Gentodenz

ref19-DENZ-50, ref19-DENZ-500

Density gradient medium for cell separation.

Applications for Gentodenz:

Separations based on density differences: fractionation of nucleic acids, proteins, polysaccarides and nucleoproteins.
Subcellular organelles can be successfully isolated on gradients of Gentodenz under either isotonic or mildly hypertonic conditions.
Separation of intact living cells.
Useful in the isolation and purification of viruses and bacteria.

Formation of gradients:

Gradients of Gentodenz can be generated as follows:
1. Formed in situ by centrifugation (self-forming gradients).
2. Layering solutions of the desired concentration into an appropriate centrifuge tube and allowing the solutions to diffuse. Using Gentodenz isotonic solution gradients can be simply prepared in a hour.
3. Freezing and thawing.
4. Gradients mixers

Extended uses:

In literature the product, the applications, the procedures are described.
Over more than 30 years the density gradient compound has been sold by several commercial companies under different trade names.

Chemical info:

Structure: non-ionic tri-iodinated derivative of benzic acid with 3 aliphatic hydrophilic side chains.
Chemical name: 5- (N-2, 3-dihydroxypropylactamido)-2, 4, 6-tri-iodo-N,N' –bis (2, 3 dihydroxypropyl) isophthalamide.

Purity: analytical grade	min 99.20 %
Molecular weight	821.14
Density	2.1 g/ml

Product description:

Gentodenz is non-ionc, non-toxic and is very water soluble.
Solutions up to 80 % (w/v) can be prepared. Aqueous solutions have a very high water activity. Most particles will are fully hydrated in solutions of Gentodenz and will band at a low density.
Solutions of Gentodenz are stable to heat and may be autoclaved, stability to autoclaving (enhanced by the addition of small amounts of Tris and EDTA.

Solutions of Gentodenz are very resistant to bacterial degradation and Gentodenz is not metabolized by mammalian cells.

The concentration and density of solutions of Gentodenz can easily be determined by measuring the refractive index.
The relationship between concentration, refractive index (ŋ) and density is linear and can be formulate:

Concentration, % (w/v)	= 607.75 ŋ – 810.13
Density (g/ml)	= 3.242 ŋ – 3.323

(Before using this equation the refractive index must be corrected for the presence of buffer or salt in the gradient medium.)

Gentodenz is a non-particulate medium;therefore the distribution of cells in a gradient can be determined using a haemocytometer, electronic particle counter or by light-scattering measurements using a spectrophotometer.

Gentodenz is also soluble in formamide and dimethyl formamide, for
non-aqueous denaturing gradients of Gentodenz.

Stability and storage:

Gentodenz in solid form is stable for a period of 5 years when stored at room temperature and protected from light. Gentodenz in solution is stable for 5 years provided that it is kept sterile and protected from light. Prolonged exposure to direct sunkight leads to release of iodine from the molecule. This effect is negligible when working with these solutions on a day to day basis.

Isotonic Gradients

Gradients solution for the preparation of essentially iso-osmotic Gentodenz gradients can be prepared using an iso-osmotic solution of Gentodenz which contains 27.6% (w/v) Gentodenz (density=1.15 g/ml) made up in buffered medium.
This solution may be diluted to desired concentration by using a buffered diluent containing either sucrose or NaCl as osmotic balancer.
The composition of these diluents is as follows: 0.75 g NaCl or 7.45 g sucrose Dissolved in 100 ml 5 mmol/Tris-HCl (pH 7.5) containing 3 mmol/l KCl and 0.3 mmol/l CaNa2 EDTA.
The relationship between density and refractive index (ŋ) can be formulated:

NaCl diluent	Sucrose diluent
Density = 3.287 ŋ – 3.383	Density = 3.410 ŋ – 3.555

Compatibility with some widely used assays:

Gentodenz does not interfere with the orcinol and diphenylamine reactions for estimation of nucleic acids, nor with the very sensitive dyebinding assays for protein and DNA.

Polysaccharides and sugars can be determined in the presence of Gentodenz using the phenol/H2SO4 assay.
Fluorimetric assays of nucleic acid and proteins can also be carried out in the presence of Gentodenz.
Gentodenz does not interfere with most assays for the marker enzymes of subcellular components, also most commercial scintillants are compatible with Gentodenz.

Removal of Gentodenz from samples:

Gentodenz can be removed from samples by dialysis, ultrafiltration and gel filtration. Cells, subcellular organelles and other particulate matter can be separated from Gentodenz by centrigugation without the risk of contaminating the pellet with Gentodenz.
Gentodenz is readily soluble in both acidic and ethanolic media. Thus in a number of cases samples can be isolated free of Gentodenz by precipitating the sample with trichloroacetic or ethanol.

Ordering information: