390. *The Standardisation of some Buffer Solutions at Elevated Temperatures.*

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This paper records measurements of the p_H values at 12·5°, 25°, 34°, 53°, 63°, 75° and 91° for the Britton–Robinson universal buffer mixture and also for buffer solutions obtained by the separate neutralisation of the constituent acids, *viz.*, citric, diethylbarbituric, and boric acid and potassium dihydrogen phosphate. The dissociation constants of the acids have been calculated at the different temperatures, and the results discussed.

SAVE for the work of Walbum (*Biochem. Z.*, 1920, **107**, 219), who determined the p_H values of some of Sørensen's buffer solutions (*ibid.*, 1909, **21**, 131; **22**, 352; *Ergebn. Physiol.*, 1912, **12**, 393) at temperatures varying from 10° to 70°, and of Kolthoff and Tekelenburg

(*Rec. trav. chim.*, 1927, **46**, 33; Kolthoff, " Saure-Basen Indicatoren," 4th ed., 1932, p. 251 *et seq.*), who also investigated buffer mixtures between 10° and 60°, little seems to have been done to ascertain the usefulness of buffer mixtures at elevated temperatures, and this is particularly true of universal buffer mixtures such as might be used to calibrate the glass electrode. As it was desired to determine the p_H values of solutions at elevated temperatures by means of the glass electrode, it was necessary to find the p_H values corresponding to different stages of neutralisation of a buffer mixture which extended over a wide p_H range. Prideaux and Ward's universal buffer mixture (J., 1924, **125**, 426) was first considered, but had the serious disadvantage that one of its constituents, *viz.*, phenylacetic acid, is appreciably volatile at the higher temperatures; Prideaux's earlier buffer mixture (*Proc. Roy. Soc.*, 1916, **92**, *A*, 463), in which acetic acid is used, is still more disadvantageous. For use at higher temperatures Britton and Robinson's universal buffer mixture (J., 1931, **1456**) was much more satisfactory : now that diethylbarbituric acid can be obtained commercially, there is no need to use the sodium salt and an equivalent amount of hydrochloric acid in preparing the mixture. All the constituents can therefore be weighed out directly, and it thus becomes a solution which is 0·02857M. with respect to each of its four constituents.

For work within smaller p_H ranges buffer solutions were prepared from each of the constituents separately, and the p_H values found when they were treated with increasing amounts of sodium hydroxide at various temperatures. 0·2N-Solutions of potassium dihydrogen phosphate, citric acid, and boric acid were used, but owing to its low solubility diethylbarbituric acid was used in 0·03N-solution.

EXPERIMENTAL.

The apparatus described in the preceding paper was employed, and the hydrogen was saturated with water vapour at each temperature before being passed very slowly through the hydrogen electrode. The pressure of hydrogen, P, surrounding the electrode was therefore equal to 1 atm. minus the saturated vapour pressure of water, p, and consequently $P = (760 - p)/760$ atm. The *E.M.F.* of the cell

$$\overset{+}{\text{N-Calomel}} \;\Big|\; \text{N-KCl} \;\Big|\; \text{Satd. KCl} \;\Big|\; 100\text{ c.c. Buffer mixture} + x\text{ c.c. } 0{\cdot}2\text{N-NaOH} \;\Big|\; \overset{-}{H_2}$$

$$= E_{\text{N-Cal.}} - E_{H_2} = E_{\text{N-Cal.}} - \boldsymbol{R}T/\boldsymbol{F}\log_e[\text{H}^{\bullet}]/\sqrt{P}$$
$$= E_{\text{N-Cal.}} + \tfrac{1}{2} \times 0{\cdot}0001984T\,.\log P + 0{\cdot}0001984T \times p_H$$

The values, $E_{\text{N-Cal.}}$, are those recorded by the authors (*loc. cit.*).

The data are given in Table I, and those of the universal buffer mixture are plotted in the figure, Britton and Robinson's original data for 18° being plotted as a broken line. It will be seen that, as the temperature is increased, the p_H values vary but little until p_H 6 is reached. Thereafter they becomes considerably lower as the temperature increases, the curves exhibiting marked departure from the rectilinearity characteristic of that at ordinary temperatures. The upper scale of the figure indicates when the different acids or acid stages undergo neutralisation with sodium hydroxide. The p_H values during the addition of the first 3 equivs. of alkali, corresponding with the neutralisation of the three stages of citric acid, change but little with temperature. This effect is seen more clearly in Table I (i), the first 42·9 c.c., and in Table I (ii), the reason being that temperature has but a slight influence on each of the three dissociation constants (Table II). Table II (i) gives the p_H values set up at the mid-points of neutralisation of each of the successive stages of neutralisation of 0·2N-citric acid [Table I (ii)], and on the assumption that these values are approximately those of p_{K_1}, p_{K_2}, and p_{K_3} respectively, it will be seen that, whereas K_1 and K_2 are almost constant, K_3 becomes somewhat smaller with rising temperature. In the remaining sections of Table II, the classical p_K values of the second and third stages of phosphoric acid and of boric and diethylbarbituric acids are given. They were calculated from Table I, (iii)—(v). The data reveal that, except for the ionisation $H_2PO_4' \rightleftharpoons H^{\bullet} + HPO_4''$, which appears scarcely to be influenced by temperature, the constants increase with temperature. (The constants calculated from p_H data set up during the first half of the neutralisation of boric acid are a little larger than those referring to the second half, a difference which becomes less apparent at the higher temperatures.) These enhanced constants explain why the curves referring to the addition of the last 4 equivs. of alkali to the universal

TABLE I.

x.	(i) Britton–Robinson universal buffer mixture.							(ii) 0·2N-Citric acid.						
	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.
0	2·38	2·42	2·47	2·53	2·55	2·59	2·64	2·12	2·13	2·13	2·14	2·14	2·14	2·14
5	2·86	2·90	2·92	2·94	2·94	2·96	2·96	2·43	2·43	2·43	2·44	2·44	2·44	2·44
10	3·36	3·36	3·36	3·35	3·35	3·35	3·35	2·70	2·70	2·70	2·70	2·70	2·70	2·70
15	3·94	3·92	3·89	3·85	3·84	3·83	3·82	2·93	2·93	2·93	2·93	2·93	2·93	2·93
20	4·42	4·40	4·37	4·35	4·34	4·33	4·33	3·15	3·15	3·15	3·15	3·15	3·15	3·15
25	4·83	4·82	4·80	4·80	4·80	4·80	4·80	3·37	3·37	3·37	3·37	3·37	3·37	3·37
30	5·25	5·27	5·28	5·28	5·30	5·31	5·33	3·59	3·59	3·60	3·60	3·60	3·60	3·60
35	5·65	5·68	5·70	5·73	5·74	5·76	5·75	3·82	3·83	3·83	3·83	3·83	3·83	3·83
40	6·10	6·10	6·10	6·10	6·10	6·10	6·10	4·04	4·05	4·05	4·05	4·05	4·06	4·06
45	6·55	6·51	6·49	6·47	6·46	6·45	6·44	4·27	4·27	4·27	4·27	4·27	4·28	4·28
50	6·96	6·90	6·85	6·80	6·77	6·73	6·71	4·44	4·44	4·45	4·46	4·46	4·47	4·47
55	7·38	7·30	7·24	7·15	7·09	7·02	6·98	4·64	4·64	4·64	4·65	4·65	4·67	4·69
60	7·82	7·71	7·61	7·48	7·42	7·33	7·27	4·82	4·85	4·87	4·87	4·87	4·88	4·92
65	8·27	8·14	8·01	7·87	7·76	7·67	7·59	5·00	5·05	5·05	5·07	5·08	5·13	5·18
70	8·77	8·63	8·48	8·35	8·25	8·16	8·07	5·21	5·25	5·28	5·32	5·34	5·36	5·42
75	9·30	9·15	9·01	8·87	8·77	8·67	8·58	5·43	5·47	5·48	5·54	5·56	5·61	5·68
80	9·90	9·71	9·55	9·40	9·30	9·16	9·06	5·65	5·67	5·69	5·75	5·78	5·83	5·90
85	10·90	10·50	10·27	10·06	9·93	9·77	9·57	5·85	5·89	5·90	5·96	6·00	6·06	6·13
90	11·60	11·25	11·02	10·68	10·48	10·24	9·95	6·11	6·14	6·16	6·24	6·25	6·33	6·40
95	11·91	11·58	11·36	10·98	10·77	10·51	10·17	6·46	6·50	6·52	6·61	6·67	6·71	6·84
100	12·10	11·79	11·56	11·14	10·97	10·69	10·34	—	—	—	—	—	—	—
	(iii) 0·2N-KH_2PO_4.							(iv) 0·03N-Diethylbarbituric acid.*						
5	5·86	5·86	5·86	5·86	5·86	5·86	5·86	6·78	6·61	6·51	6·35	6·25	6·13	5·99
10	6·21	6·21	6·21	6·20	6·20	6·20	6·20	7·08	6·95	6·86	6·67	6·56	6·47	6·28
15	6·45	6·44	6·42	6·42	6·42	6·42	6·40	7·27	7·16	7·05	6·89	6·74	6·64	6·47
20	6·64	6·64	6·62	6·62	6·62	6·62	6·61	7·42	7·32	7·21	7·01	6·90	6·79	6·62
25	6·82	6·81	6·80	6·80	6·80	6·80	6·79	7·56	7·44	7·33	7·14	7·03	6·91	6·73
30	6·99	6·98	6·98	6·98	6·98	6·98	6·97	7·66	7·52	7·43	7·25	7·11	7·01	6·84
35	7·17	7·17	7·16	7·16	7·15	7·15	7·15	7·73	7·63	7·53	7·33	7·22	7·11	6·93
40	7·41	7·40	7·39	7·38	7·38	7·38	7·38	7·84	7·72	7·63	7·44	7·30	7·20	7·02
45	7·75	7·75	7·74	7·74	7·74	7·74	7·74	7·91	7·81	7·74	7·52	7·39	7·28	7·11
50	9·23	9·26	9·24	9·16	9·10	9·00	8·94	8·02	7·91	7·79	7·59	7·47	7·37	7·19
55	10·84	10·74	10·61	10·37	10·23	10·07	9·84	8·10	7·97	7·90	7·67	7·56	7·45	7·27
60	11·19	11·06	10·93	10·68	10·53	10·36	10·11	8·17	8·08	7·97	7·78	7·65	7·53	7·35
65	11·43	11·27	11·13	10·88	10·71	10·53	10·29	8·27	8·15	8·06	7·87	7·73	7·61	7·45
70	11·58	11·45	11·28	10·99	10·83	10·66	10·39	8·38	8·26	8·15	7·94	7·82	7·71	7·54
75	11·74	11·56	11·41	11·10	10·92	10·75	10·49	8·48	8·36	8·25	8·05	7·93	7·84	7·66
80	11·87	11·68	11·51	11·18	11·01	10·84	10·55	8·58	8·47	8·34	8·18	8·08	7·96	7·80
85	11·98	11·78	11·61	11·26	11·09	10·89	10·63	8·72	8·61	8·48	8·33	8·21	8·09	7·94
90	12·10	11·87	11·69	11·33	11·15	10·95	10·67	8·88	8·79	8·69	8·52	8·38	8·26	8·11
95	12·20	11·98	11·76	11·38	11·20	11·00	10·71	9·20	9·06	8·93	8·77	8·67	8·52	8·35
100	12·27	12·02	11·82	11·44	11·24	11·04	10·74	—	—	—	—	—	—	—

(v) 0·2N-HBO_2.

x.	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.	x.	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.
5	7·43	7·47	7·46	7·44	7·38	7·37	7·37	55	9·38	9·29	9·20	9·09	8·99	8·96	8·86
10	7·89	7·89	7·86	7·81	7·75	7·74	7·73	60	9·48	9·37	9·31	9·18	9·10	9·04	8·94
15	8·19	8·15	8·12	8·05	8·00	7·98	7·94	65	9·59	9·48	9·39	9·28	9·20	9·15	9·04
20	8·41	8·38	8·32	8·27	8·19	8·14	8·11	70	9·68	9·59	9·50	9·39	9·31	9·23	9·15
25	8 61	8·55	8·48	8·42	8·36	8·31	8·26	75	9·77	9·68	9·59	9·49	9·41	9·34	9·25
30	8·75	8·69	8·65	8·55	8·47	8·45	8·37	80	9·91	9·81	9·72	9·62	9·54	9·47	9·36
35	8·91	8·85	8·77	8·67	8·61	8·56	8·49	85	10·06	9·95	9·89	9·78	9·68	9·60	9·49
40	9·04	8·97	8·89	8·77	8·71	8·66	8·58	90	10·21	10·14	10·03	9·91	9·83	9·75	9·64
45	9·16	9·08	8·99	8·89	8·81	8·76	8·68	95	10·52	10·40	10·32	10·16	10·07	9·93	9·80
50	9·27	9·18	9·10	8·97	8·91	8·86	8·76		—	—	—	—	—	—	—

* 0·03N-NaOH used for titration.

buffer mixture are depressed as the temperature is increased. The fact that the curves begin to diverge with the addition of the fourth equiv. is evidently due to incipient neutralisation of diethylbarbituric acid.

The very slight effect of temperature on the three dissociation constants of citric acid explains why Kolthoff and Tekelenburg (*loc. cit.*) observed that temperatures up to 60° led to an almost negligible variation in the p_H values of mixtures involving disodium hydrogen citrate, whilst the increase in K_3 of phosphoric acid accounts for the appreciable lowerings in the p_H values of disodium hydrogen phosphate–sodium hydroxide buffer mixtures at temperatures up to

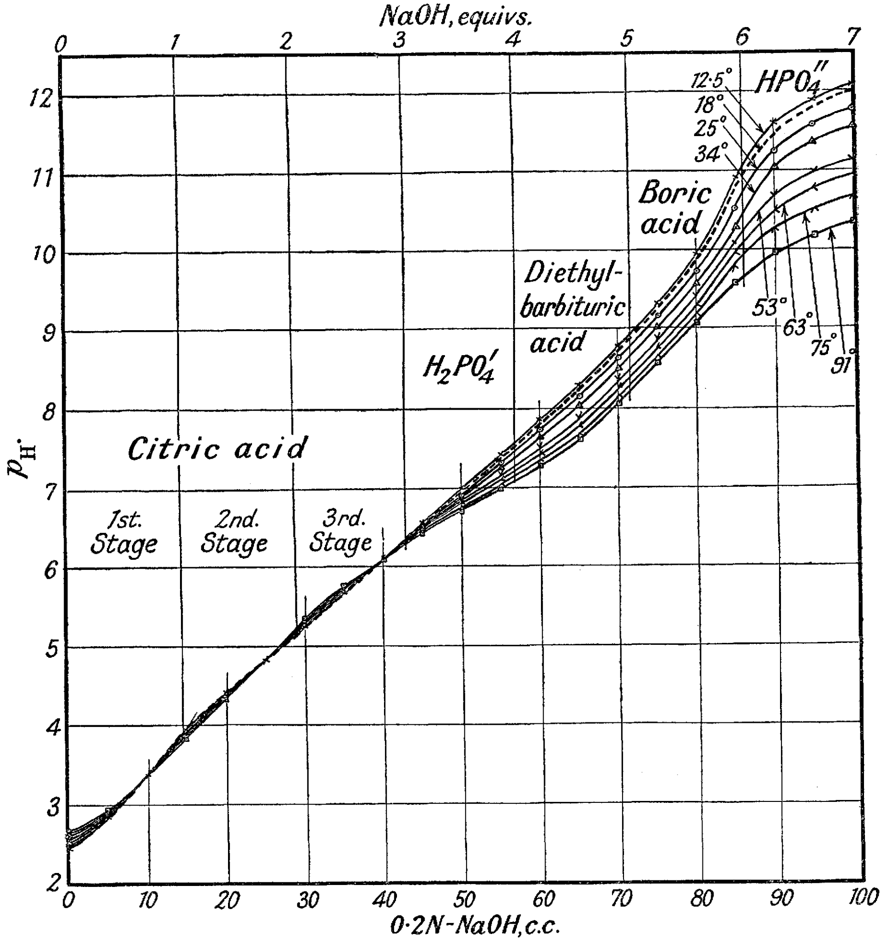

TABLE II.

(i) p_{H} Values at 0·5, 1·5, and 2·5 equivs. of NaOH per mol. of citric acid.

NaOH, mols.	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.
0·5	3·01	3·01	3·01	3·01	3·01	3·01	3·01
1·5	4·44	4·44	4·45	4·46	4·46	4·47	4·49
2·5	5·78	5·81	5·84	5·90	5·94	5·99	6·06

(ii) $p_{K_2(\mathrm{class.})}$ for $H_2PO_4' \rightleftharpoons H^{\cdot} + HPO_4''$.

Neutn., %.	12·5°.	25°.	34°.	53°.	63°.	75°.	91°.
30	6·82	6·81	6·79	6·79	6·79	6·79	6·77
50	6·82	6·81	6·80	6·80	6·80	6·80	6·79
70	6·80	6·80	6·79	6·79	6·78	6·78	6·78

(iii) $p_{K_3(\mathrm{class.})}$ for $HPO_4'' \rightleftharpoons H^{\cdot} + PO_4'''$.

25	11·85	11·71	11·64	11·47	11·46	11·41	11·32
50	11·84	11·71	11·62	11·52	11·43	11·41	11·36
75	11·85	11·66	11·60	11·51	11·46	11·38	11·37

(iv) $p_{K(\mathrm{class.})}$ for $HBO_2 \rightleftharpoons H^{\cdot} + BO_2'$.

25	9·09	9·03	8·96	8·90	8·84	8·79	8·74
50	9·27	9·18	9·10	8·97	8·91	8·86	8·76
75	9·29	9·20	9·11	9·01	8·93	8·86	8·77

(v) $p_{K(\mathrm{class.})}$ for diethylbarbituric acid.

25	8·04	7·92	7·81	7·62	7·51	7·39	7·21
50	8·02	7·91	7·79	7·59	7·47	7·37	7·19
75	8·00	7·88	7·77	7·57	7·45	7·36	7·18

60°. The gradual increase in the dissociation of boric acid with temperature similarly accounts for the lower p_H values indicated by Walbum's borax mixtures at elevated temperatures.

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