RediGrad

Instructions

RediGrad™ is composed of colloidal silica covalently coated with silane. Centrifugation of RediGrad results in spontaneous formation of a density gradient due to the heterogeneity of particle sizes in the medium.

RediGrad can be used for formation of gradients either by the use of convenient gradient mixers or by high speed centrifugation. In the latter case, the sample can be pre-mixed with the medium and then separated on the gradient created *in situ*. In this way, gradient formation and sample separation can be achieved in a single operation.

Intended use

RediGrad is a medium for cell preparation by density gradient centrifugation.

The products are intended for research use only, and shall not be used in any clinical or *in vitro* procedures for diagnostic purposes. Where your procedures require sterile products, do not use RediGrad.

Safety

For use and handling of the products in a safe way, please refer to the Safety Data Sheet.

Physical properties

RediGrad has the following combination of properties[1]:

- Low osmolality (max. 30 mOsm/kg H_2O) permitting precise adjustment to physiological conditions without significant interference from the medium.
- Impermeable to biological membranes, resulting in no change of buoyant density of particles during centrifugation.
- Spontaneous formation of gradient during centrifugation, allowing mixing of large sample volumes in the centrifuge tubes.
- Low viscosity (max. 15 cP) resulting in rapid formation of gradients and particle separation.

Properties	RediGrad
Composition	Silica sol with covalently linked silane
Density (g/ml)	1.130 ± 0.005
Osmolality (mOsm/kg H_2O)	max. 30
Viscosity (cP)	max. 15 at 20°C
pH	9.4 ± 0.5 at 20°C to 25°C
Endotoxin (EU/ml)	max. 2

1 Data on File.

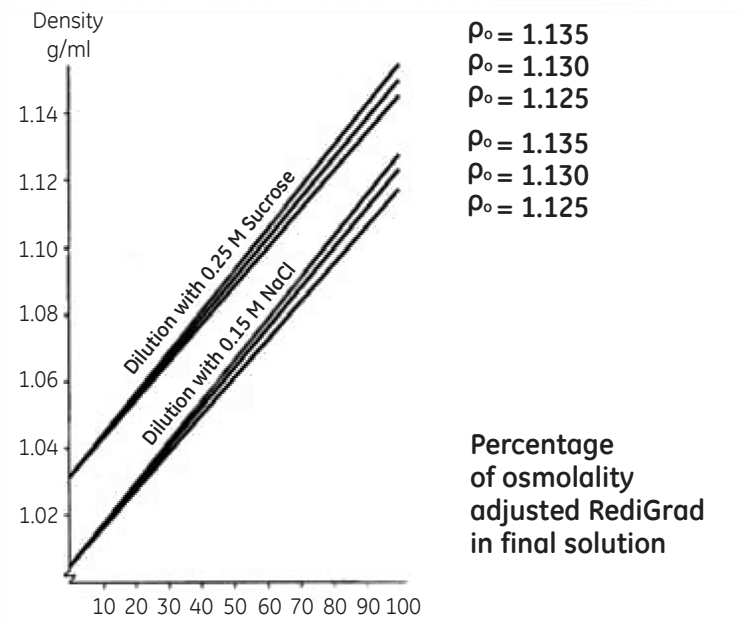

Figure 1: Dilution of stock osmolality adjusted RediGrad (340 mOsm/kg H_2O) with saline or sucrose solution. ρ_0 is the density of RediGrad from the bottle.

Instructions for use

Preparation of gradient material

RediGrad is best used in balanced salt solutions, physiological saline, or 0.25 M sucrose.

The low osmolality of RediGrad permits this parameter to be controlled by the user without significant interference from the density medium itself. The addition of 9 parts (v/v) of RediGrad to 1 part (v/v) of either 1.5 M NaCl, 10× concentrated cell culture medium, or 2.5 M sucrose will result in a solution adjusted to about 340 mOsm/kg H_2O. Solutions of different osmotic pressure can be produced by adjusting the relative volumes of RediGrad and salt or sucrose solution [1]. The final adjustment to the required osmolality can be carried out by the addition of salts or distilled water. When precise osmotic pressures are required, it is recommended that the osmolality of the solutions be measured in an osmometer. Concentrations other than 10× physiological saline may also be used satisfactorily[2].

1 Vincent, R., Nadeau, D. Anal. Biochem, 141 (1984) 322–328
2 Timonen, T., Reynolds, C.W., Ortaldo, J.R., et al. J. Immunol. Methods 51 (1982) 269–277

Centrifugation with RediGrad

RediGrad will form self-generated gradients by centrifugation at approximately 10,000 g_{av} (in 0.15 M saline) or 25,000 g_{av} (in 0.25 M sucrose) in fixed-angle rotor heads after 15 minutes.

Density determination of RediGrad gradients

Measurement of the density of RediGrad solutions after gradient fractionation can be carried out easily using a refractometer. Refractive index has a linear correlation with the density of a RediGrad solution.

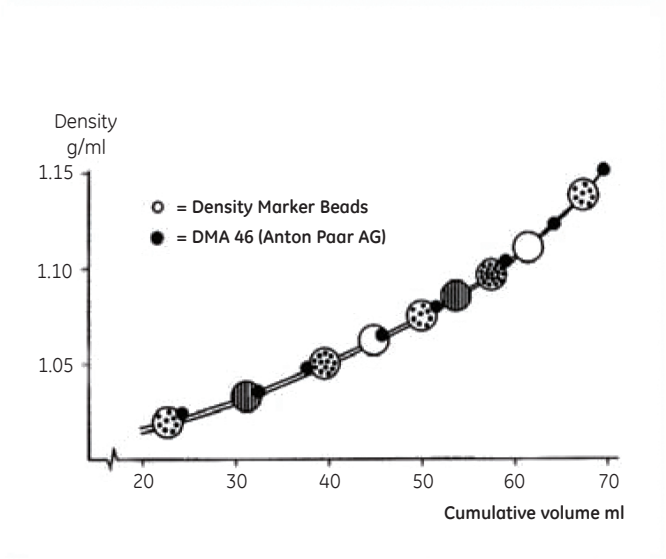

Figure 2: Correlation of recorded densities of a RediGrad gradient in 0.15 M NaCl using Density Marker Beads and a digital densitometer (DMA 46, Anton Paar A.G.).

Removal of RediGrad after centrifugation

If it is desirable to remove the gradient medium from the biological material, this may be performed by following one of the procedures outlined below.

- Cells can be recovered free from particles of RediGrad by dilution with physiological saline and centrifugation to collect the cells.

- Gel filtration or ion exchange chromatography can also be used to separate biological material from RediGrad.

Point for practical use

Storage

RediGrad may be stored unopened for up to 5 years at room temperature. When opened, it should be stored at 2°C to 8°C.

Care and cleaning of equipment

Solutions containing silica usually give a pellet at the bottom of the centrifuge tube and deposits of silica on the walls of tubing used for fractionation. These deposits may be difficult to remove when dry and it is recommended that all equipment is washed thoroughly immediately after use. Spillage of RediGrad can be removed by washing with water.

Ordering information

Related products	Quantity	Code No
Percoll™ PLUS	250 ml	17544502
Percoll PLUS	1 l	17544501
Percoll	250 ml	17089102
Percoll	1 l	17089101
Percoll	6 x 1 l	17089109
Ficoll-Paque™ PLUS	6x100 ml	17144002
Ficoll-Paque PL	6x500 ml	17144003
Ficoll-Paque PREMIUM	6x100 ml	17544202
Ficoll-Paque PREMIUM	6x500 ml	17544203
Ficoll-Paque PREMIUM 1.084	6x100 ml	17544602
Ficoll-Paque PREMIUM 1.073	6x100 ml	17544652

Related literature	Code No
Cell Separation Media, Methodology and applications	18111569
Isolation of mononuclear cells, Methodology and applications	18115269

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