assuming that HCO_3^- reacts with OH^- to form CO_3^{2-} and water.

When oxygen was rigorously excluded from the solution (Fig. 3), the fall in *p*H with time in the region of the extra ionization was still observed but no keto acid formation could be detected. This would indicate that reactions (2)–(4) here are not the only ones which occur in this system.

We thank the Royal Society for a grant to purchase some of the apparatus used, and the Medical Research Council for financial support for one of us (S. S. T.). Miss Anna Straker helped in preparing the manuscript and figures.

[1] Tate, S. S., Grzybowski, A. K., and Datta, S. P., *J. Chem. Soc.*, 3905 (1965).
[2] Patnaik, R. K., and Pani, S., *J. Indian Chem. Soc.*, **38**, 709 (1961).
[3] Patnaik, R. K., and Pani, S., *J. Indian Chem. Soc.*, **38**, 364 (1961).
[4] Patnaik, R. K., and Pani, S., *J. Indian Chem. Soc.*, **34**, 619 (1957).
[5] Warner, R. C., and Weber, I., *J. Amer. Chem. Soc.*, **75**, 5086 (1953).
[6] Shnarevich, A. I., *Russ. J. Inorg. Chem.*, **8**, 1083 (1963).
[7] Friedemann, T. E., and Haugen, G. E., *J. Biol. Chem.*, **147**, 415 (1943).
[8] McArdle, B., *Biochem. J.*, **66**, 144 (1957).
[9] El Hawary, M. F. S., and Thompson, R. H. S., *Biochem. J.*, **53**, 340 (1953).

WEBER RATIO IN GUSTATORY CHEMORECEPTION; AN INDICATOR OF SYSTEMIC (DRUG) REACTIVITY

By Prof. ROLAND FISCHER, FRANCES GRIFFIN, ROBERT C. ARCHER, STEPHEN C. ZINSMEISTER and Prof. PHILIP S. JASTRAM

Research Division, Department of Psychiatry, College of Medicine and Department of Physics, Ohio State University, Columbus

THE exponential nature of the perceptual process has been amply documented through the years from Fechner's discovery in 1860 to Stevens's[1] proposed modification of it in 1961. In order to propagate quantitative information, the nerve cells cannot increase the intensity of the impulses: they only increase their frequency. Katz[2] and Adrian[3] found that this frequency is proportional to the logarithm of the 'intensity' of the stimulus. It appears now that Stevens's power law describes sensation magnitude before adaptation to a test stimulus, whereas the Fechnerian discriminability law describes it after adaptation[4].

Re-examination of Data and Models

Let us examine the logarithmic function implied in the Weber–Fechner law in one sense modality: gustatory chemoreception. We focus specifically on the data of Lemberger[5] since they have been repeatedly used to support models and mechanisms in gustatory chemoreception.

Lemberger obtained 40 successive 'just-noticeable differences' (jnd) for sodium saccharinate. Beidler[6] plotted these data according to his "fundamental taste equation"[7], an adaptation of the Langmuir adsorption isotherm, with the assumption that "each jnd is proportional to an equal increment of neural activity"; specifically C/R was plotted against C, where C is the concentration and R is the number of the successive 'just-noticeable differences'. A closer inspection reveals that the linear relationship applies for the concentration range between the 11th and the 36th jnd alone.

Duncan[8] also re-examined Lemberger's data for sodium saccharinate and re-plotted her first 22 jnd's; he found a pronounced curvature in the range of the first nine jnd's instead of a linear relationship implied by Beidler. It is apparent, therefore, that Lemberger's data in this lower range do not fit the fundamental taste equation[7] nor is there a linear relationship beyond the 36th jnd.

In summary, Lemberger, Beidler and Duncan used the same set of data to fit their three different models. (1) Lemberger[5] (p. 308) showed that the taste response closely follows a logarithmic curve deviating from the Fechner law only in the lower and higher ranges. (2) Beidler[6] used Lemberger's data to fit his fundamental taste equation, whereas (3) Duncan[8] used them for the demonstration of a "hyperbolic rather than a logarithmic relationship between the magnitude of the response and the stimulus intensity".

It was of interest to us to plot Lemberger's data without trying to fit them to a model. Such an 'assumptionless' plot of the 40 successive jnd's of Lemberger for sodium saccharinate relates the concentrations of the lower limits on the abscissa to their corresponding upper limits on the ordinate. Our plot (Fig. 1) illustrates a closely linear relationship between the lower and upper limits of successive jnd's within the range spanning jnd 7 to jnd 38. Our 'assumptionless' plot apparently illustrates a non-linear relationship for the first 6 successive jnd's followed by a linear relationship to the 38th jnd—just 2 jnd's from the maximum response.

We may define a jnd in taste as a 'signal' and its lower concentration limit 'noise' as Barlow[9] did for light stimuli. The well-known Weber ratio or signal/noise ratio[10] in these terms is independent of the units of measurement and thus is a dimensionless number. (Instead of equal increments of neural activity, we prefer to define Weber ratios as 'dimensionless numbers', that is, 'similarity criteria' or 'dimensionless ratios' rather than 'units'.) We plotted the size of each successive jnd, ΔS, of Lemberger against the concentration of its lower limits, S; the Weber ratio, $\Delta S/S$, is illustrated in Fig. 2 on a log-log scale. A distinctly non-linear relationship exists for the first six successive jnd's resembling a 'half moon'; a linear one for the range between the 7th and the 36th jnd revealing the prevalence of a steady signal/noise ratio. The non-linearity of the 'half-moon' portion of the curve, that is, the first six jnd's with a lack of steady signal/noise ratio, does not support models based on the Langmuir adsorption isotherm.

We proceeded to calculate the mean Weber ratio for sodium saccharinate—between the 7th and the 36th jnd—and found it to be $11{\cdot}02 \pm 0{\cdot}08$ per cent (*S.E.*); the corresponding value for sucrose, again using Lemberger's data, between the 7th and the 22nd jnd, that is, the linear portion of the curve, was larger, namely, $15{\cdot}64 \pm 0{\cdot}14$ per cent (*S.E.*).

The obvious hypothesis suggested itself: the larger Weber ratio for sucrose may be due mainly to its being a less-effective competitor than sodium saccharinate for receptor sites in the presence of tap-water electrolytes. Our subsequent experiments, however, did not substantiate such a hypothesis.

A Comparison of Methodologies in jnd Determination

Lemberger's criteria of taste testing are fully described in her paper[5]. The main features of her procedure are recapitulated only to enable us to compare them with ours. Specifically, she was her own subject, used tap-water as a rinse, a temperature of about 19° C for solutions and

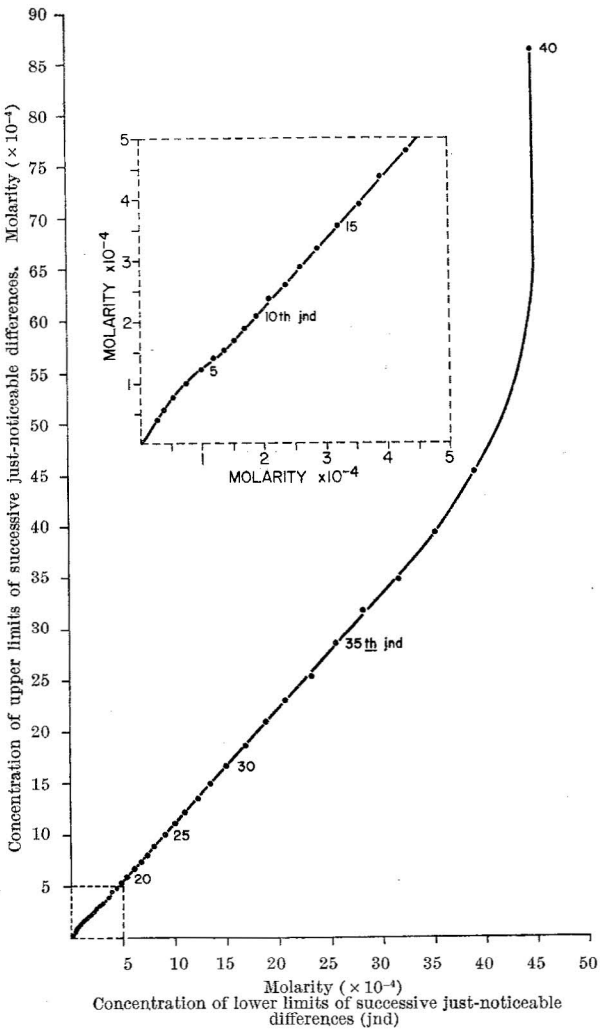

Fig. 1. Relation between upper (ordinate) and lower concentration limits (abscissa) of successive just-noticeable differences (jnd) in the taste sensation of sodium saccharinate calculated from Lemberger's data (ref. 5, Table 8). The plot of the first 18 jnd's is magnified in the upper left corner for better visibility

rinses and a tediously slow sorting procedure—with one jnd determination apparently requiring 2–3 days. Lemberger's limits of accuracy for determining a jnd were based on obtaining at least 75 per cent differentiations.

Our modification of the Harris–Kalmus threshold determination procedure was adapted to just-noticeable taste difference testing and involved the use of distilled water for solutions as well as for rinsing between cups and the final 100 per cent correct sorting technique[11]. The following adaptations were introduced in order to make the method, originally designed for the determination of taste thresholds, suitable for jnd measurements. The original procedure involved taste thresholds, represented by solution numbers each of which corresponded to a concentration the double of the previous concentration. In our adaptation: (1) a threshold—the baseline or lower limit of the first jnd—was determined to a resolution of 1 per cent of a whole solution number; the same was true during subsequent jnd determinations, that is, also for testing the upper limit of each jnd (the standard errors of our procedure are to be found in Tables 1, 2 and 3); (2) during the determination of the upper concentration limit of each jnd we used—instead of distilled water placebos—the upper concentration limit of the preceding jnd; (3) the temperatures of all solutions and rinses were thermostatically controlled to either 19 or 22 ± 0·1° C, the former corresponding to Lemberger's experimental conditions and the latter to our standard laboratory procedure. Our subjects were trained, that is, 'learned', laboratory personnel constantly exposed for from 1 to more than 2 years to taste testing practice, and one additional subject who was a novice in taste testing. Lemberger's procedure required a criterion of 75 per cent correct sorting against our requirement of 100 per cent; this difference, however, cannot account for more than 0·5 per cent change in the values of the concentration limits for jnd's; (4) the concentrations are expressed linearly as molarities in Tables 1, 2 and 3, whereas, in fact, the resolutions of the upper and lower limits of the jnd's are determined to 1 per cent of a concentration corresponding to the next higher solution number, a member of an exponential series with a base of 2 (ref. 12).

The first eleven successive jnd's are well within the linear portion of a Weber ratio plot as evidenced by the fact that even up to the fifteenth jnd the plot remains linear. Therefore, we restricted ourselves to the determination of the first eleven successive jnd's.

Attempts to reproduce Lemberger's Data

Our attempts to reproduce Lemberger's data, specifically the 'half-moon' part of the $\Delta S/S$ plot (Fig. 2), were unsuccessful. We set out, therefore, to determine the influence of each variable in the procedure on the outcome of the experimental data to account for our inability to reproduce her data. We studied the influence of three main variables possibly involved in the initial 'half-moon' feature of Fig. 2, namely, temperature, type of rinse water and chemical structure of the compound.

The importance of the difference in temperature of the compound solutions, rinses and placebos was determined by comparing Weber ratios obtained at a temperature of 19° C (as used by Lemberger) and at a temperature of 22° C. Table 1 presents the first eleven jnd's for sodium saccharinate obtained with solutions, rinses and placebos at 19° C and Table 2 presents jnd's obtained at 22° C. Temperature was found to be of no significance since the mean Weber ratio at 19° C of 33·03 ± 2·6 per cent (distilled water) and 33·23 ± 7·52 per cent (tap-water), com-

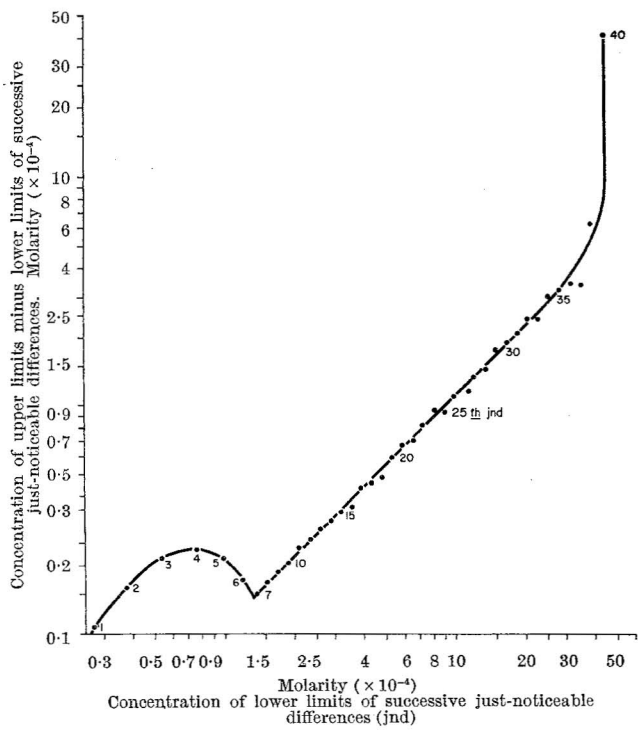

Fig. 2. Difference thresholds for sodium saccharinate. Logarithmic relation between the magnitude (in molarity) of successive just-noticeable differences on the ordinate and the concentration of their corresponding lower limits on the abscissa. Plot based on data of Lemberger (ref. 5)

Table 1. Upper and Lower Limits and their Concentration Differences of the First 11 Successive jnd's expressed in Molarity with Corresponding Weber Ratios as a Percentage for Sodium Saccharinate at 19° C using Distilled Water or Tap-water Rinses

	Distilled water rinse				Tap-water rinse			
Successive jnds	Conc. of lower limit ($M \times 10^5$) (S)	Conc. of upper limit ($M \times 10^5$)	Conc. diff. ($M \times 10^5$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)	Conc. of lower limit ($M \times 10^5$) (S)	Conc. of upper limit ($M \times 10^5$)	Conc. diff. ($M \times 10^5$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)
1	2·53125	3·5625	1·03125	40·74	2·4844	3·5156	1·3012	41·51
2	3·5625	4·5000	0·9375	26·32	3·5156	4·4531	0·9375	26·67
3	4·5000	6·1875	1·6875	37·50	4·4531	6·0000	1·5469	34·74
4	6·1875	8·0625	1·8750	30·30	6·0000	7·9688	1·9688	32·81
5	8·0675	9·3750	1·3125	16·28	7·9688	10·3125	2·3437	29·41
6	9·3750	14·0625	4·6875	50·00	10·3125	14·0625	3·7500	36·36
7	14·0625	17·8125	3·7500	26·67	14·0625	17·6250	3·5625	25·33
8	17·8125	24·3750	6·5625	36·84	17·6250	24·3750	6·7500	38·30
9	24·3750	31·8750	7·6000	31·18	24·3750	31·8750	7·5000	30·77
10	31·8750	42·0000	10·1250	31·76	31·8750	37·5000	5·6250	17·65
11	42·0000	57·0000	15·0000	35·71	37·5000	57·0000	19·5000	52·00
	Mean Weber ratio: 33·03 ± 2·64 per cent ($S.E.$)				Mean Weber ratio: 33·23 ± 7·52 per cent ($S.E.$)			

Table 2. Upper and Lower Limits and their Concentration Differences of the First 11 Successive jnd's expressed in Molarity with Corresponding Weber Ratios as a Percentage for Sodium Saccharinate at 22° C using Distilled Water or Tap-water Rinses

	Distilled water rinse				Tap-water rinse			
Successive jnds	Conc. of lower limit ($M \times 10^5$) (S)	Conc. of upper limit ($M \times 10^5$)	Conc. diff. ($M \times 10^5$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)	Conc. of lower limit ($M \times 10^5$) (S)	Conc. of upper limit ($M \times 10^5$)	Conc. diff. ($M \times 10^5$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)
1	2·3906	3·28125	0·89065	37·26	2·3906	3·28125	0·89065	37·26
2	3·28125	4·2656	0·98435	29·99	3·28125	4·21875	0·9375	28·57
3	4·2656	5·53125	1·26565	29·67	4·21875	5·53125	1·3125	31·11
4	5·53125	7·40625	1·87525	33·90	5·53125	7·5000	1·96875	35·59
5	7·40625	9·3750	1·96875	26·58	7·5000	9·3750	1·8750	25·00
6	9·3750	13·1250	3·7500	40·00	9·3750	13·1250	3·7500	40·00
7	13·1250	16·8750	3·7500	28·57	13·1250	16·6875	3·5625	27·14
8	16·8750	22·1250	5·2500	31·11	16·6875	22·5000	5·8125	34·83
9	22·1250	30·0000	7·8750	35·59	22·5000	30·0000	7·5000	33·33
10	30·0000	37·1250	7·1250	23·75	30·0000	37·5000	7·5000	25·00
11	37·1250	53·2500	16·1250	43·43	37·5000	53·2500	15·7500	42·00
	Mean Weber ratio: 32·71 ± 1·79 per cent ($S.E.$)				Mean Weber ratio: 32·71 ± 5·58 per cent ($S.E.$)			

pared with the mean Weber ratio at 22° C of 32·71 = 1·79 per cent (distilled water) and 32·71 ± 5·58 per cent (tap-water), are clearly of the same order of magnitude.

Secondly, the influence of the type of rinse water was examined. Both sodium saccharinate and sucrose jnd's were determined with tap-water and distilled water rinses (Tables 1, 2 and 3). No significant differences were found in mean Weber ratios for sodium saccharinate at 19° C, 33·23 ± 2·6 per cent (distilled water) and 33·23 ± 7·52 per cent (tap-water); for sodium saccharinate at 22° C, 32·71 ± 1·79 per cent (distilled water) and 32·71 ± 5·58 per cent (tap-water); for sucrose at 22° C, 41·06 ± 1·02 per cent (distilled water) and 40·48 ± 1·14 per cent (tap-water).

Our investigation of the third variable, the possible influence of the chemical structure of a compound on the mean Weber ratio, was a comparison of the size of mean Weber ratios for sucrose (an un-ionized compound) with the data for sodium saccharinate (an ionized compound) at 22° C for the two-rinse water conditions (Tables 2 and 3).

A survey of our experimental data allows us to state the following: (1) A 3° C difference in temperature has no effect on the mean Weber ratios for sodium saccharinate other than to increase slightly the standard error at the lower temperature. (2) The type of rinse water has no influence on the mean Weber ratios for sodium saccharinate; however, the effect of a tap-water rinse is to increase the standard error (see Tables 1 and 2); (3) the tap-water rinse has no effect on the mean Weber ratios for either of the two compounds at 22° C; however, the standard error with tap-water rinse for the ionized compound (sodium saccharinate) is increased more than for sucrose, the un-ionized compound (see Tables 2 and 3).

In spite of controlling temperature and type of rinse water, we are unable to reproduce the initial curvilinear portion of Lemberger's sodium saccharinate curve ('half-moon') nor to establish a single factor to account for the lack of this initial curvilinear portion (from the first to the sixth jnd) in our data. Nor can we find any explanation for the 2·5–3 times smaller size of Lemberger's mean Weber ratios for sucrose and sodium saccharinate, respectively, within the linear range of her data (from the 7th to the 36th jnd) compared with our mean Weber ratios.

Further differences between Lemberger's and our methodologies, however, may account for our inability to reproduce the curvilinear 'half-moon' (the range between the first and sixth jnd) and for the fact that her Weber ratios were 2·5–3 times smaller than ours:

(1) Our average daily performance consisted of determining up to 20 jnd's; whereas Lemberger, according to her description, can be assumed to have taken 2–3 days for the determination of one jnd.

(2) Our subject, as already mentioned, was 'learned', that is, his threshold for quinine due to conditions of well-defined practice suddenly decreased three thresholds in solution numbers, that is, eight-fold from the point of view of concentration[12] before this experimentation (each successive solution number represents a doubling concentration). We can define his taste sensitivity prior to learning: taste threshold for quinine in solution numbers equals 3, whereas that for 6-*n*-propylthiouracil (PROP) equals solution number 6. The sucrose threshold of our subject was solution number 15, quite similar to that of Lemberger, whose threshold 16, expressed in our solution numbers, corresponds to a concentration of $2{\cdot}4 \times 10^{-2}$ M. Lemberger could not have been a 'learned' subject before her experimentation since, according to our experience, learning is apparently restricted to taste practice with quinine under defined conditions and it is not known whether learning can be achieved by tasting sodium saccharinate under Lemberger's experimental conditions. We also attempted to reproduce Lemberger's data with an 'unlearned' subject also slow in taste testing, who, under our experimental conditions, displayed from 25 to 100 per cent variation in the size of his first jnd for sodium saccharinate. We do not know whether the extent of variability is a characteristic of unlearned subjects or an individual characteristic of this 'slow' subject.

Although Lemberger repeated the determination of each jnd until the standard error was minimal for it, she

Table 3. UPPER AND LOWER LIMITS AND THEIR CONCENTRATION DIFFERENCES OF THE FIRST 11 SUCCESSIVE JND'S EXPRESSED IN MOLARITY WITH CORRESPONDING WEBER RATIOS AS A PERCENTAGE FOR SUCROSE AT 22° C USING DISTILLED WATER OR TAP-WATER RINSES

	Distilled water rinse				Tap-water rinse			
Successive jnds	Conc. of lower limit ($M \times 10^{-2}$) (S)	Conc. of upper limit ($M \times 10^{-2}$)	Conc. diff. ($M \times 10^{-2}$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)	Conc. of lower limit ($M \times 10^{-2}$) (S)	Conc. of upper limit ($M \times 10^{-2}$)	Conc. diff. ($M \times 10^{-2}$) (ΔS)	Weber ratio (per cent) ($\Delta S/S \times 100$)
1	0·780	1·080	0·300	38·46	0·780	1·080	0·300	38·46
2	1·080	1·560	0·480	44·44	1·080	1·560	0·480	44·44
3	1·560	2·160	0·600	38·46	1·560	2·160	0·600	38·46
4	2·160	3·120	0·960	44·44	2·160	3·120	0·960	44·44
5	3·120	4·320	1·200	38·46	3·120	4·320	1·200	38·46
6	4·320	6·144	1·824	42·22	4·320	6·144	1·824	42·22
7	6·144	8·544	2·400	39·06	6·144	8·640	2·496	40·625
8	8·544	12·480	3·936	46·07	8·640	12·480	3·840	44·44
9	12·480	17·280	4·800	38·46	12·480	16·896	4·416	35·38
10	17·280	24·960	7·680	44·44	16·896	22·656	5·760	34·09
11	24·960	34·176	9·216	36·92	22·656	32·640	9·984	44·07
	Mean Weber ratio: 41·06 ± 1·02 per cent ($S.E.$)				Mean Weber ratio: 40·48 ± 1·14 per cent ($S.E.$)			

did not repeat the whole procedure of successive jnd determinations. We are unable, therefore, to calculate her standard error for reproducing the whole curve or to compare her data with that of our 'unlearned' subject.

The relation between the magnitude of stimulation and sensory response is tabulated for the six sets of data in Tables 1, 2 and 3, of which the latter deserves special attention. With arithmetically increasing concentration limits there is a corresponding geometrical increase in magnitude of jnd. This increase, evident in Table 3, is progressing in a quantum fashion, whereby each quantal unit is exactly the double of the previous one. The sinusoidal fluctuation in the magnitude of the first eleven Weber ratios (see Table 3), alternating with troughs of 38·46 per cent and peaks of 44·44 per cent, correspond to a period of 30 min.

In summary, a graphic and experimental re-examination of Lemberger's data led us to the elucidation of methodological factors in jnd determination influencing the size of a dimensionless ratio, the Weber ratio. For both sodium saccharinate and sucrose, we found a straight line plot for the Weber ratios with rhythmical doubling of concentration limits and sinusoidal fluctuations in the Weber ratios.

Size of a Weber Ratio as an Indicator of Systemic Reactivity

Recently it was found that a statistically significant relationship exists (within a group of 48 acutely ill female schizophrenic patients) between extrapyramidal side effects induced by a low cumulative dose of trifluoperazine and taste sensitivity. Side effects displayed in response to higher trifluoperazine dosage were more likely to occur in taste-insensitive patients[13].

We wondered whether the size of a Weber ratio, instead of taste threshold, would also be related to systemic (drug) reactivity. Our pilot experiments indicate that trifluoperazine-induced tranquillization increases, whereas 'Psilocybin'-induced (central) sympathetic stimulation decreases, the size of an individual's Weber ratio for sodium saccharinate (see Table 4).

Our sample consisted of 12 acutely ill mental patients, 7 of whom were on high and 5 on low trifluoperazine medication with a male to female ratio 1 : 1; 7 healthy volunteers of whom 5 were under the influence of 'Psilocybin' and a week later without any drug. 'Psilocybin' was chosen because it produces a transient state of central sympathetic stimulation[14], thus contrasting the tranquillizing effect of trifluoperazine and enabling us to compare the size of Weber ratios under these two extreme systemic conditions.

We used sodium saccharinate at 22° C with distilled water rinses when determining a jnd approximately two thresholds above the sodium saccharinate threshold of a patient but still well within the linear portion of the Weber ratio plot.

Even individual variations in reactivity to trifluoperazine or to 'Psilocybin' seem to be mirrored in the size of the Weber ratio. To give a specific example: a taste-sensitive volunteer reacted to 12·5 mg 'Psilocybin' more intensely than another less taste-sensitive subject to 15 mg of the same drug; their Weber ratios at the peak of drug experience were 29 and 39 per cent respectively.

One set of data requires comment, namely, the approximately 20 per cent higher mean Weber ratio of the 7 healthy volunteers in Table 4 (48·9 ± 1·8 per cent) as compared with the size of the mean Weber ratio for sodium saccharinate in Table 2 (32·71 ± 1·79 per cent). The difference may be accounted for by our observation that continuous taste practice diminished the size of the Weber ratio of our subject in Table 2, whereas the 7 volunteers in Table 4 were not previously exposed to such practice. The nearly identical size of the Weber ratio (48·9 ± 1·8 per cent) in our 7 volunteers may be accounted for by one factor: the sodium saccharinate baseline used in all 7 subjects was identical, namely, $9{\cdot}38 \times 10^{-5}$ M.

Table 4. RELATION BETWEEN DEGREE OF TRANQUILLIZATION OR SYMPATHETIC STIMULATION, THAT IS, SYSTEMIC (DRUG) REACTIVITY AND THE SIZE OF THE WEBER RATIO FOR SODIUM SACCHARINATE AT 22° C WITH DISTILLED WATER RINSES

Subjects	Mean Weber ratio for sodium saccharinate at 22° C
(1) Patients on a high daily dose of trifluoperazine (30–60 mg). $N = 5$, 25–40 years of age	217 ± 9 per cent ($S.E.$)
(2) Patients on a low daily dose of trifluoperazine (5–20 mg). $N = 7$, 25–40 years of age	132 ± 13 per cent ($S.E.$)
(3) Healthy volunteers without any drug. $N = 7$, 21–28 years of age	48·9 ± 1·8 per cent ($S.E.$)
(4) Healthy volunteers, 100 min after the ingestion of 10–15 mg of 'Psilocybin'*, same subjects as (3). $N = 5$, 21–28 years of age	34 ± 2·7 per cent ($S.E.$)

* Only subjects previously exposed to 'Psilocybin' were included to eliminate effects related to "novelty of experience".

There are two other factors which may influence the size of a Weber ratio, namely, taste sensitivity and sex. Sensitive tasters have smaller Weber ratios than insensitive tasters; women are, under otherwise identical conditions, more sensitive tasters than men[15]. That excitation diminishes the size of the Weber ratio should not be surprising in view of the data of Kimura[16] and Chernetski[17]. The latter author found that systemic sympathetic stimulation enhances afferent activity recorded from the glossopharyngeal nerve during application of gustatory stimuli to a frog's tongue. This article also offers an explanation for the sympathetic enhancement of peripheral gustatory activity noted by Kimura in the rat.

We have dealt with methodological and systemic factors related to the Weber ratio in gustatory chemoreception. The constancy within limits of this dimensionless ratio reminds us that we live in a world in which the absolute magnitudes of stimuli go unperceived, a perceptually compressed but "the best of all possible worlds".

The work was supported by U.S. Public Health Service National Institutes of Health, general research support grants administered by the Ohio State University College of

Medicine. We thank the following for discussions, assistance and interest: Dr. J. Gander, Dr. R. Kaelbling, Dr. W. Knopp, Miss B. Mayers, Dr. B. Pasamanick, Miss M. Rockey, Dr. J. Sandberg and Dr. A. Sollberger (Cleveland). We also thank Dr. R. Bircher and Dr. L. B. Achor, Sandoz Pharmaceuticals, Hanover, N.J., for providing us with samples of 'Psilocybin'.

[1] Stevens, S. S., *Science*, **133**, 80 (1961).
[2] Katz, B., *Electric Excitation of Nerve* (Oxford University Press, London, 1939).
[3] Adrian, E. D., *The Basis of Sensation* (Christopher, London, 1928).
[4] Green, E. E., *Science*, **138**, 1274 (1962).
[5] Lemberger, F., *Pflüger's Arch. Ges. Physiol.*, **123**, 293 (1908).
[6] Beidler, L. M., 1 (see Fig. 10, p. 6) in *Proc. Second Symp. Physiol. Psychol.*, March 19–21, Office of Naval Research, Dept. of the U.S. Navy, *O.N.R. Symposium Report ACR*–30 (1958).
[7] Beidler, L. M., *J. Gen. Physiol.*, **38**, 133 (1954).
[8] Duncan, C. J., *J. Theoret. Biol.*, **5**, 114 (1963).
[9] Barlow, H. B., *J. Physiol.*, **136**, 469 (1957).
[10] Rushton, W. A. H., in *Sensory Communication*, edit. by Rosenblith, W. A., 169–182 (M.I.T. Press, Cambridge, Mass., and Wiley, New York, 1961).
[11] Fischer, R., Griffin, F., England, S. J. M., and Pasamanick, B., *Med. Exp.*, **4**, 356 (1961).
[12] Fischer, R., and Griffin, F., *Arzneimittelforschung* ("Drug Research"), **14**, 673 (1964).
[13] Fischer, R., Knopp, W., and Griffin, F., *Arzneimittelforschung* ("Drug Research" (in the press).
[14] Hofmann, A., Heim, R., Brack, A., *et al.*, chap. 5 in *Les Champignons Hallucinogènes du Mexique*, edit. by Heim, R., and Wasson, D. (*Archives du Muséum National d' Histoire Naturelle*, Paris, Série 7, **6**, 247, 1958).
[15] Glanville, V., Kaplan, A. R., and Fischer, R., *J. Gerontol.*, **19**, 474 (1964).
[16] Kimura, K., *Kumamoto Med. J.*, **14**, 95 (1961).
[17] Chernetski, K. E., *J. Neurophysiol.*, **27**, 493 (1964).

IMPORTANCE OF MEMBRANES IN PROTEIN BIOSYNTHESIS

By Dr. RICHARD W. HENDLER

Laboratory of Biochemistry, Section on Cellular Physiology; National Heart Institute; National Institutes of Health; Bethesda, Maryland

THREE years ago, on the basis of a wide variety of observations reported in the literature from this and other laboratories, a general model for protein synthesis was proposed which suggested that ribosomes are membrane-associated when they carry out their synthetic activities in the living cell[1]. Since that time many new investigations bearing on the validity of such a functional cytological relationship have appeared. Since, at the moment, these important and related papers have not been considered as a unit, it seemed of distinct value to assemble the information so that a clear picture could be drawn of the existing evidence. Although many earlier reports demonstrated that membrane preparations, obtained principally from bacteria, were capable of efficient amino-acid incorporation into protein, the question of whether or not this represented a different mode of incorporation than the more usual free ribosome mediated systems was not amply considered. Furthermore, some systems actively studied, notably *E. coli*, were capable of efficient amino-acid incorporation even though an intracytoplasmic membrane system does not appear to be present and the ribosomes are considered to exist free in the cytoplasm. For the generality of the proposed ribosome-membrane model to extend to such systems as *E. coli*, the possibility was considered that a certain proportion of the ribosomes of *E. coli* were attached to the plasma membrane at any time. These attached ribosomes would be responsible for protein synthesis in the intact cell and due to a lability of their linkage to the membrane would be dissociated by the forces used to break the cell and afterwards would appear as free ribosomes.

In a series of papers, Hendler and Tani presented evidence that the foregoing description does apply to *E. coli* and that the generality of the membrane-ribosome structure for protein synthesis is not compromised by the finding of high amounts of incorporated amino-acids in free ribosome fractions[2–4].

In several investigations dating from 1958, the group at the Chester Beatty Institute, including Drs. Butler, Hunter, Godson, Crathorn, and Goodsall, examined the site of amino-acid incorporation in *B. megaterium*[1]. They showed that the membrane represented the major site of incorporation, that an appreciable fraction of the cellular RNA sedimented with and was held by the membrane, and that ribosomal particles could be released from the membrane by washing with buffers of low ionic strength. Schlessinger recently re-examined this system and confirmed that an appreciable fraction of the total RNA was bound to membranes (more than 50 per cent[5]). Most of this RNA was released from the membranes by lowering the magnesium concentration from 10^{-2} M to 0. Similarly, 0·2 per cent DOC or subsequent incubation alone was capable of releasing the bound RNA. The RNA released by DOC treatment sedimented in a sucrose gradient principally as polysomes of 4 or more ribosomes. The isolated membranes were capable of incorporating up to 6 times more amino-acid into protein per unit weight of RNA than the ribosome–polysome mixture obtained from a crude extract. Schlessinger pointed out that there was no direct evidence that the polysomes found in the DOC extracts of membrane were present in the membranes as polysomes. In yeast, a membrane–ribosome fraction was described which could incorporate sulphur-35 into stable peptide linkage 5–7½ times faster than free ribosomal particles on a per unit RNA basis[6].

Moore and Umbreit, in a preliminary communication[7], reported that in *S. fecalis*, when a washed membrane fraction and a ribosomal fraction were compared, it was found that the membrane-bound ribosomes were about 5-fold more effective than cytoplasmic ribosomes for the incorporation of radioactive amino-acids. This work has continued and Moore and Umbreit have demonstrated that the incorporation of amino-acids in the free ribosome fraction is proportional to their phospholipid concentration. Phospholipid is found only in membranes of *S. fecalis* and is absent from cell walls and cytoplasm. Consequently, these authors conclude that the results indicate a membrane constituent to be the limiting factor in protein synthesis in this system even for the 'free' ribosome fraction[8].

The localization of ribosomes at membrane sites is further demonstrated by studies involving the incorporation of labelled precursors into RNA in *E. coli*[9] and *Streptococcus fecalis*[10]. In HeLa cells infected with polio virus, it has been shown that the polio-specific polysomes are also membrane-attached[11]. Another study having a possible bearing on this question is that of Neu and Heppel[12]. They found that during the formation of EDTA-lysozyme spheroplasts of *E. coli*, all the latent ribonuclease activity was released from the unlysed cells. β-Galactosidase was not so released, but could be obtained after lysis. If, as is commonly thought, the latent RNase is a part of the *E. coli* ribosome, then this investigation would be consistent with a membrane localization of the ribosomes. Another alternative considered by these authors is that the ribonuclease may actually not be part of the ribosome, but could exist free in a pericytoplasmic compartment.

The sensitivity of micro-organisms to antibiotics also points to a functional association of membranes and