympani response to glucose was too weak to allow reliable measurement, especially in the case where the response to glucose may have been depressed. The stimuli employed with hamsters are 1.0M fructose, 1.0M sucrose, 1.0M glucose and 0.40M NaCl. It would have been desirable to include 0.05M HAc as a stimulus in this experiment but that would have increased the number of comparisons between stimuli from 16 to 25. This represents a 90 minute increase in experimental testing time which would have jeopardized the possibility of completing the entire set of comparisons with any given preparation.

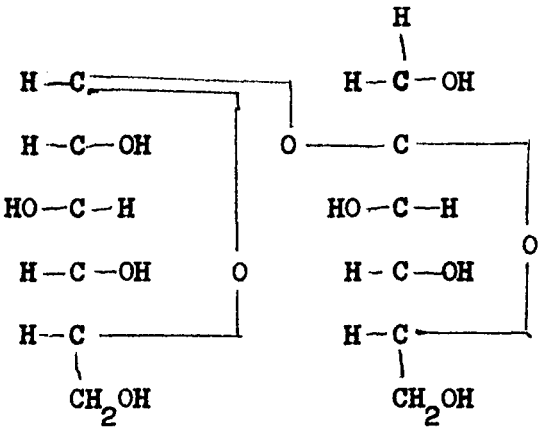
Several predictions can be made about the possible depression effect between sugars based upon the molecular structure of these substances:



D - Glucose



D - Fructose



Sucrose

It can be seen that the sucrose molecule is a combination of the glucose molecule and the fructose molecule. If it can be assumed that the structure of a molecule is related to its binding to a receptor site, it can be predicted that sucrose will have a greater depression effect upon fructose and glucose than these substances would have upon each other. In addition, sucrose will have a greater depression effect upon fructose and glucose than these substances will have upon sucrose. These predictions reflect the possibility that sucrose can theoretically bind with a fructose and glucose receptor site but that these latter substances may not be able to bind with a sucrose receptor site.

CHAPTER II

METHODS

I. General

Subjects. The Ss in Experiment I and II were female albino rats 150-300 grams. The Ss in Experiment III were female albino rats, 150-225 grams, and hamsters, 100-125 grams.

Procedure. Ss were anesthetized with sodium pentobarbital (Nembutal, 60 mg/ml). Doses were 65 mg/kg for the rat and 80 mg/kg for the hamster; supplemental doses of 25 mg/kg were administered to the hamster. The trachea was cannulated and the Ss head was placed in a holder that allowed access to the Ss tongue. The left chorda tympani nerve was exposed from its exit from the lingual nerve until its passage into the bulla region. The nerve was cut and placed upon a wick Ag-AgCl electrode which led to a Tektronix 532 oscilloscope through a Grass P511G amplifier. The output from the oscilloscope led to an audio amplifier and through an integrated circuit (rise time = 0.11 seconds, fall time = 0.025 seconds) to a Sanborn recorder (Figure 1).

Stimuli were presented through a flow system that included three cylindrical separatory funnels (two at 125 ml, one at 500 ml) and a flow chamber that fitted securely over the anterior portion of the Ss tongue (Figure 2). This system allowed a rapid change from one stimulus to another stimulus, prevented the influence of saliva from modifying the taste response and assured that, in a given preparation, the same area of the tongue was stimulated throughout the experiment. Solutions were prepared from reagent grade chemicals with the exception of sucrose which was of commercial variety; distilled water was used in all phases of this research.

Figure 1. Diagram of the recording apparatus. The preparation including the stimulus funnels and the amplifier were enclosed in an aluminum shielded cage,

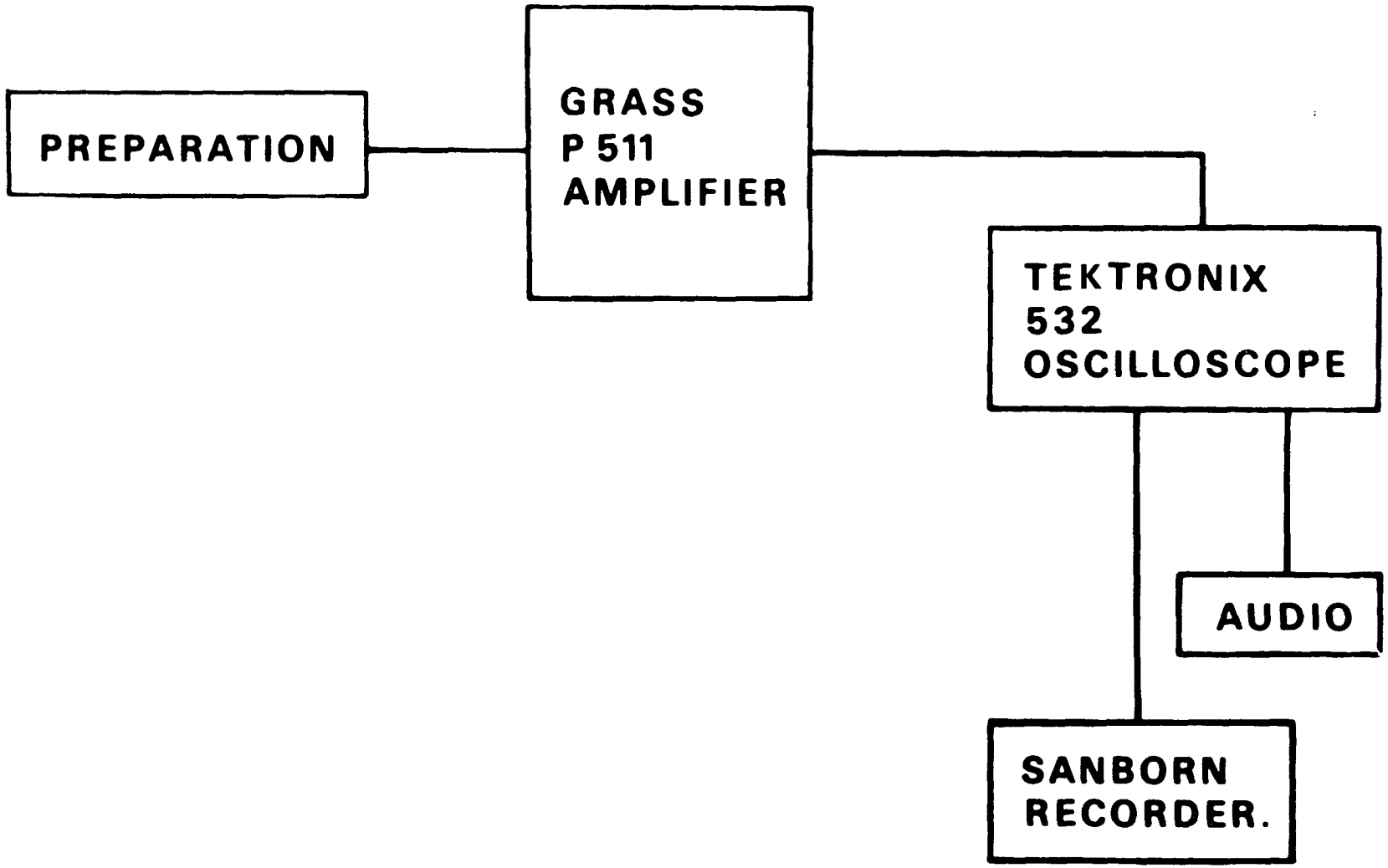
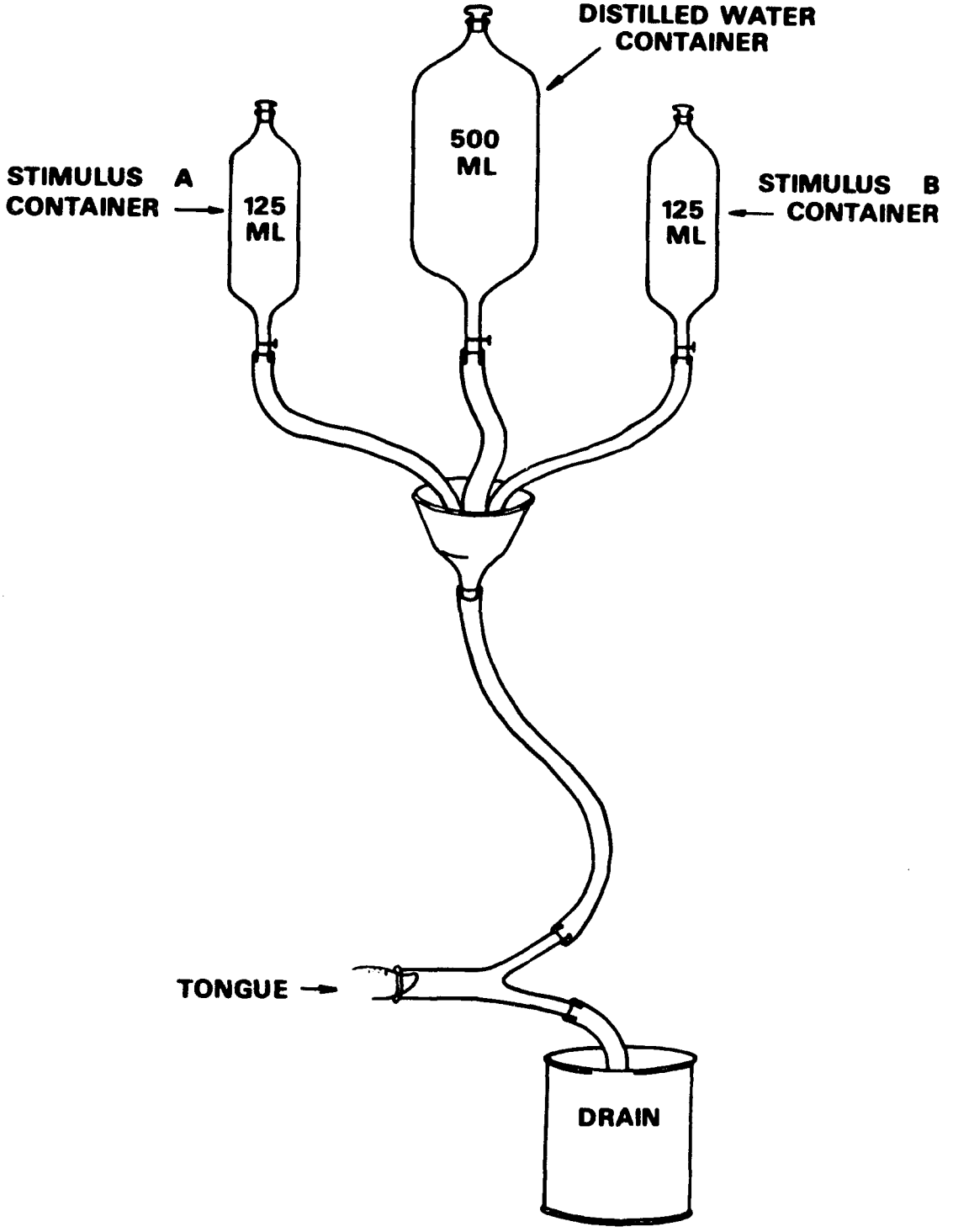

PREPARATION
GRASS
P 511
AMPLIFIER
TEKTRONIX
532
OSCILLOSCOPE
AUDIO
SANBORN
RECORDER.

Figure 2. Arrangement of the stimulation system that provided a method to control the flow of taste solutions over the tongue.


DISTILLED WATER
CONTAINER
500
ML
STIMULUS A
CONTAINER
125
ML
125
ML
STIMULUS B
CONTAINER
TONGUE
DRAIN

2. Experiment I: Reliability of the Depression Effect.

The stimuli used in this experiment were 0.3M K_2SO_4, 0.3M NaCl, 0.3M NaCl, and 0.05M HAc. To test the effect of Stimulus A upon Stimulus B, the following sequence of stimulus presentations was used:

1. Stimulus B - 3 seconds
2. Water rinse - 60 seconds
3. Stimulus A - 3 seconds
4. Water rinse - 5 seconds
5. Stimulus B - 3 seconds
6. Water rinse - 60 seconds
7. Stimulus B - 3 seconds

The above sequence was repeated twice for each of the sixteen stimulus pairs in this experiment.

3. Experiment II: Depression as a Function of Stimulus Concentration and Duration of the Intervening Water Rinse.

The aim of this experiment was to examine the effects of two variables, stimulus concentration and duration of the intervening water rinse upon the depression effect. The stimuli employed in this experiment were 0.3M LiCl, 0.05M HAc and four concentrations of NaCl, 0.05M, 0.10M, 0.20M, and 0.40M. Four sets of comparisons were made with these stimuli: (1) the effect of 0.3M LiCl upon the NaCl concentration; (2) the effect of 0.05M HAc upon the NaCl concentrations; (3) the effect of the NaCl concentrations upon 0.3M LiCl; and (4) the effect of the NaCl concentrations upon 0.05M HAc. Each comparison within a set was made twice at three water rinse intervals: 3, 5, and 10 seconds. Due to the length of the testing procedure it was not possible to test all four sets of comparisons with each preparation. However, any given set of comparisons was

completed on the same preparation.

4. Experiment III: Sugars and the Depression Effect.

Stimuli in this experiment when rats were used as Ss were 0.05M NaCl, 0.05M HAc, 1.0M fructose and 1.0M sucrose. In the hamster portion of this experiment the stimuli employed were 1.0M fructose, 1.0M sucrose, 1.0M glucose, and 0.40M NaCl. The sequence of stimulus presentations in this experiment followed the general plan outlined for Experiment I.

CHAPTER III

RESULTS

The responsiveness of the chorda tympani nerve to the various taste solutions was obtained through an integrated circuit. In order to have consistency in measuring the integrated records across the different preparations, a ratio procedure was used to interpret these records. The responsiveness (maximum displacement from baseline) to Stimulus B after Stimulus A was compared with the responsiveness to Stimulus B after the 60 second water rinse. This comparison is expressed in the following ratio:

$$\frac{\text{Stimulus B after Stimulus A}}{\text{Stimulus B after 60 second water rinse}} \times 100 = \text{Effect of Stimulus A upon Stimulus B}$$

If this ratio is equal to 100, then Stimulus A does not have an effect upon Stimulus B. If this ratio is greater than 100, then Stimulus A potentiated the response to Stimulus B; if the ratio is less than 100, then Stimulus A depressed the response to Stimulus B.

Experiment I: Reliability of the Depression Effect.

The purpose of this experiment was to examine the reliability and existence of an interaction between taste stimuli when all stimulus pairs are presented to all Ss. A measure of reliability across Ss can be obtained with analysis of variance procedures. A one-way, repeated measures analysis of variance was applied to the data by considering each stimulus pair combination as a different treatment condition. This analysis (Table 2) revealed a significant treatment effect ($F = 7.72$, p 0.001). The variation attributable to differences between Ss was

Figure 3. Sample integrated records from the chorda tympani nerve from three different preparations. Time base, 1 cm = 20 seconds.

a. Stimulus A - 0.3M K_2SO_4

   Stimulus B - 0.3M NaCl

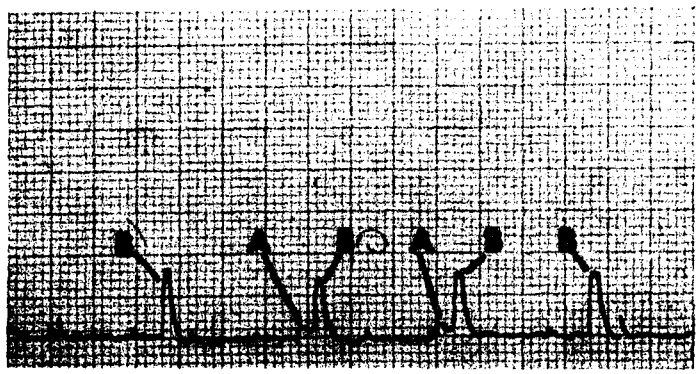
b. Stimulus A - 1.0M Fructose

   Stimulus B - 0.05M HAc

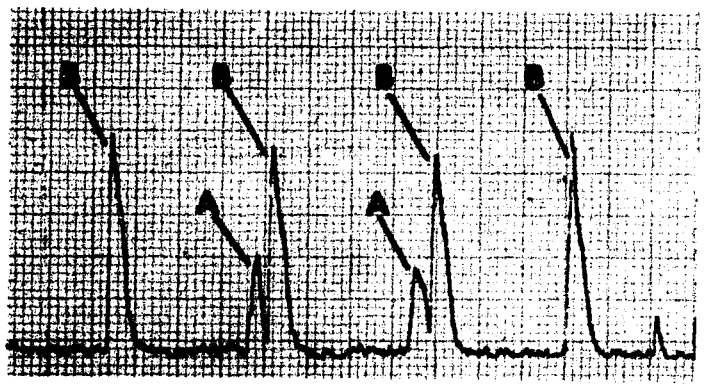
c. Stimulus A - 0.2M NaCl

   Stimulus B - 0.05M HAc, 10 second water rinse.

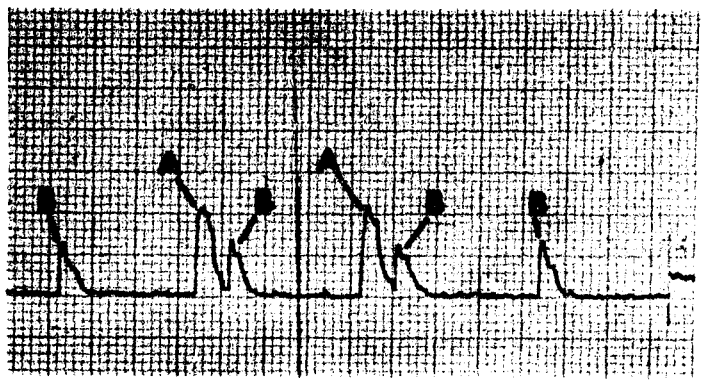
A



B



C



Source	SS	df	MSq	F
Between	3533.02	6		
Within	26384.25	96		
Treatment	15530.67	15	1035.37	7.72 ***
Residual	10853.58	81	133.99	
Total	29917.27			

*** $p < 0.001$

Table 2. Analysis of variance summary table for the case where each stimulus pair is treated as a different treatment condition: Experiment I.

only 12% of the total variation while 88% of the total variation is attributable to within groups differences. This indicates that individual differences between Ss did not markedly influence any effect that may occur when stimuli are presented to the tongue in a successive contrast paradigm. Further partitioning of the within groups variation shows that the treatment effect accounted for 53% of the total variation and that the remaining 35% of the total variation was error variation. Although the treatment effect was statistically significant, the error variation may be considered large and, therefore, deserves comment. One possible source of error in this experiment may be attributed to the measurement of the responses to 0.3M K_2SO_4. This substance produced the weakest integrated response of the four stimuli employed in this experiment and, therefore, any fluctuations in the response to 0.3M K_2SO_4 would produce greater variability in measurement than fluctuations in the response to any of the other stimuli. For example, a response to 0.3M LiCl or 0.3M NaCl may have been 20 units (arbitrary scale) while the response to 0.3M K_2SO_4 is 5 units. A one unit change to either 0.3M LiCl or 0.3M NaCl would represent a 5% change in responsiveness whereas, this one unit change in responsiveness to 0.3M K_2SO_4 would represent a 20% change in responsiveness. Thus, the normal expected changes in responsiveness had a greater effect upon 0.3M K_2SO_4 than upon 0.3M LiCl or 0.3M NaCl. The stimulus 0.05M HAc exhibited a degree of responsiveness intermediate between the 0.3M LiCl-0.3M NaCl group and 0.3M K_2SO_4. Since the error variability in this experiment was large, it was decided to construct confidence intervals about the mean for each of the 16 stimulus pairs (Table 3). This procedure was used to reliably note the existence of a depression effect between stimuli. If the upper confidence

limit was less than 100 (indication of no effect), it was concluded that a statistically significant depression effect existed. Similarly, if the upper confidence limit was greater than 100, it was concluded that a depression effect did not exist for this stimulus pair. (The case of a potentiation effect between two stimuli did not occur in this experiment. It would have been tested by comparing the lower confidence limit of the mean in question with 100.) On this basis, the stimulus that produced the strongest depression effect was 0.3M K_2SO_4, depressing every stimulus except 0.3M LiCl. The other stimuli all depressed 0.05M HAc and 0.3M K_2SO_4; they did not produce a depression effect upon 0.3M LiCl or 0.3M NaCl. The stimuli 0.3M K_2SO_4 and 0.05M HAc were depressed by all stimuli; 0.3M NaCl was depressed only by 0.3M K_2SO_4 and 0.3M LiCl was not depressed by any of the stimuli. Examination of these results with respect to the predictions based upon Hellekant's (1969) results shows large discrepancies between the two sets of data. It was predicted that 0.3M K_2SO_4 and 0.3M LiCl would exert the strongest depression effects. The prediction holds for 0.3M K_2SO_4 but not 0.3M LiCl which exerted a relatively moderate depression effect. The second prediction stated that 0.3M NaCl and 0.05M HAc would exert weak depression effects. The results do not provide clear support for this prediction; 0.05M HAc exerted the weakest depression effect whereas, 0.3M NaCl exerted a strong depression effect, especially upon 0.05M HAc and 0.3M K_2SO_4. The third and fourth predictions are completely negated by the results. Together, they stated that the stimuli most likely to be depressed are 0.3M NaCl and 0.3M LiCl and the stimuli least likely to be depressed are 0.3M K_2SO_4 and 0.05M HAc. The present results show the exact

		Stimulus A				
		NaCl	LiCl	HAc	K_2SO_4	
Stimulus B	NaCl	$\bar{X}$ = 87.3 (106)	92.07 (111)	93.07 (112)	79.81 (98.8)	Mean 88.06
	LiCl	91.17 (110)	96.0 (114)	96.42 (115.41)	86.76 (105)	92.59
	HAc	63.85 (82)	73.64 (92)	76.5 (95)	78.35 (97)	78.07
	K_2SO_4	62.42 (81.41)	63.92 (82.91)	79.57 (98.56)	63.57 (82.56)	67.37
	Mean	76.18	81.40	86.39	77.12	

Table 3. Means and, in parenthesis, the upper bound of the 99% confidence interval for each of the stimulus combinations. To be read, the effect of Stimulus A upon Stimulus B.

opposite findings; 0.3M K_2SO_4 and 0.05M HAc were the most easily depressed stimuli while 0.3M NaCl was depressed only by 0.3M K_2SO_4 and 0.3M LiCl was not depressed by any stimulus. On the basis of these results it may be concluded that the interaction of taste stimuli does, in some cases, result in a depressed response to the second stimulus and that this interaction is not profoundly influenced by individual differenced between Ss. The largest discrepancies between the results of the present experiment and Hellekant's (1969) experiment may possibly be due to species differences. This possibility will be discussed in Chapter IV.

In the present experiment it is possible to assign two properties to each stimulus; its ability to produce a depression effect and a stimulus' susceptibility to depression by another stimulus. Graphic presentation of the data in terms of effect productivity and effect susceptibility is given in Figure 4 and Figure 5 respectively. It is clear that the four stimuli employed in this experiment did not differ considerably with respect to effect productivity but that clear differences are apparent with respect to effect susceptibility. In order to determine if these differences are statistically meaningful, a two way analysis of variance was performed with effect productivity as one factor (Stimulus A in Table 3) and effect susceptibility as the second factor (Stimulus B in Table 3). Significance was obtained for the production factor ($F = 4.06$, p 0.01) and the susceptibility factor ($F = 26.81$, p 0.001); the production X susceptibility interaction was not statistically significant (Table 4). The results of the

Newman-Keuls procedure to possibly identify the source of these differences by making individual comparisons between the means within each factor are presented in Table 5. There were no significant differences between stimuli with respect to effect productivity, but with respect to effect susceptibility, three significant differences were obtained. Both 0.3M LiCl and 0.3M NaCl were significantly less susceptible to depression than 0.3M K_2SO_4 ($p < 0.01$), and 0.3M LiCl was less susceptible to depression than 0.05M HAc ($p < 0.05$). The results of this analysis indicate that it is possible to significantly differentiate between certain stimuli in their susceptibility to depression but there are no statistically significant differences between stimuli in their ability to produce a depression effect, i.e., the strongest depresser, 0.3M NaCl produces an effect that is not significantly different from the effect produced by the weakest depresser, 0.05M HAc.

Experiment II: The Effect of Stimulus Concentration and Intervening Water Rinse Duration Upon the Interaction Between Stimuli.

The analysis of results for this experiment followed the same form as in the analysis for Experiment I. First, confidence limits were constructed about the stimulus pair means to identify the location(s) of an interaction effect and then analysis of variance procedures, including a Newman-Keuls analysis, were used to determine the influence of the variables in question upon the interaction between stimuli.

Mean and the upper 99% confidence limit for the stimulus pairs in each of the four sections of this experiment: (1) the effect of 0.3M LiCl upon NaCl concentrations; (2) the effect of HAc upon NaCl at

Figure 4. The ability of each stimulus to produce a depression effect; Experiment I. To be read, the effect of Stimulus A upon Stimulus B.

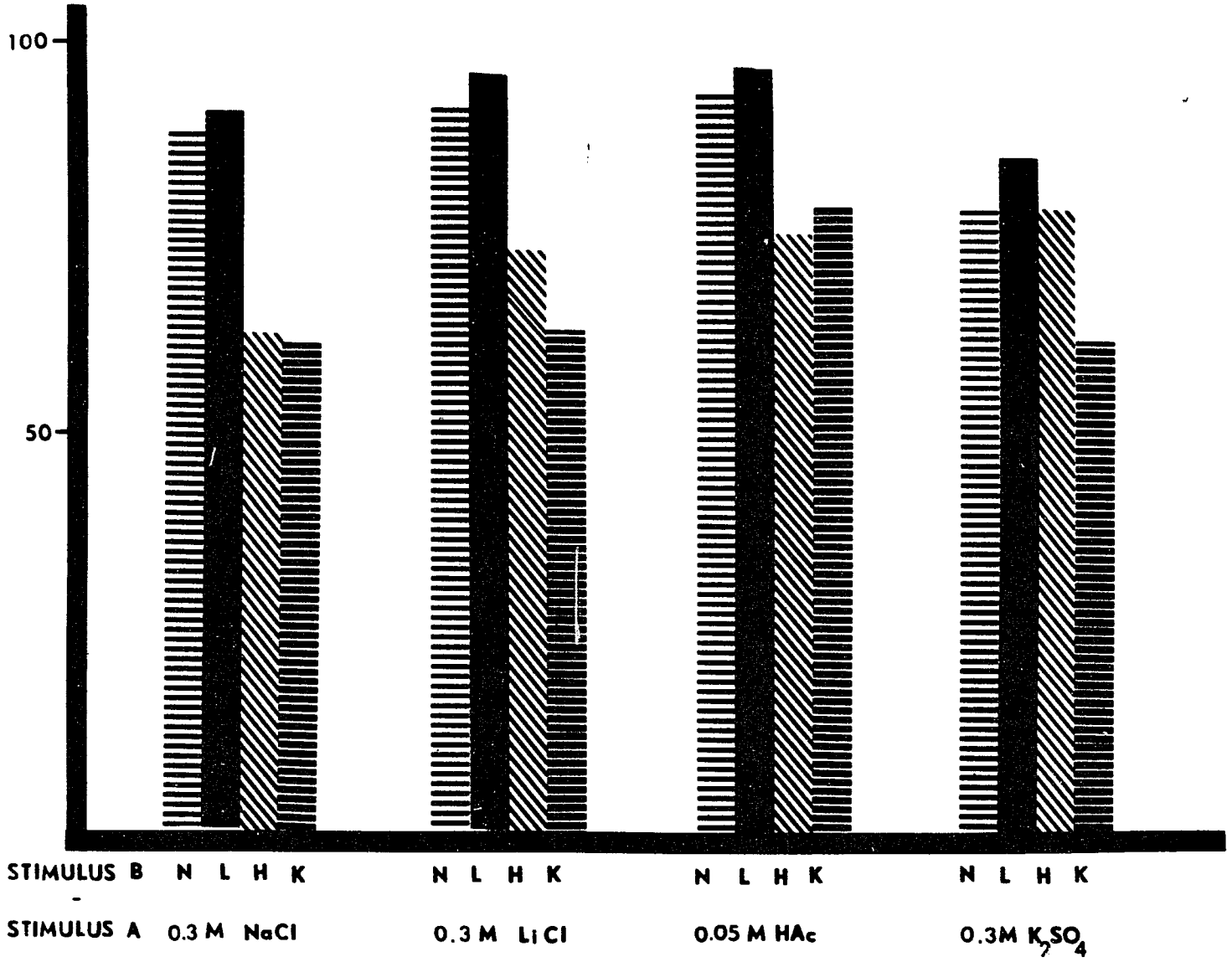
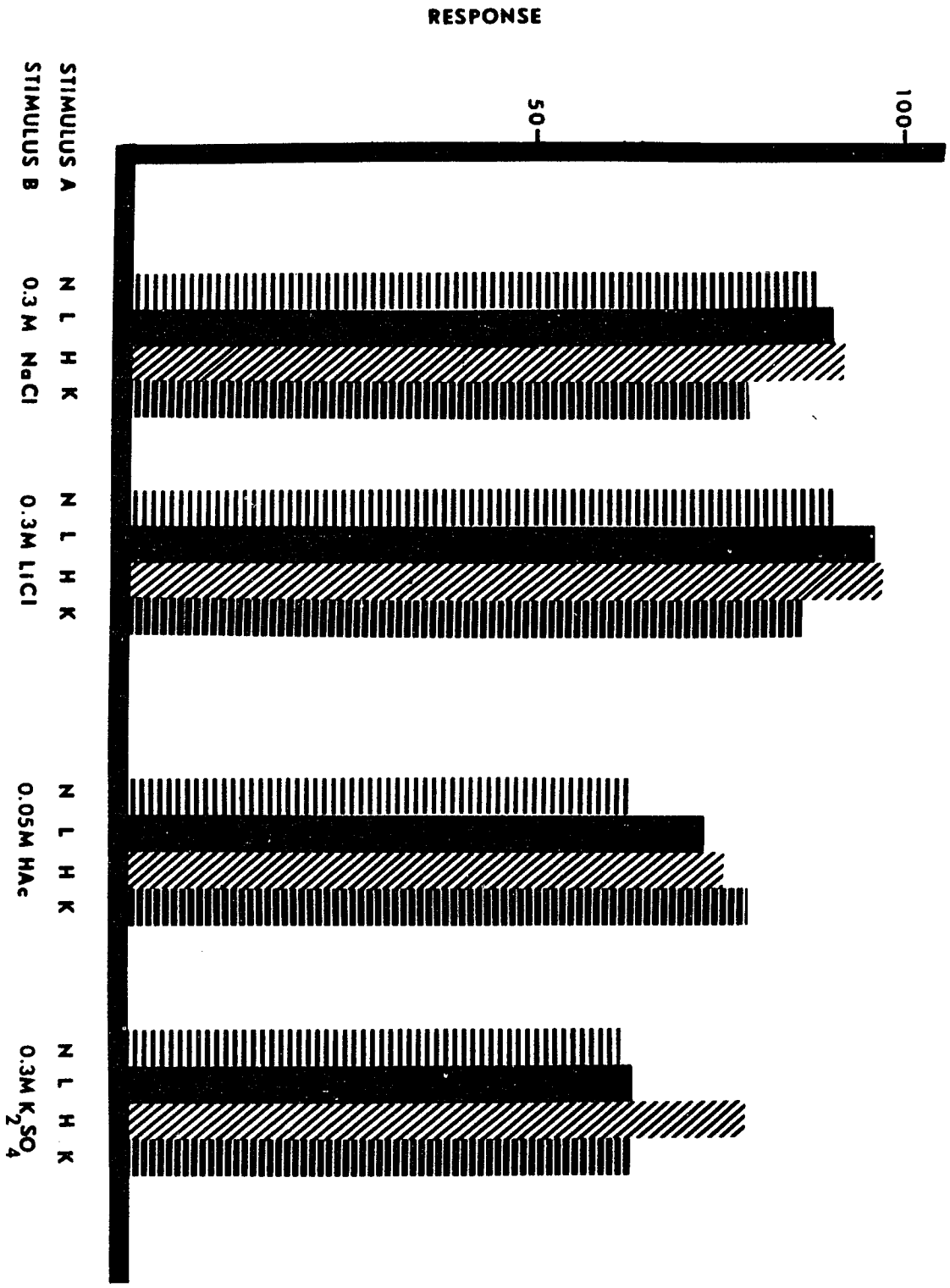

100
50
STIMULUS B N L H K N L H K N L H K N L H K
STIMULUS A 0.3 M NaCl 0.3 M LiCl 0.05 M HAc 0.3M K_2SO_4

Figure 5. The susceptibility of each stimulus to depression by another stimulus; Experiment I. To be read, the effect of Stimulus A upon Stimulus B.


RESPONSE
50
100
STIMULUS A
STIMULUS B
N L H K
0.3 M NaCl
N L H K
0.3M LiCl
N L H K
0.05M HAc
N L H K
0.3M K_2SO_4

Source	SS	df	MSq	F
Productivity	1827.26	3	609.08	4.06 **
Susceptibility	12056.54	3	4018.84	26.81 ***
Prod. X Susc.	1646.87	9	182.98	1.22
Error	14386.64	96	149.86	

** $p < 0.01$

*** $p < 0.001$

Table 4. Analysis of variance summary table for effect productivity and effect susceptibility.

a.

	NaCl	K_2SO_4	LiCl	HAc
NaCl	-	NS	NS	NS
K_2SO_4		-	NS	NS
LiCl			-	NS

b.

	K_2SO_4	HAc	NaCl	LiCl
K_2SO_4	-	NS	**	**
HAc		-	NS	*
NaCl			-	NS
LiCl				

* $p < 0.05$

** $p < 0.01$

NS Not significant

Table 5. Newman-Keuls analysis for individual comparisons between stimuli for a. effect productivity and b. effect susceptibility.

various concentrations; (3) the effect of various NaCl concentrations upon 0.3M LiCl; and (4) the effect of various NaCl concentrations upon 0.05M HAc are presented in Table 6 and Table 7 (see also Figure 6 and Figure 7). Both 0.3M LiCl and 0.05M HAc had a significant depression effect upon 0.05M NaCl and 0.10M NaCl at the 3 seconds and the 5 seconds water rinse interval. At 0.4M NaCl, 0.3M LiCl produced a significant depression effect at the 3 seconds water rinse interval. The ability of NaCl to significantly depress 0.3M LiCl occurred only at 0.4M NaCl with a 3 seconds water rinse. However, all concentrations of NaCl were able to produce a significant depression effect upon 0.05M HAc at the 3 seconds and 5 seconds water rinse intervals. No significant depression effect of HAc occurred at the 10 seconds water rinse interval.

In summary, stimulus concentration and water rinse duration can both profoundly influence the interaction between taste stimuli. However, the degree of their influence is ultimately dependent upon the specific stimuli in question.

a.

		NaCl Concentration			
		0.05M	0.10M	0.20M	0.40M
Intervening Water Rinse Duration (Seconds).	3	24.2 (47.3)	33.0 (56.1)	66.3 (89.4)	64.9 (68.0)
	5	50.24 (73.37)	76.1 (99.2)	91.0 (114.1)	91.7 (114.8)
	10	83.7 (106.8)	89.5 (112.6)	105.7 (128.8)	108.8 (131.9)

b.

		NaCl Concentration			
		0.05M	0.10M	0.20M	0.40M
Intervening Water Rinse Duration (Seconds).	3	49.9 (72.0)	65.2 (87.32)	74.5 (96.62)	79.2 (101.32)
	5	65.8 (87.9)	71.6 (93.7)	82.6 (104.7)	83.7 (105.8)
	10	84.7 (106.8)	91.9 (114.2)	92.6 (114.72)	101.9 (123.0)

Table 6a. Means and, in parenthesis, the upper bound of the 99% confidence interval, for the effect of 0.3M LiCl upon NaCl as a function of NaCl concentration and intervening water rinse duration.

Table 6b. Means and, in parenthesis, the upper bound of the 99% confidence interval, for the effect of 0.05M HAc upon NaCl as a function of NaCl concentration and intervening water rinse duration.

a.

		NaCl Concentration			
		0.05M	0.10M	0.20M	0.40M
Intervening Water Rinse Duration (Seconds).	3	88.5 (115.6)	89.5 (116.6)	77.7 (104.8)	68.5 (95.6)
	5	100.0 (127.1)	102.0 (129.1)	90.0 (117.1)	77.5 (107.6)
	10	104.9 (131.9)	104.0 (131.1)	104.3 (131.4)	107.6 (134.7)

b.

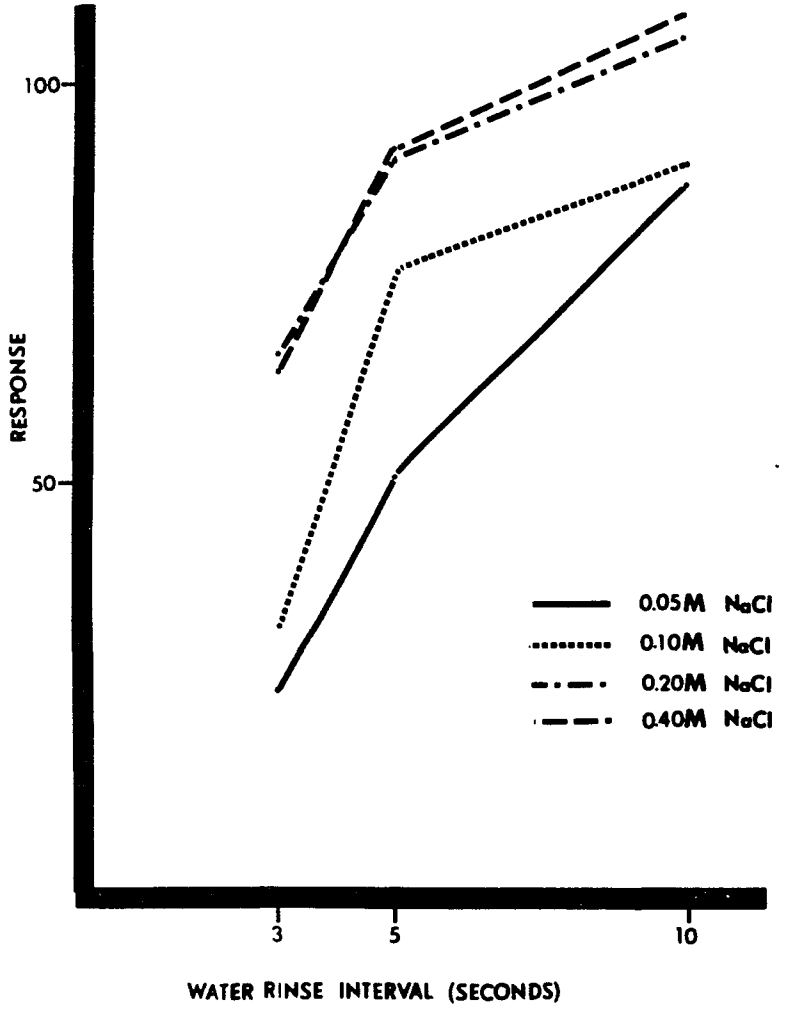
		NaCl Concentration			
		0.05M	0.10M	0.20M	0.40M
Intervening Water Rinse Duration (Seconds).	3	60.7 (82.5)	59.5 (81.3)	39.6 (60.4)	31.5 (53.3)
	5	71.1 (92.9)	63.6 (85.4)	55.5 (76.3)	54.5 (75.3)
	10	85.6 (107.4)	84.3 (105.1)	86.3 (108.1)	78.3 (100.1)

Table 7a. Means and, in parenthesis, the upper bound of the 99% confidence interval, for the effect of NaCl upon 0.3M LiCl as a function of NaCl concentration and intervening water rinse duration.

Table 7b. Means and, in parenthesis, the upper bound of the 99% confidence interval, for the effect of NaCl upon 0.05M HAc as a function of NaCl concentration and intervening water rinse duration.

Figure 6a. The effect of 0.3M LiCl as a function of NaCl concentration and intervening water rinse duration.

Figure 6b. The effect of 0.05M HAc upon NaCl as a function of NaCl concentration and intervening water rinse duration.


100
50
RESPONSE
3
5
10
WATER RINSE INTERVAL (SECONDS)
0.05M NaCl
0.10M NaCl
0.20M NaCl
0.40M NaCl

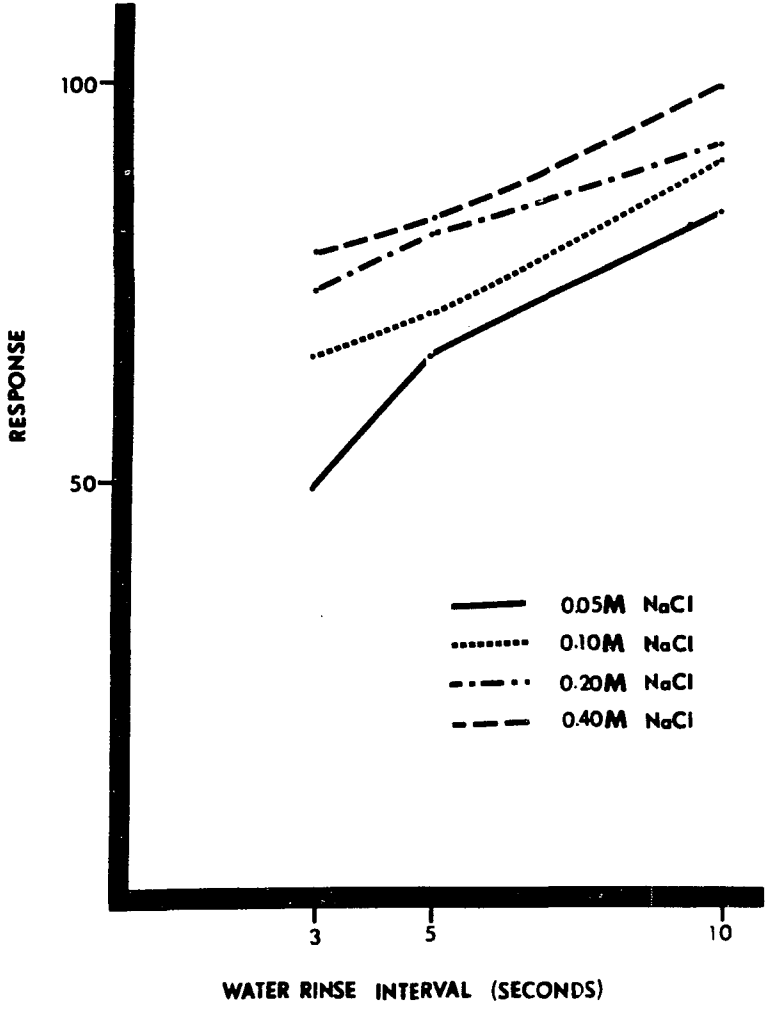
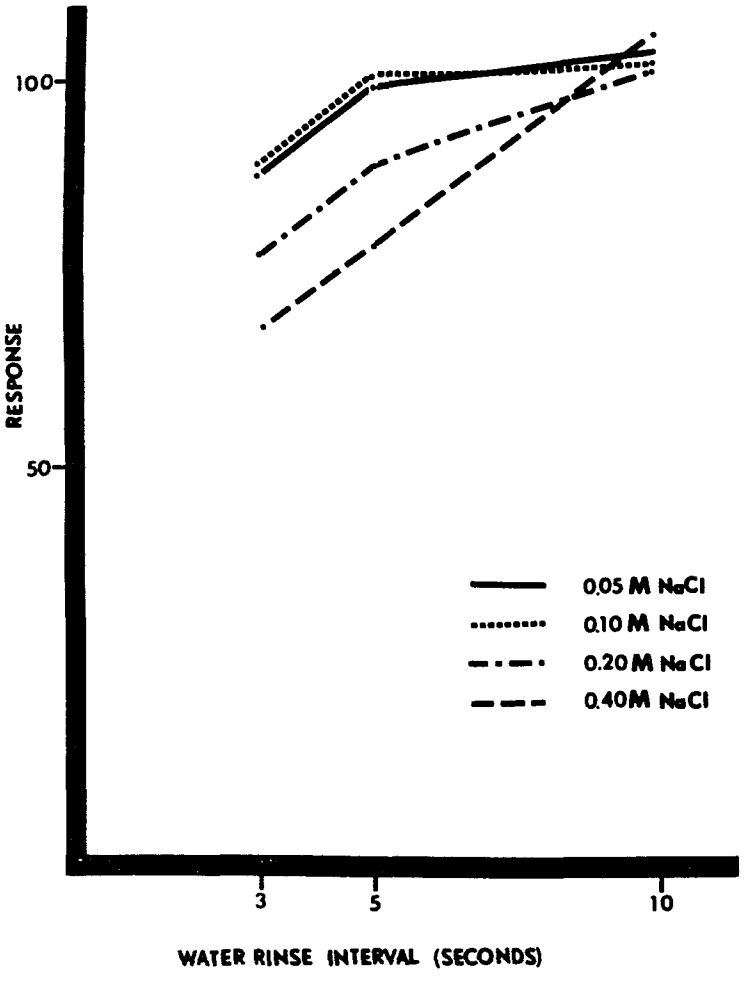
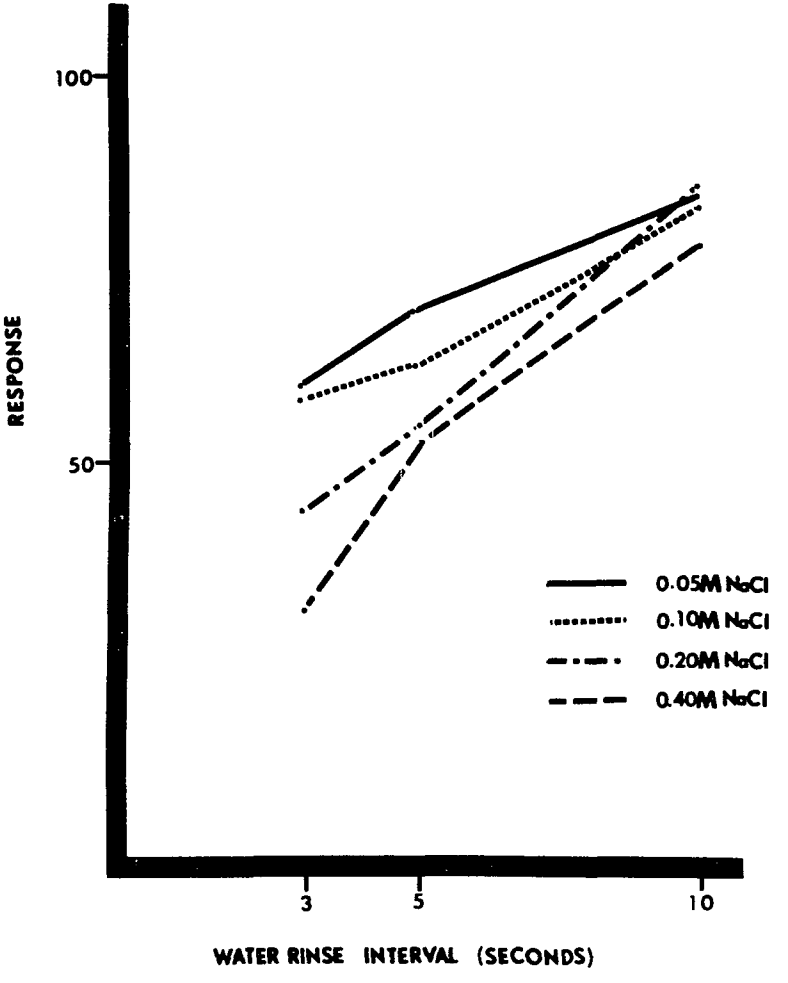

100
50
RESPONSE
3
5
10
WATER RINSE INTERVAL (SECONDS)
0.05M NaCl
0.10M NaCl
0.20M NaCl
0.40M NaCl

Figure 7a. The effect of NaCl upon 0.3M LiCl as a function of NaCl concentration and intervening water rinse duration.

Figure 7b. The effect of NaCl upon 0.05M HAc as a function of NaCl concentration and intervening water rinse duration.


100
50
RESPONSE
0.05 M NaCl
0.10 M NaCl
0.20 M NaCl
0.40M NaCl
3
5
10
WATER RINSE INTERVAL (SECONDS)


100
50
RESPONSE
0·05M NaCl
0.10M NaCl
0.20M NaCl
0.40M NaCl
3
5
10
WATER RINSE INTERVAL (SECONDS)

Experiment IIIa: The Effect of Sugars on the Interaction Effect In Rats.

As in Experiment I, an estimate of the reliability of the depression effect across Ss was obtained with a one-way repeated measures analysis of variance (Table 8). The results of this analysis showed a significant treatment effect ($F = 12.03$, $p < 0.001$). Only 2% of the total variability was attributable to individual differences between Ss while 98% of the total variability was attributable to treatment and error variability. The treatment effects accounted for 74% of the total variability. On this basis, it is again concluded that the interaction effect between taste stimuli is a reliable phenomenon across Ss.

Means and their respective 99% confidence limits for the 16 stimulus pairs are presented in Table 9. It should be noted that a significant interaction effect existed in 12 of the 16 cases; only the 1.0 fructose - 0.05M NaCl, 1.0M fructose - 0.05 HAc, 1.0M sucrose - 0.05M NaCl, and the 1.0M sucrose - 0.05M HAc stimulus pairs failed to show a significant depression effect. The stimulus that produced the greatest depression effect was 0.05M HAc while 1.0M fructose and 1.0M sucrose were equally ineffective in depressing the response to another stimulus (Figure 8). The stimulus that was least susceptible to depression was 0.05M HAc while the stimulus most susceptible to depression was 1.0M fructose (Figure 9).

Source	SS	df	MSq	F
Between	629.63	4		
Within	31060.70	75		
Treatment	23311.24	15	1554.08	12.03 ***
Residual	7749.46	60	129.15	
Total	31690.33			

*** p = 0.001

Table 8. Analysis of variance summary table for the case where each stimulus pair is considered as a different treatment condition: Experiment IIIa

		Stimulus A			
		NaCl	HAc	Fructose	Sucrose
Stimulus B	NaCl	58.70 (82.20)	59.10 (72.60)	90.60 (114.10)	94.54 (118.04)
	HAc	75.42 (98.92)	52.00 (75.50)	94.80 (118.30)	93.20 (116.70)
	Fructose	52.20 (75.70)	55.90 (79.40)	57.78 (81.28)	60.20 (83.70)
	Sucrose	46.10 (69.60)	46.20 (69.70)	76.20 (99.70)	61.44 (84.94)

Table 9. Means and, in parenthesis, the upper bound of the 99% confidence interval for each of the stimulus combinations: Experiment IIIa. To be read, the effect of Stimulus A upon Stimulus B.

Figure 8. The ability of each stimulus to produce a depression effect; Experiment IIIa. To be read, the effect of Stimulus A upon Stimulus B.

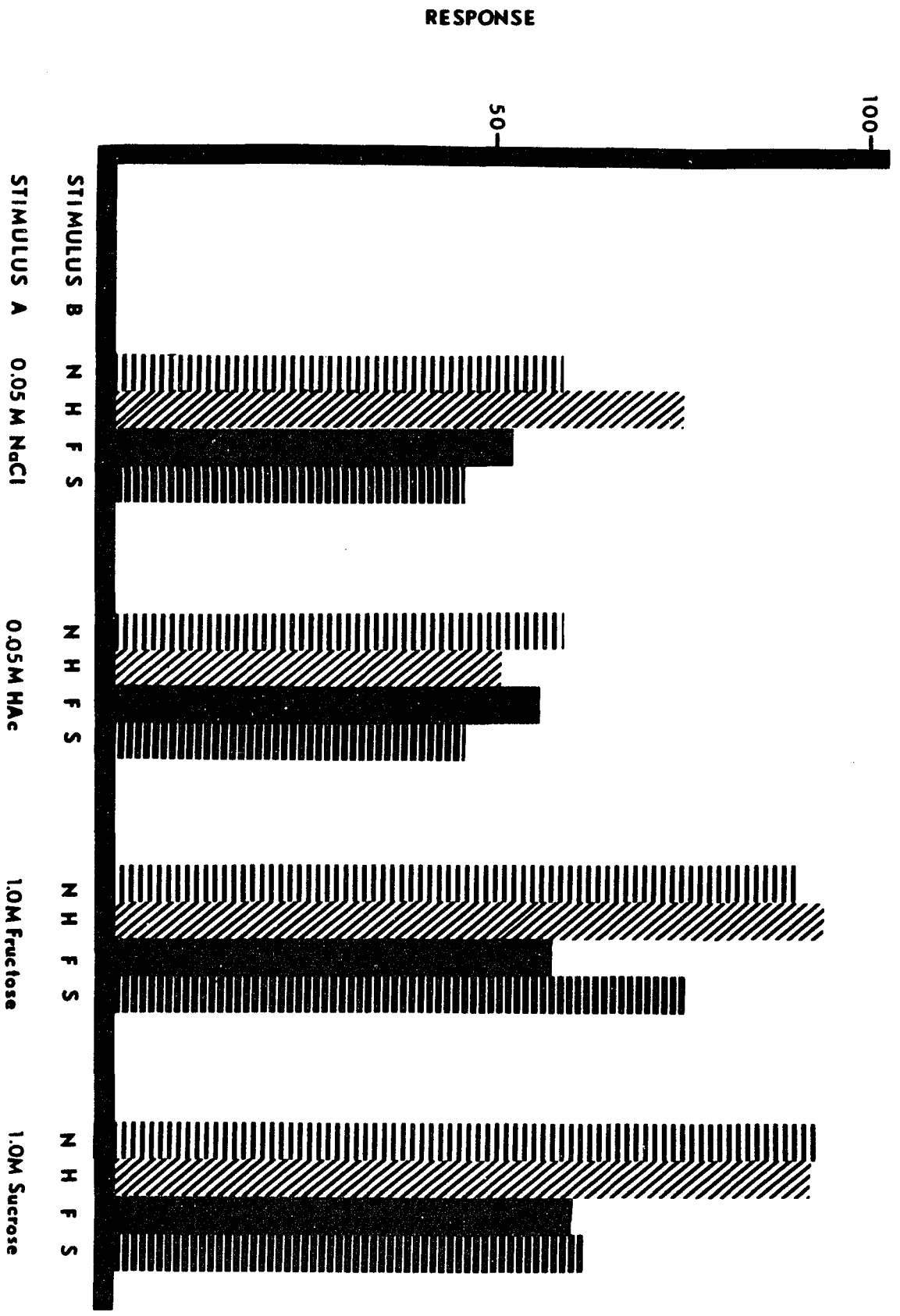
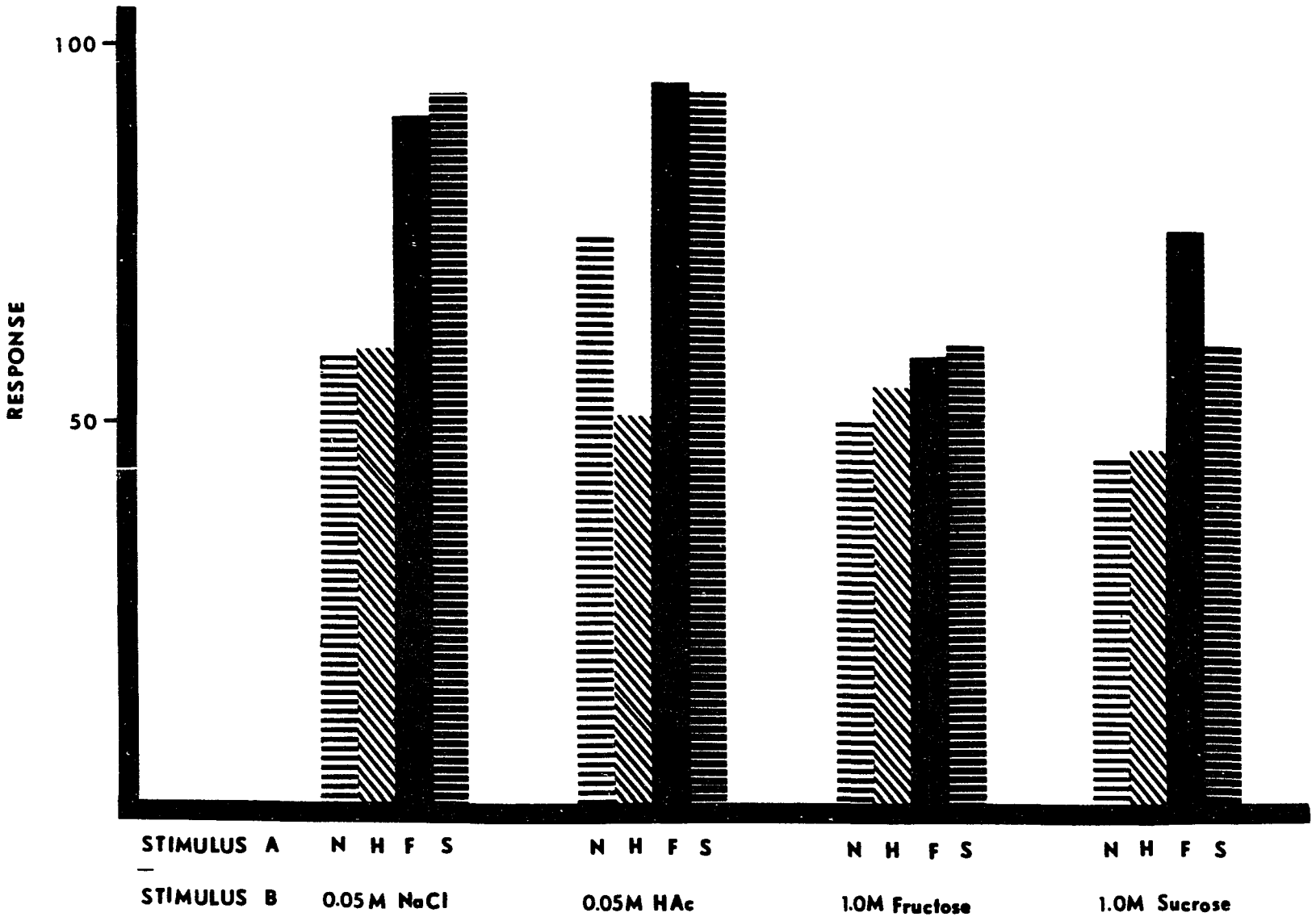

RESPONSE
50
100
STIMULUS A
0.05 M NaCl
0.05M HAc
1.0M Fructose
1.0M Sucrose
STIMULUS B
N H F S
N H F S
N H F S
N H F S

Figure 9. The susceptibility of each stimulus to depression by another stimulus; Experiment IIIa. To be read, the effect of Stimulus A upon Stimulus B.


100
50
RESPONSE
STIMULUS A
N H F S
N H F S
N H F S
N H F S
STIMULUS B
0.05 M NaCl
0.05M HAc
1.0M Fructose
1.0M Sucrose

Experiment IIIb. The Effect of Sugars on the Interaction Effect in Hamsters.

Of the eleven preparations attempted with the hamster, data were collected in only three cases. The results for this experiment are presented in Table 10 and Figures 10 and 11. Examination of the data show several consistent trends across stimuli:

1. 1.0M glucose is the stimulus most readily depressed by all other stimuli in this experiment.
2. 1.0M fructose is the second most depressed stimulus by all other stimuli in this experiment.
3. The stimulus that produces that strongest depression effect is 1.0M sucrose. This stimulus produced the greatest depression effect upon all stimuli except 0.4M NaCl.
4. The stimulus least susceptible to depression is 1.0M sucrose.

The predictions for the interaction between sugars outlined in Chapter I, page 10 are:

1. 1.0M sucrose will produce a greater depression effect upon 1.0M glucose than the effect produced by 1.0M fructose.
2. 1.0M sucrose will produce a greater depression effect upon 1.0M fructose than the effect produced by 1.0M glucose.
3. 1.0M sucrose will produce a greater depression effect upon 1.0M fructose than the depression effect exerted by 1.0M fructose upon 1.0M sucrose.
4. 1.0M sucrose will produce a greater depression effect upon 1.0M glucose than the depression effect exerted by 1.0M glucose upon 1.0M sucrose.

In order to test these predictions, t-tests for correlated samples were conducted. The results of these tests provided support for three of the four predictions. The effect of 1.0M sucrose upon 1.0M glucose was 19.5 and the effect of 1.0M fructose upon 1.0M glucose was 29.66. Although the differences between these means are in the predicted direction, there is no statistical support for this prediction: $t(obs) = 1.27$, $t(p<.10$, one tail $= 1.88)$. The effect of sucrose upon 1.0M fructose is 36.33 and the effect of 1.0M glucose upon 1.0M fructose is 66.3. This difference is statistically significant, $t = 5.2$, $t(p< 0.025) = 4.30$, thus providing support for the second prediction. The effect of 1.0M sucrose upon 1.0M fructose is 36.33 and the effect of 1.0M fructose upon 1.0M sucrose is 75.33. This difference is statistically significant; $t = 3.55$, $p< 0.05$.The effect of 1.0M sucrose upon 1.0M glucose is 19.5 and the effect of 1.0 glucose upon 1.0M sucrose is 78.83. This difference is also statistically significant; $t = 11.00$, $p< 0.005$.

			Stimulus A			
			NaCl	Glucose	Fructose	Sucrose
Stimulus B	NaCl	X =	54.0	71.83	78.00	55.50
		SD =	23.0	8.12	5.56	17.67
	Glucose	X =	48.3	44.66	29.66	19.5
		SD =	8.5	16.41	13.27	12.13
	Fructose	X =	52.25	66.33	44.83	36.33
		SD =	12.36	20.25	12.57	20.03
	Sucrose	X =	65.75	78.83	75.33	46.00
		SD =	5.28	20.31	9.81	15.80

Table 10. Means and standard deviations for the 16 stimulus pairs in Experiment IIIb. To be read, the effect of Stimulus A upon Stimulus B.

Figure 10. The ability of each stimulus to produce a depression effect; Experiment IIIb. To be read, the effect of Stimulus A upon Stimulus B.

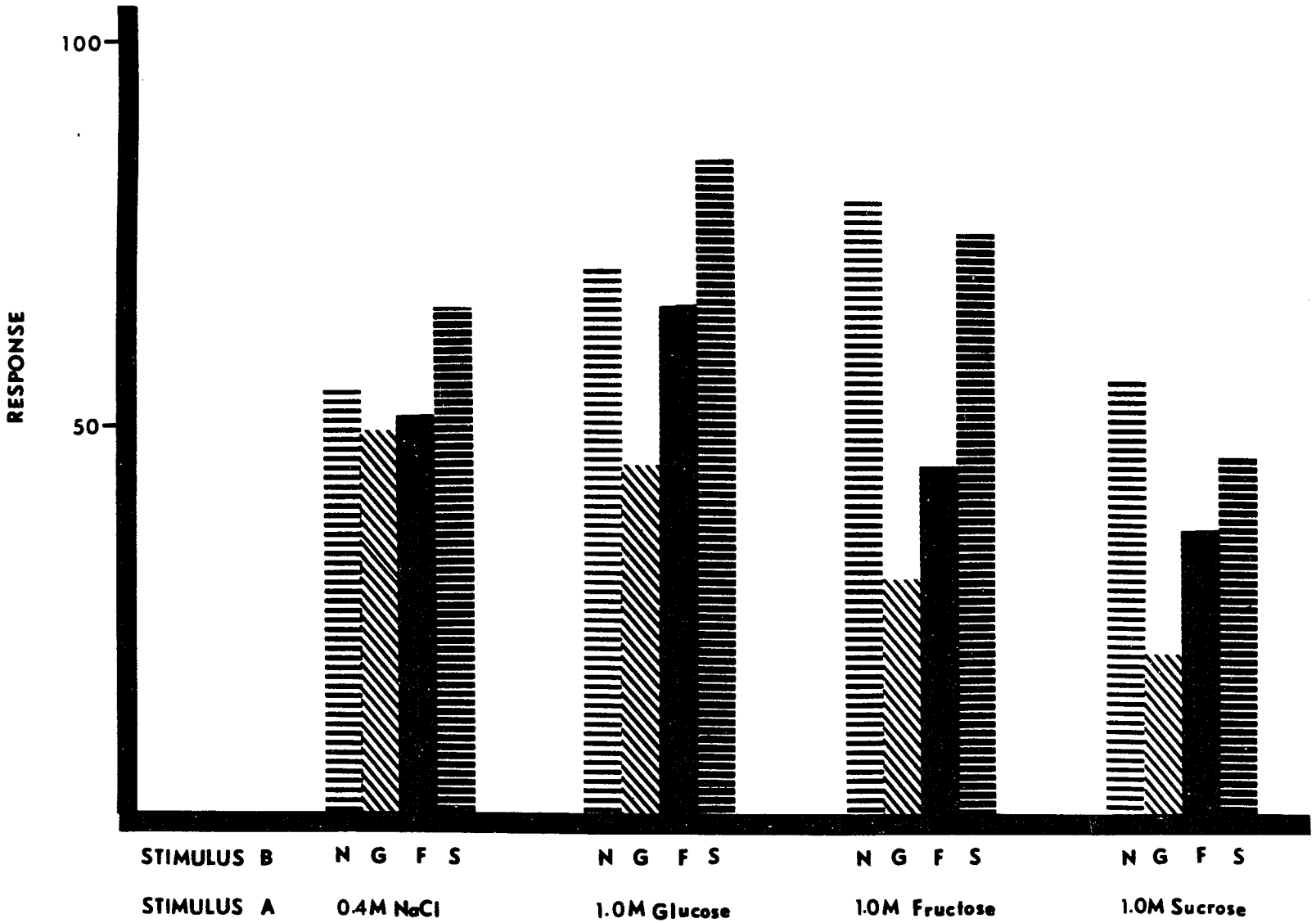
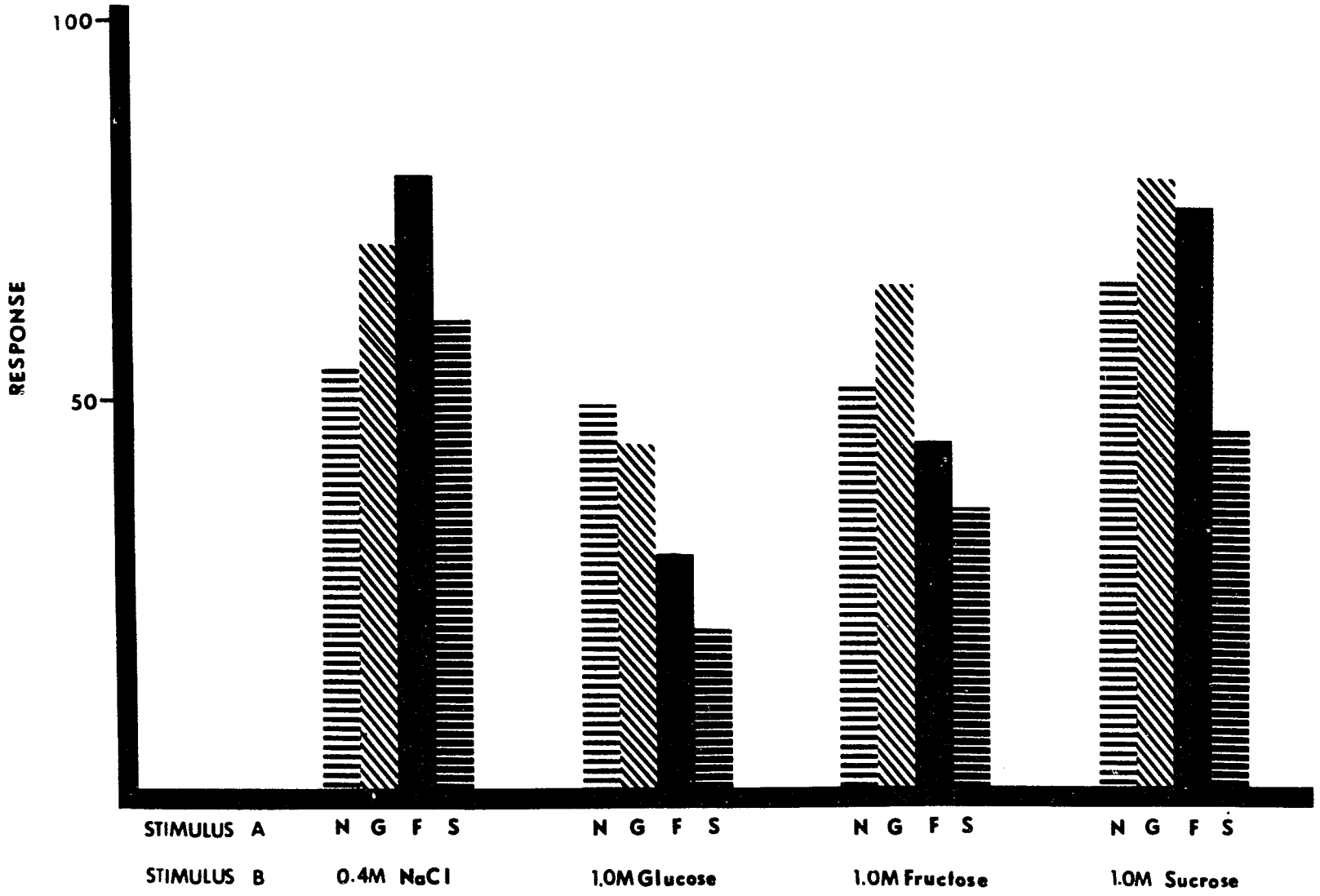

100
50
RESPONSE
STIMULUS B
N G F S
N G F S
N G F S
N G F S
STIMULUS A
0.4M NaCl
1.0M Glucose
1.0M Fructose
1.0M Sucrose

Figure 11. The susceptibility of each stimulus to depression by another stimulus; Experiment IIIb. To be read, the effect of Stimulus A upon Stimulus B.


100
50
RESPONSE
STIMULUS A
N G F S
N G F S
N G F S
N G F S
STIMULUS B
0.4M NaCl
1.0M Glucose
1.0M Fructose
1.0M Sucrose

CHAPTER IV

DISCUSSION

A. Reliability of the Depression Effect.

The results of Experiment I and Experiment IIIa show that the depression effect resulting from the interaction between two stimuli is a reliable phenomenon. That is, a greater portion of the variability in these experiments can be attributed to treatment effects than to individual differences between Ss. The major import of demonstrating reliability of the interaction effect between stimuli is methodological. It is now possible to design experiments with a large number of stimuli because, since the effect is reliable, all stimulus combinations do not have to be presented to all Ss. The problem of the number and type of stimuli employed in a gustation experiment is crucial. Beidler and Gross (1971) estimate that the human tongue may be sensitive to 4,000 to 10,000 different stimuli. However, most investigators severely limit the number of stimuli employed in any given experiment. For example, Hellekant (1969) employed 13 stimuli and Erickson, Doetsch and Marshall (1965) used 11 stimuli (8 of which were salts) to investigate a possible neural coding system. It appears imperative that future research in this area attempt to employ as many different types of stimuli as possible.

B. Species Differences.

Experiment I is instructive because it reveals differences between the cat and the rat with respect to the interaction between taste stimuli (Table 11). While the stimulus 0.3M K_2SO_4 produced a

strong depression effect in both the Hellekant (1969) and the present experiments, its effects were much more profound in Hellekant's (1969) experiment. For example, in the latter experiment, the effect of 0.3M K_2SO_4 upon 0.3M NaCl and 0.3M LiCl were 14 and 40.5, respectively; in the present experiment, these same stimulus effects were 79.8 and 86.7. This same pattern also applies to 0.3M NaCl and 0.3M LiCl; they exerted a greater depression effect upon each other in the Hellekant (1969) experiment than in the present experiment. However, the stimuli, 0.05HAc and 0.3M K_2SO_4 were far more susceptible to depression in the present experiment than in the Hellekant (1969) experiment. In the latter experiment these stimuli were depressed only in selected cases 0.3M K_2SO_4, quinine hydrochloride and choline chloride depressed K_2SO_4 and only HAc exerted a marked depression effect upon HAc, 49.0, while in the present experiment 0.3M K_2SO_4 and 0.05M HAc were always significantly depressed (Table 3).

Another difference between the results of the present research and Hellekant's (1969) results is the time course of the depression effect. In Experiment II of the present research indicated that there was no significant interaction between 0.3M LiCl and 0.05M HAc, and the various concentrations of NaCl at the 10 second water rinse interval. Working with rats, Smith and Frank (1972) also reported that with a 10 second water rinse between applications of 0.1M NaCl, there was no interaction between the two stimulations. Hellekant's (1968) values (water rinse interval when response to Stimulus A is 2/3 of its maximum response) were 13 seconds and 10 seconds for 0.3M LiCl and 0.3M NaCl, respectively.

		Stimulus A			
		NaCl	LiCl	HAc	K_2SO_4
Stimulus B	NaCl	87.3 (70.02)	92.07 (48.5)	93.07 (91.0)	79.81 (14.0)
	LiCl	91.16 (55.5)	96.0 (31.5)	96.42 (97.5)	86.76 (40.5)
	HAc	63.85 (105.7)	73.64 (97.6)	76.5 (49)	78.35 (98.0)
	K_2SO_4	62.42 (110.7)	63.92 (109.5)	79.57 (89.0)	63.57 (65.5)

Table 11. Means of the stimulus combinations in Experiment I and, in parenthesis, the means for these stimulus combinations reported by Hellekant (1969). To be read, the effect of Stimulus A upon Stimulus B.

The fact that the interaction patterns between stimuli are different in the cat and the rat is not surprising in view of the previous literature that substantiates differences in responsiveness to the same stimuli in these species. Beidler, Fishman and Hardiman (1956) reported that inorganic chloride salts, particularly NaCl, is an effective taste stimulus for the rat but not the cat. This result is consistent with the fact that NaCl is more depressed in the cat than in the rat; the receptive field for NaCl may be larger in the rat than in the cat. Pfaffmann (1955) reported that, for the rat, NaCl and HCl were typically more effective as taste stimuli than KCl, whereas, in the cat KCl and HCl were more effective taste stimuli than NaCl. In addition, Pfaffmann (1955) found that the cat is more responsive to quinine than to sucrose while, in the rat, quinine and sucrose were of equal effectiveness as taste stimuli. Beidler and Gross (1971) have summarized the status of species differences with the following statement: "Species differences may be attributed to a quantitative difference rather than a qualitative difference in response profiles" (p.107). In the present investigation, the differences in the interaction patterns may be interpreted to mean that the number of receptor sites for a given stimulus differs in the two species and that the strength of the bind formed between a stimulus and the receptor also differs in the two species.

C. Sugars and the Depression Effect.

In Experiment III, the interrelationships between selected sugar stimuli were examined. The predictions, based upon the configuration of the sugar molecules, were confirmed in three out of the four cases

supporting the notion that the molecular structure of a substance influences the type of interaction this stimulus will have with other stimuli. However, examination of the data shows a pattern of interaction that cannot be readily explained by the molecular structure hypothesis. The stimulus, 1.0M sucrose exerted a strong depression effect upon 0.4M NaCl in Experiment IIIb. If molecular structure were somehow involved in this interaction then it would be expected that either 1.0M fructose or 1.0M glucose would also exert a strong depression effect upon 0.4M NaCl; this was not the case. An alternative explanation for the strong depression effects exerted by 1.0M sucrose is that the size of a molecule is important in determining the degree of depression exerted by a particular stimulus. This explanation is attractive because it accounts for the strong depression effects exerted by sucrose upon 1.0M fructose, 1.0M glucose and 0.4M NaCl. A molecule of large size does not necessarily have to bind with a receptor site to block another molecule from combining with that receptor site but essentially the 'entrance' to a particular receptor site. A suggestion for future research is that molecules of different sizes be employed to determine the relevance of that factor in determining the interaction between taste stimuli.

D. Stimulus Concentration and Water Rinse Duration.

The results of Experiment II show that the variables, stimulus concentration and water rinse duration are orderly variables with respect to the interaction between stimuli. These results support the model that the interaction between taste stimuli is a function of a stimulus binding to a receptor site.

The fact that stimulus concentration has its predicted effect (page 11) shows that the number of receptor sites involved in the interaction effect is a crucial variable. Second, the orderly function associated with the water rinse duration is congruent with the idea that the purpose of the water rinse is to break stimulus-receptor site binds. A suggestion for future research is testing the interaction between stimuli with water rinse as an independent variable to determine if stimulus-receptor site bind strength is a viable method for classifying taste stimuli.

E. Interaction Patterns: Specificity-Generality.

There are several interaction patterns that occur when two stimuli are presented in a successive contrast design. In this section, the interaction patterns that can be explained by assuming that stimuli and receptor sites have characteristics of specificity or non-specificity (generality) will be discussed. The eventual purpose of examining interaction patterns is to identify characteristics of stimuli and/or receptor sites that may be related to the neural coding problem.

One type of interaction that occurs is that Stimulus A depresses its own response greater than it depresses the response to any other stimulus ($a\downarrow a > a\downarrow b$). The explanation for this effect is that Stimulus A is specific in character and will only form a bind with its own receptor sites. An example of this effect is the stimulus, 1.0M fructose, Experiment IIIa (Figure 11). A method for validating this effect would be to use fructose as the adapting stimulus in a cross-adaptation experiment. It is predicted that fructose, because it has a low affinity for foreign receptor sites, would not produce a strong

cross-adaptation effect. Other stimuli that show this interaction pattern are 0.3M K_2SO_4 and 0.05M HAc (Figure 4), and 1.0M glucose (Figure 10).

Another type of interaction is that Stimulus A depresses itself to a greater extent than Stimulus B depresses Stimulus A ($a\downarrow a > b\downarrow a$). This is similar to the first effect; the crucial difference is that in the present case we are examining the action of other stimuli upon Stimuli A whereas, in the first case, the action of Stimulus A upon other stimuli was examined. The explanation for this effect is that the receptor sites for Stimulus A are specific in character, that is, they will not form a bind with any stimulus except Stimulus A. Similarly, if this effect does not occur, this indicates that the receptor sites for Stimulus A are non-specific (general). Examples of the occurrence of this effect are NaCl and HAc in Experiment IIIa (Figure 9) and NaCl and sucrose in Experiment IIIb (Figure 11). Examples of the non-occurrence of this effect are listed below:

Experiment I	Experiment IIIa	Experiment IIIb
0.3M K_2SO_4 → 0.3M NaCl	0.05M HAc → 1.0M fructose	1.0M fructose → 1.0M glucose
0.3M NaCl → 0.3M LiCl	0.05M HAc → 1.0M sucrose	1.0M sucrose → 1.0M glucose
0.3M K_2SO_4 → 0.3M LiCl	0.05M NaCl → 1.0M fructose	1.0M sucrose → 1.0M fructose
0.3M NaCl → 0.05M HAc	0.05M NaCl → 1.0M sucrose	
0.3M LiCl → 0.05M HAc		

For the 16 interaction patterns that could be considered for this case, only four indicate any type of receptor specificity. The remaining cases suggest a degree of non-specificity or communality of receptor sites. This communality is further evidence that the neural

coding of taste quality is not governed by a specificity principle but rather by a spectrum principle as proposed by Pfaffmann (1955) and Erickson (1963).

F. Interaction Patterns: Stimulus-Receptor Site Binds.

The non-occurrence of the second type of interaction effect is expressed as a a a b or, more conveniently, b a a a; Stimulus B depresses Stimulus A to a greater degree than Stimulus A depresses itself. Examples of this case are listed above. The explanation for this effect or pattern requires the additional considerations of receptive field size and the strength of the bind formed between a stimulus and a receptor site. The receptive field for a stimulus has been previously defined as the total collection of receptor sites on the taste cell membrane to which a stimulus may bind. It does not imply any regional or geographic communality. The concept of stimulus-receptor site bind is operationally defined as the duration of the water rinse necessary to prevent an interaction between two stimuli. (This is similar to the value, τ, as defined by Hellekant (1968), (page 5). The explanation for this effect assumes that Stimulus B is able to form a more effective (stronger) stimulus-receptor site bind than Stimulus A for receptor sites that are common to both stimuli. Therefore, the five second water rinse will remove more Stimulus A receptor bonds than Stimulus B receptor bonds. Consequently, when Stimulus A is re-applied to the tongue, fewer receptor sites will be available after Stimulus B than after Stimulus A with the result of a greater reduction in chorda tympani activity after Stimulus B than after Stimulus A. In summary, the model that has been proposed to account for the third type of interaction

effect included the strength of the bind formed between a stimulus and a receptor site and the sharing of receptor sites.

A fourth type of interaction pattern that exists between two stimuli is the non-reciprocal relationship, Stimulus A depresses the response to Stimulus B greater than Stimulus B depresses the response to Stimulus A.

One explanation for this non-reciprocal relationship is that the receptive fields for A and B intersect (Stimulus A and Stimulus B share some receptor sites) and that the receptive field for A is larger than the receptive field for B. If this is the case, then sharing between A and B will have a greater effect on the stimulus with the smaller receptive field, in this case, Stimulus B. For example, if the receptive field for A is 10 arbitrary units and the receptive field for B is 5 arbitrary units and the intersection between A and B is 2 units, then the effect of this intersection concerns 20% of A's receptive field and 40% of B's receptive field. Thus, Stimulus A will block proportionately more of B's receptor than Stimulus B will block A's receptor sites.

G. Stimulus Domain for Taste.

A problem that has prevented progress in understanding the taste process is the lack of a well defined stimulus domain for taste. In other sense systems, e.g. vision, the physical characteristics of the effective stimulus are defined, e.g. wavelengths. This allows the investigator to vary the physical stimulus in some known and controlled manner and observe changes in the sense system. However, in taste this is not possible since only minimal progress has been made in understanding the types and classes of effective gustatory stimuli.

Erickson, et al. (1965) developed a model for understanding the stimulus domain for taste by correlating the amount of activity across different chorda tympani fibers for a variety of stimuli. The model achieved some success; stimuli that showed extremely high correlations (e.g. NaCl and LiCl, r = 0.91) could not be discriminated by rats in a behavioral situation while stimuli with low correlations (KCl and NaCl, r = 0.02) were able to be discriminated by rats. Smith and McBurney (1969) found that nitrate and sulfate salts did not cross-adapt to NaCl as did chloride and bromide salts. This suggests the possibility that two independent salt qualities exist. Smith and Frank (1972) also found two independent salt qualities when studying cross-adaptation in the rat's chorda tympani nerve. Sodium and lithium salts formed one cross-adaptation category and magnesium, calcium, ammonium and potassium belonged to another cross-adaptation category. Smith and Frank (1972) believed that the cation was responsible for the cross-adaptation effect. This appears to contradict the report of Smith and McBurney (1969) who differentiated between salts on the basis of the anion present. However, Smith and McBurney (1969) were able to discriminate between salts based upon the presence of a particular cation except in the case of sulfate and nitrate salts where the anion appears to have greater influence than the cation. Andersen and Hartman (1971) conducted a factor analytic study of the activity of single rat chorda tympani fibers. They concluded that a resolution of stimuli based upon the four basic taste qualities was inadequate. However, a resolution based upon five factors did provide a satisfactory solution accounting for 92.6%

of the total variance. The five factors identified by Anderson and Hartman (1971) are sweet, sour, bitter and two salt factors; the sodium and lithium salts, and salts of large cations (calcium, magnesium, potassium and ammonium). The results of the present research also indicate the presence of two salt categories. In Figure 5 are shown the interaction patterns of Experiment I. The patterns associated with NaCl and LiCl are extremely similar. This pattern of interaction is quite different from the K_2SO_4 pattern of interaction. This result not only confirms the existence of two independent salt qualities but also suggests that the successive contrast design may be profitably used to classify stimuli into different categories.

Clarification of the nature of the stimulus domain for taste is essential to an understanding of the gustatory neural coding process.

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