

Table 2.2.3.2. – pH of reference buffer solutions at various temperatures

Temperature (°C)	Potassium tetraoxalate 0.05 M	Potassium hydrogen tartrate saturated at 25 °C	Potassium dihydrogen citrate 0.05 M	Potassium hydrogen phthalate 0.05 M	Potassium dihydrogen phosphate 0.025 M + disodium hydrogen phosphate 0.025 M	Potassium dihydrogen phosphate 0.0087 M + disodium hydrogen phosphate 0.0303 M	Disodium tetraborate 0.01 M	Sodium carbonate 0.025 M + sodium bicarbonate 0.025 M	Calcium hydroxide, saturated at 25 °C
	$C_4H_3K_3O_{13}, 2H_2O$	$C_4H_5K_3O_6$	$C_6H_7K_2O_7$	$C_8H_5K_2O_4$	$KH_2PO_4 + Na_2HPO_4$	$KH_2PO_4 + Na_2HPO_4$	$Na_2B_4O_7, 10H_2O$	$Na_2CO_3 + NaHCO_3$	$Ca(OH)_2$
15	1.67		3.80	4.00	6.90	7.45	9.28	10.12	12.81
20	1.68		3.79	4.00	6.88	7.43	9.23	10.06	12.63
25	1.68	3.56	3.78	4.01	6.87	7.41	9.18	10.01	12.45
30	1.68	3.55	3.77	4.02	6.85	7.40	9.14	9.97	12.29
35	1.69	3.55	3.76	4.02	6.84	7.39	9.10	9.93	12.13
$\frac{\Delta pH^{(1)}}{\Delta t}$	+ 0.001	- 0.0014	- 0.0022	+ 0.0012	- 0.0028	- 0.0028	- 0.0082	- 0.0096	- 0.034

(1) pH variation per degree Celsius.

**Potassium dihydrogen phosphate 0.025 M + disodium hydrogen phosphate 0.025 M.** Dissolve 3.39 g of  $KH_2PO_4$  and 3.53 g of  $Na_2HPO_4$ , both previously dried for 2 h at  $120 \pm 2$  °C, in carbon dioxide-free water R and dilute to 1000.0 ml with the same solvent.

**Potassium dihydrogen phosphate 0.0087 M + disodium hydrogen phosphate 0.0303 M.** Dissolve 1.18 g of  $KH_2PO_4$  and 4.30 g of  $Na_2HPO_4$ , both previously dried for 2 h at  $120 \pm 2$  °C, in carbon dioxide-free water R and dilute to 1000.0 ml with the same solvent.

**Disodium tetraborate 0.01 M.** Dissolve 3.80 g of  $Na_2B_4O_7, 10H_2O$  in carbon dioxide-free water R and dilute to 1000.0 ml with the same solvent. Store protected from atmospheric carbon dioxide.

**Sodium carbonate 0.025 M + sodium hydrogen carbonate 0.025 M.** Dissolve 2.64 g of  $Na_2CO_3$  and 2.09 g of  $NaHCO_3$  in carbon dioxide-free water R and dilute to 1000.0 ml with the same solvent. Store protected from atmospheric carbon dioxide.

**Calcium hydroxide, saturated at 25 °C.** Shake an excess of calcium hydroxide R with carbon dioxide-free water R and decant at 25 °C. Store protected from atmospheric carbon dioxide.

**STORAGE**

Store buffer solutions in suitable chemically resistant, tight containers, such as type I glass bottles or plastic containers suitable for aqueous solutions.

Table 2.2.4.1

Reaction	pH	Indicator	Colour
Alkaline	> 8	Litmus paper red R	Blue
		Thymol blue solution R (0.05 ml)	Grey or violet-blue
Slightly alkaline	8.0 – 10.0	Phenolphthalein solution R (0.05 ml)	Colourless or pink
		Thymol blue solution R (0.05 ml)	Grey
Strongly alkaline	> 10	Phenolphthalein paper R	Red
		Thymol blue solution R (0.05 ml)	Violet-blue
Neutral	6.0 – 8.0	Methyl red solution R	Yellow
		Phenol red solution R (0.05 ml)	
Neutral to methyl red	4.5 – 6.0	Methyl red solution R	Orange-red
Neutral to phenolphthalein	< 8.0	Phenolphthalein solution R (0.05 ml)	Colourless; pink or red after adding 0.05 ml of 0.1 M base
Acid	< 6	Methyl red solution R	Orange or red
		Bromothymol blue solution RI	Yellow
Faintly acid	4.0 – 6.0	Methyl red solution R	Orange
		Bromocresol green solution R	Green or blue
Strongly acid	< 4	Congo red paper R	Green or blue

01/2005:20204

01/2005:20205

## 2.2.4. RELATIONSHIP BETWEEN REACTION OF SOLUTION, APPROXIMATE pH AND COLOUR OF CERTAIN INDICATORS

To 10 ml of the solution to be examined, add 0.1 ml of the indicator solution, unless otherwise prescribed in Table 2.2.4.-1.

## 2.2.5. RELATIVE DENSITY

The relative density  $d_{t_2}^{t_1}$  of a substance is the ratio of the mass of a certain volume of a substance at temperature  $t_1$  to the mass of an equal volume of water at temperature  $t_2$ .

Unless otherwise indicated, the relative density  $d_{20}^{20}$  is used. Relative density is also commonly expressed as  $d_4^{20}$ . Density  $\rho_{20}$ , defined as the mass of a unit volume of the substance at 20 °C may also be used, expressed in kilograms per cubic metre or grams per cubic centimetre

( $1 \text{ kg}\cdot\text{m}^{-3} = 10^{-3} \text{ g}\cdot\text{cm}^{-3}$ ). These quantities are related by the following equations where density is expressed in grams per cubic centimetre:

$$\rho_{20} = 0.998203 \times d_{20}^{20} \text{ or } d_{20}^{20} = 1.00180 \times \rho_{20}$$

$$\rho_{20} = 0.999972 \times d_4^{20} \text{ or } d_4^{20} = 1.00003 \times \rho_{20}$$

$$d_4^{20} = 0.998230 \times d_{20}^{20}$$

Relative density or density are measured with the precision to the number of decimals prescribed in the monograph using a density bottle (solids or liquids), a hydrostatic balance (solids), a hydrometer (liquids) or a digital density meter with an oscillating transducer (liquids and gases). When the determination is made by weighing, the buoyancy of air is disregarded, which may introduce an error of 1 unit in the 3<sup>rd</sup> decimal place. When using a density meter, the buoyancy of air has no influence.

*Oscillating transducer density meter.* The apparatus consists of:

- a U-shaped tube, usually of borosilicate glass, which contains the liquid to be examined;
- a magneto-electrical or piezo-electrical excitation system that causes the tube to oscillate as a cantilever oscillator at a characteristic frequency depending on the density of the liquid to be examined;
- a means of measuring the oscillation period ( $T$ ), which may be converted by the apparatus to give a direct reading of density, or used to calculate density using the constants  $A$  and  $B$  described below.

The resonant frequency ( $f$ ) is a function of the spring constant ( $c$ ) and the mass ( $m$ ) of the system:

$$f^2 = \frac{1}{T^2} = \frac{c}{m} \times \frac{1}{4\pi^2}$$

Hence:

$$T^2 = \left( \frac{M}{c} + \frac{\rho \times V}{c} \right) \times 4\pi^2$$

$M$  = mass of the tube,

$V$  = inner volume of the tube.

Introduction of 2 constants  $A = c/(4\pi^2 \times V)$  and  $B = M/V$ , leads to the classical equation for the oscillating transducer:

$$\rho = A \times T^2 - B$$

The constants  $A$  and  $B$  are determined by operating the instrument with the U-tube filled with 2 different samples of known density, for example, degassed *water R* and air. Control measurements are made daily using degassed *water R*. The results displayed for the control measurement using degassed *water R* shall not deviate from the reference value ( $\rho_{20} = 0.998203 \text{ g}\cdot\text{cm}^{-3}$ ,  $d_{20}^{20} = 1.000000$ ) by more than its specified error. For example, an instrument specified to  $\pm 0.0001 \text{ g}\cdot\text{cm}^{-3}$  shall display  $0.9982 \pm 0.0001 \text{ g}\cdot\text{cm}^{-3}$  in order to be suitable for further measurement. Otherwise a re-adjustment is necessary. Calibration with certified reference materials is carried out regularly. Measurements are made using the same procedure as for calibration. The liquid to be examined is equilibrated in a thermostat at  $20 \text{ }^\circ\text{C}$  before introduction into the tube, if necessary, to avoid the formation of bubbles and to reduce the time required for measurement.

Factors affecting accuracy include:

- temperature uniformity throughout the tube,
- non-linearity over a range of density,
- parasitic resonant effects,
- viscosity, whereby solutions with a higher viscosity than the calibrant have a density that is apparently higher than the true value.

The effects of non-linearity and viscosity may be avoided by using calibrants that have density and viscosity close to those of the liquid to be examined ( $\pm 5$  per cent for density,  $\pm 50$  per cent for viscosity). The density meter may have functions for automatic viscosity correction and for correction of errors arising from temperature changes and non-linearity.

Precision is a function of the repeatability and stability of the oscillator frequency, which is dependent on the stability of the volume, mass and spring constant of the cell.

Density meters are able to achieve measurements with an error of the order of  $1 \times 10^{-3} \text{ g}\cdot\text{cm}^{-3}$  to  $1 \times 10^{-5} \text{ g}\cdot\text{cm}^{-3}$  and a repeatability of  $1 \times 10^{-4} \text{ g}\cdot\text{cm}^{-3}$  to  $1 \times 10^{-6} \text{ g}\cdot\text{cm}^{-3}$ .

01/2005:20206

## 2.2.6. REFRACTIVE INDEX

The refractive index of a medium with reference to air is equal to the ratio of the sine of the angle of incidence of a beam of light in air to the sine of the angle of refraction of the refracted beam in the given medium.

Unless otherwise prescribed, the refractive index is measured at  $20 \pm 0.5 \text{ }^\circ\text{C}$ , with reference to the wavelength of the D-line of sodium ( $\lambda = 589.3 \text{ nm}$ ); the symbol is then  $n_{\text{D}}^{20}$ .

Refractometers normally determine the critical angle. In such apparatus the essential part is a prism of known refractive index in contact with the liquid to be examined.

Calibrate the apparatus using certified reference materials.

When white light is used, the refractometer is provided with a compensating system. The apparatus gives readings accurate to at least the third decimal place and is provided with a means of operation at the temperature prescribed. The thermometer is graduated at intervals of  $0.5 \text{ }^\circ\text{C}$  or less.

01/2005:20207

## 2.2.7. OPTICAL ROTATION

Optical rotation is the property displayed by chiral substances of rotating the plane of polarisation of polarised light.

Optical rotation is considered to be positive (+) for dextrorotatory substances (i.e. those that rotate the plane of polarisation in a clockwise direction) and negative (–) for laevorotatory substances.

The specific optical rotation  $[\alpha_m]_{\lambda}^t$  is the rotation, expressed in radians (rad), measured at the temperature  $t$  and at the wavelength  $\lambda$  given by a 1 m thickness of liquid or a solution containing  $1 \text{ kg}/\text{m}^3$  of optically active substance. For practical reasons the specific optical rotation  $[\alpha_m]_{\lambda}^t$  is normally expressed in milliradians metre squared per kilogram ( $\text{mrad}\cdot\text{m}^2\cdot\text{kg}^{-1}$ ).

The Pharmacopoeia adopts the following conventional definitions.

The *angle of optical rotation* of a neat liquid is the angle of rotation  $\alpha$ , expressed in degrees ( $^\circ$ ), of the plane of polarisation at the wavelength of the D-line of sodium ( $\lambda = 589.3 \text{ nm}$ ) measured at  $20 \text{ }^\circ\text{C}$  using a layer of 1 dm; for a solution, the method of preparation is prescribed in the monograph.