

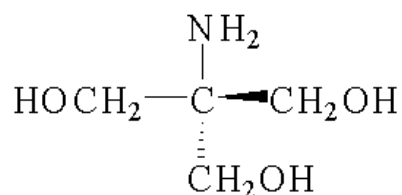
## Product Information

### Tris(hydroxymethyl)aminomethane; Tris

Technical Bulletin No. 106B

#### IMPORTANT NOTICE

The "Tris" described in this bulletin is Tris(hydroxymethyl)aminomethane. It is not the "Tris" used to flame-proof fabric. The latter, Tris(2,3-dibromopropyl)phosphate, has been reported to be a cancer suspect agent.



Trizma<sup>®</sup> is Sigma's registered trademark applied to various compounds of tris(hydroxymethyl)-aminomethane (tris) prepared by Sigma. For example, Trizma HCl is the completely neutralized crystalline hydrochloride salt of tris. Trizma Base is the pure tris itself.

Trizma and its salts have been useful as buffers in a wide variety of biological systems. Uses include pH control *in vitro*<sup>1,2</sup> and *in vivo*,<sup>3,4</sup> for body fluids and as an alkalizing agent in the treatment of acidosis of the blood.<sup>5</sup> Trizma has been used as a starting material for polymers, oxazolones (with carboxylic acids) and oxazolidines (with aldehydes).<sup>6</sup> Trizma does not precipitate calcium salts and is of value in maintaining solubility of manganese salts.<sup>7</sup>

#### PRIMARY BASIMETRIC STANDARD

Trizma Base meets many of the requirements for a primary basimetric standard. It is pure, essentially stable, relatively non-hygroscopic and has a high equivalent weight. Trizma Base can be dried at 100°C for up to 4 hours.<sup>8</sup> It can be used for the direct standardization of a strong acid solution; the equivalence point can be determined either potentiometrically or by use of a suitable indicator: [3-(4-Dimethylamino-1-naphthylazo)-4-methoxybenzenesulfonic acid, Product No. D5407].

#### Tris(hydroxymethyl)aminomethane (Trizma Base) -- Physical Data

Empirical Formula: C<sub>4</sub> H<sub>11</sub> N O<sub>3</sub>

Molecular Weight: 121.14

Equivalent Weight: 121.14

pH of 0.05 M aqueous solution: 10.4\*

K<sub>b</sub> = 1.202 × 10<sup>-6</sup> at 25° (pK<sub>a</sub> = 8.08)<sup>9</sup>

### Trizma HCl – Physical Data

Empirical Formula: C<sub>4</sub> H<sub>12</sub> Cl N O<sub>3</sub>

Molecular Weight: 157.6

Equivalent Weight: 157.6

pH of 0.05 M aqueous solution: 4.7\*

\*Trizma HCl in solution will produce a pH of approximately 4.7, but will have little if any buffering capacity. Trizma Base in solution will produce a pH of approximately 10.4, but will have little if any buffering capacity. Blending Trizma Base and Trizma HCl will produce any desired pH between 7 and 9, with a reasonable buffering capacity.

### Available Grades

Trizma Base and Trizma HCl are both available in Molecular Biology, Electrophoresis and SigmaUltra grades. Additionally, Trizma Base is available in Tissue Culture, Plant Tissue Culture and ACS grades, as well as a grade that meets all analytical and packaging requirements as per USP. Sigma also offers a variety of salts of Trizma Base.

### EFFECTS OF TEMPERATURE ON pH

In general, as the solution decreases in temperature from 25°C to 5°C, the pH increases an average of 0.03 pH units per °C. As the solution increases in temperature from 25°C to 37°C, the pH decreases an average of 0.025 pH units per °C.

### EFFECTS OF CONCENTRATION ON pH

Increasing the total tris concentration from 0.05 M to 0.5 M will increase the pH by about 0.05 pH units. Decreasing the concentration from 0.05 M to 0.005 M will decrease the pH by about 0.05 pH units.

If Trizma components are thoroughly desiccated, it is possible to weigh the correct ratio and achieve a predicted pH with an accuracy of  $\pm 0.05$ . Thus, under careful conditions, the use of a pH meter for confirming the pH is unnecessary. The [Mixing Table](#) below shows the proportions to use to obtain a desired pH.

### STABILITY

Our experience indicates that Trizma buffer solutions can be autoclaved.

### TRIZMA "PRESET pH" COMPOUNDS

Sigma offers "Preset pH" Trizma dry mixtures from which pH-specific solutions may be prepared without adjustment. For example, Trizma 8.2 will yield a pH of 8.2 ( $\pm 0.05$ ) when dissolved in water to a concentration of 0.05 M at 25°C. Each package of Trizma Preset pH is labeled with its average molecular weight and the pHs which will result at 5°, 25°, and 37°C at a concentration of 0.05 M. See the [Mixing Table](#) below for the Product Nos. and pHs available.

### HOW TO MAKE A BUFFER SOLUTION

If a pH meter can be used, tris HCl buffers are usually prepared by "titrating" Trizma Base with hydrochloric acid. The Trizma Base is dissolved in water nearly to volume; a solution of hydrochloric acid is gradually added to achieve the desired pH, and the resulting tris HCl buffer is diluted to the appropriate final volume.

Alternatively, the following table also specifies the amounts of Trizma HCl and Trizma Base required to prepare 0.05 M buffer solutions at various pH and temperature conditions. To obtain the desired 0.05 M buffer, dissolve and dilute the indicated quantities of Trizma HCl and Trizma Base to one liter with water.

**TRIZMA MIXING TABLE**

Product Number	Trizma Preset Compound	pH at 5°C	pH at 25°C	pH at 37°C	Grams/liter for 0.05 M Tris HCl	Grams/liter for 0.05 M Tris Base
T3503	7.0	7.55	7.00	6.70	7.28	0.47
T3628	7.1	7.66	7.10	6.80	7.13	0.57
T3753	7.2	7.76	7.20	6.91	7.02	0.67
T3878	7.3	7.89	7.30	7.02	6.85	0.80
T4003	7.4	7.97	7.40	7.12	6.61	0.97
T4128	7.5	8.07	7.50	7.22	6.35	1.18
T4253	7.6	8.18	7.60	7.30	6.06	1.39
T4378	7.7	8.26	7.70	7.40	5.72	1.66
T4503	7.8	8.37	7.80	7.52	5.32	1.97
T4628	7.9	8.48	7.90	7.62	4.88	2.30
T4753	8.0	8.58	8.00	7.71	4.44	2.65
T4878	8.1	8.68	8.10	7.80	4.02	2.97
T5003	8.2	8.78	8.20	7.91	3.54	3.34
T5128	8.3	8.88	8.30	8.01	3.07	3.70
T5253	8.4	8.98	8.40	8.10	2.64	4.03
T5378	8.5	9.09	8.50	8.22	2.21	4.36
T5503	8.6	9.18	8.60	8.31	1.83	4.65
T5628	8.7	9.28	8.70	8.42	1.50	4.90
T5753	8.8	9.36	8.80	8.51	1.23	5.13
T5878	8.9	9.47	8.90	8.62	0.96	5.32
T6003	9.0	9.56	9.00	8.70	0.76	5.47

Product Number	Trizma Preset Compound	pH at 5°C	pH at 25°C	pH at 37°C	Grams/liter for 0.05 M Tris HCl	Grams/liter for 0.05 M Tris Base
T6128	9.1	9.67	9.10	8.79	0.69	5.53

Also, by using an appropriate weak acid with Trizma Base to make pH adjustments, buffers over an extended range may be prepared. Please consult our catalog for extensive listings.

### TRIZMA FISH GRADES

Trizma type 7.2 FT (Product No. T8508), Trizma type 7.4 FT (Product No. T1753) and Trizma type 8.3 FT (Product No. T1878), are prepared by Sigma for experimental use in water conditioning for aquariums and transport of fish. Many varieties of tropical fish have been found to respond well to transportation in Trizma buffered water, in closed and open systems, for periods ranging from 12 hours to several weeks. Trizma 7.2 and 7.4 are offered for use with fresh-water fish and will produce a pH of approximately 7.2 and 7.4, respectively, with many natural fish waters. Trizma 8.3 is prepared for use with marine species and will produce a pH of approximately 8.3.

Experiments with Trizma buffered waters in marine fish transport indicate that a concentration of from 10 to 30 grams (1/3 to 1 ounce) per gallon of water is effective in preventing significant changes in pH and markedly depresses CO<sub>2</sub> buildup over considerable periods.<sup>7</sup> Mortality of the fish was greatly reduced. Also, putrefication of dead fish was retarded. Since CO<sub>2</sub> control alone does not seem to explain the significant results, Trizma apparently has other desirable properties. We have been told that concentrations closer to 30 grams (1 ounce) per gallon of water are preferable for many species.<sup>10</sup>

Some breeders and shippers of fish prefer to operate at pH levels other than those provided by Trizma 7.2, 7.4 or 8.3. The weights of Trizma HCl and Trizma Base necessary to produce any desired pH between 7.0 and 9.1 are given in the [Trizma Mixing Table](#) above.

Note: Although reports indicate that Trizma is safe in the concentrations indicated, anyone planning to use it should conduct tests to validate the safety of this application.

### ELECTRODES

Reports indicate that certain pH electrodes do not give accurate pH readings when used with tris buffers.<sup>11,12</sup> This is believed to be caused by a reaction between tris and the linen fiber junction of the reference electrode, resulting in high liquid-junction potentials and long equilibration times.

The Leeds and Northrup Company cautions about using silver/silver chloride reference electrodes in tris solutions containing proteins. The interaction between the influxing silver ions and the proteins can cause spurious pH readings. Consult your electrode manufacturer if there is any uncertainty concerning the compatibility of your electrodes with tris buffer.

Sigma offers a Glass-Calomel Combination Electrode for use with tris solutions through a pH range of 0-12 and a temperature range of -5°C to 80°C. This electrode eliminates most errors frequently associated with pH measurements of tris solutions. It is supplied with a standard 30-inch lead, a choice of three diameters (4mm, 6mm, 8mm), and a choice of four connectors (ferrule-pin, BNC coaxial, Radiometer after 1975 and/or combination Belling-Lee coaxial). Other cable lengths, connectors and adapters may be available on a special-

order basis.

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