

6.2 Calibration Standards



All hygroscopic chemicals in crystal form should be dried in an oven set at 105°C for 3 hours to remove any traces of absorbed water. All prepared solutions must be mixed thoroughly with a magnetic stirrer or inverted manually at least ten times or until all crystals are completely dissolved inside the solution.

The following compounds can be used to prepare calibration standard solutions in BioTector.

To prepare a 1000mgC/l Total Organic Carbon (TOC) standard solution, use one of the following:

- Potassium Hydrogen Phthalate, $C_8H_5KO_4$, 2.13g (99.9% purity) in one liter of deionised water. Water solubility: 80 g/L at 20°C.
- Acetic acid, $C_2H_4O_2$, 2.51g (99.8% purity) in one liter of deionised water. Water solubility: Miscible in all proportions.
- Glucose, $C_6H_{12}O_6$, 2.53g (99% purity) in one liter of deionised water. Water solubility: 512g/L at 25°C.

To prepare a 1000mgC/l Total Inorganic Carbon (TIC) standard solution, use one of the following:

- Sodium Carbonate, CNa_2O_3 , 8.84g (99.9% purity) in one liter of deionised water.
- Sodium Hydrogen Carbonate, $CHNaO_3$, 7.04g (99.5% purity) in one liter of deionised water.
- Potassium Carbonate, CK_2O_3 , 11.62g (99.0% purity) in one liter of deionised water.

To prepare a 1000mgN/l Total Nitrogen (TN) standard solution, use one of the following:

- Nitric Acid, HNO_3 , 6.43g (70% purity) in one liter of deionised water.
- Cesium Nitrate, $CsNO_3$, 14.05g (99% purity) in one liter of deionised water.
- Sodium Nitrate, $NaNO_3$, 6.07g (99% purity) in one liter of deionised water.

To prepare a 1000mgP/l Total Phosphorus (TP) standard solution, use one of the following:

- Potassium Dihydrogen Phosphate, H_2KPO_4 , 4.43g (99% purity) in one liter of deionised water.
- Phosphoric Acid, H_3PO_4 , 3.72g (85% purity) in one liter of deionised water.



The quantity of concentrated chemical required to prepare stock calibration solutions will change with the % purity of the chemical used. If the purity of the chemical is different than the figures displayed above, the necessary quantity needs to be recalculated from the purity of the chemical. See example in the following page.



Depending on the system analysis ranges (see System Range Data menu), every BioTector requires specific calibration standard solutions. The required concentration of the calibration standard solutions can be identified in Span Calibration menu. As the sample pressure normalization (line equalization function) is not activated for Calibration/Manual Valve, it is recommended that the calibration standard solution flask is placed at the same height with the Sample Pump.

The calculation of the quantities required to prepare Potassium Hydrogen Phthalate (KHP) standard solutions with various purities are given as an example below:

Name:	Potassium Hydrogen Phthalate		
Formula:	C ₈ H ₅ KO ₄		
Carbon, 12	x8	=	96
Oxygen, 16	x4	=	64
Potassium, 39	x 1	=	39
Hydrogen, 1	x5	=	5
Total weight		=	204.22 g/mol

47% of KHP is Carbon. Purity of the KHP is 99.9%. Therefore, to prepare a 1000 mgC/l standard solution, add 2.13g of KHP in a flask and add enough deionised water to make it exactly 1 liter solution.

Note that the quantities required change with the % purity of the chemical used. Table 10 below gives the KHP quantities required at various % purity for the preparation of 1000mgC/l calibration standard.

Table 10 Quantity of KHP required to prepare 1000 mgC/l standard at various purities.

% Purity of KHP	Quantity of KHP (grams)
	to prepare 1000 mgC/l Standard
100	2.127
99.9	2.129
99.5	2.138
99.0	2.149
95.0	2.239
90.0	2.364

To prepare standard solutions containing more than 1000 mgC/l, the required solvent can be mixed directly with deionised water. Table 11 below gives the required quantity of KHP for various concentration standard solutions to be mixed with deionised water and added enough deionised water to make the solution exactly 1 liter.

Table 11 Quantity of KHP required to prepare various concentration TOC standard solutions.

TOC Standard Solution Concentration (mgC/l)	Quantity of 99.9% KHP (grams) to be added 1 Liter DI Water
1000	2.129
1250	2.661
1500	3.194
2000	4.258
5000	10.645
10000	21.290

Preparation of Calibration Standard Solutions:



Use eye protection and gloves.

Standards solutions greater than 1000mg/l can be prepared directly without any dilution by simply mixing the necessary quantity solvent or salt with deionised water. Standard solutions below 1000mg/l concentration should be prepared by dilution technique. First a 1000mg/l standard stock solution should be prepared, and then the required lower concentration standard solution should be prepared by applying the necessary dilution procedures:

- For example, to prepare a 50mgC/l TOC only standard solution, first weigh 50 grams of the 1000mgC/l stock standard. Add 50 grams of the 1000mgC/l standard into a one-liter flask and add enough deionised water to make the solution exactly 1 liter.
- For example, to prepare a 50mgC/l TOC and 10 mgN/l TN mixture standard, add 50 grams of the 1000mgC/l standard and 10 grams of the 1000mgN/l standard together into a one-liter flask, and add enough deionised water to make the solution exactly 1 liter.
- For example, to prepare a 50mgC/l TOC, 10 mgN/l TN and 5 mgP/l TP mixture standard, add 50 grams of the 1000mgC/l standard, add 10 grams of the 1000mgN/l standard and add 5 grams of the 1000mgP/l standard together into a one-liter flask, and finally add enough deionised water to make the solution exactly 1 liter.
- For increased accuracy, standard solutions below 5mg/l (ppm) concentration should be prepared with two or more steps dilution. For example, to prepare a 1mgC/l standard, first prepare a 100mgC/l standard by adding 100 grams of the 1000mgC/l standard into a one-liter flask and by adding enough deionised water to make it exactly a 1 liter solution. Then add 10 grams of the 100mgC/l standard into a one-liter flask and add enough deionised water to make it exactly 1 liter.
- Standard solutions at $\mu\text{g/l}$ (ppb) levels should be prepared with several dilution steps. For instance, a 1mgC/l (1000 $\mu\text{g/l}$) standard should be prepared with two or more steps dilution as described above. To prepare a 50 $\mu\text{g/l}$ standard, add 50 grams of the 1000 $\mu\text{g/l}$ standard into a one-liter flask, and add enough deionised water to make it exactly 1 liter.

Shelf Life and Storage of Calibration Standard Solutions:

- ❖ TOC standards prepared from Potassium Hydrogen Phthalate is typically stable for a month once it is kept in a closed glass container and refrigerated at 4°C.
- ❖ All other standards such as TOC prepared from Acetic Acid, TIC, TN and TP standard solutions are recommended to be used within 48 hours of manufacture.