himac APPLICATION
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Purification of plasmid DNA using S-cap and ultracentrifuge

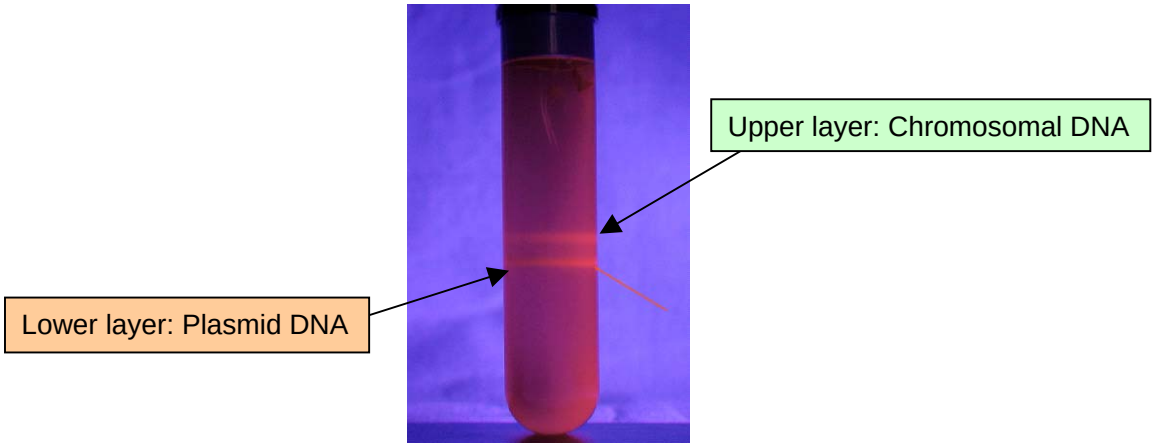
CP-WX series preparative ultracentrifuge/S-12AL cap

The 12PA tubes and the 40PA tubes are popularly used for purification of plasmid DNA by means of an ultracentrifuge because they are thin and the separated samples can be easily collected. However, the load applied to the tube cap is high due to the centrifugation at the RCF as high as hundreds of thousands of x g and thus the number of components is as many as six and the setting method is very complicated. To meet the customers' demand for the simpler tube cap, we developed the "3 & 3" S-Cap that requires only 3 components and 3-step operation for setting. Following is our experiment report on purification of plasmid DNA by means of the S-Cap.

Experiment

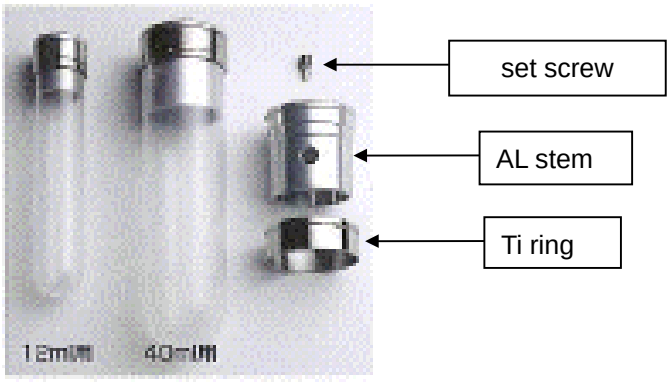
1. Conditions for centrifugation
   Centrifuge: CP100WX preparative ultracentrifuge
   Rotor: P70AT2 angle rotor (12 ml x 12)
   Tube: 12PA tube
   Cap: S-12AL cap
   Speed: 55,000 rpm (Maximum RCF: 279,000 x g)
   Time: About 17 hours
   Temperature: 20 °C
   Sample: TE solution containing coarsely extracted plasmid DNA: 7.3 ml
   Cesium chloride: 7.0 g
   Ethidium bromide (10 mg/ml): 210 µl

2. Result



Features of S-Cap

- **<3 & 3>** ------ Only 3 components (stem, ring and screw) and 3-step operation for setting




hand press

Setting the S-12AL cap to the 12PA tube

1. Put the AL stem on the tube filled with 7.5 ml of sample. Fit the AL stem in the tube with a hand press.

2. Put the Ti ring on the tube from the bottom and press-fit it with a hand press.

3. Inject the remainder of the sample into the center of the AL stem with a syringe and tighten the set screw.

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Plasmid DNA fractionation

1. Remove the set screw from the AL stem.

2. Stick the syringe needle (22 G) into the side of the tube and suck the lower-layer band (see the photo on the previous page).

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Removal of the S-cap

1. Screw the tube setter into the threaded portion at the center of the AL stem.

2. Hold the circumference of the Ti ring securely by hand. Pull up the AL stem shaking the tube setter gently from side to side.

For more information, visit our website at:
http://www.hitachi-koki.com/himac.contact/index.htm

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