



PureCep™ Sperm Wash Media Kit

For laboratory procedures only; other uses must be qualified by the end user.

Product Description	REF Number	Unit Size
PureCep™ Sperm Wash Media Kit	PA-PC80-K	12 x 12 mL
PureCep™ Sperm Wash Media Kit	PA-PC80-K4	3 x 12 mL

DESCRIPTION

An 80% (v/v) sterile colloidal suspension of silica particles stabilized with covalently bound hydrophilic silane supplied with HEPES-buffered Human Tubal Fluid (HTF). The components of this kit will allow for the highly efficient separation of motile sperm from the ejaculate of most semen specimens. It is fast, cost-effective, and needs very little equipment or expertise to perform. This is a preferred method for normozoospermic semen specimens.

MATERIALS PROVIDED IN THE SPERM SEPARATION MEDIA KIT

1. PureCep™ Gradient:

80% PureCep™ with HEPES-buffered Human Tubal Fluid (HTF-HEPES) with Human Serum Albumin, 5 mg/mL.

2. PureCep™ Sperm Washing Medium:

HEPES-buffered Human Tubal Fluid with Human Serum Albumin, 5 mg/mL.

PACKAGING, STORAGE, AND EXPIRATION

PA-PC80-K is assembled with 8 x 12-mL PureCep™ 80% (v/v) (REF #LPHTF-B) and 4 x 12-mL PureCep™ Sperm Washing Medium (REF #SWHTF-A).

PA-PC80-K4 is assembled with 2 x 12-mL PureCep™ 80% (v/v) and 1 x 12-mL PureCep™ Sperm Washing Medium (REF #SWHTF-A).

All PureCep™ components must be stored at 2°C to 8°C. The product is stable until the expiration date shown on the label or within 30 days of the Date of First Use provided that proper aseptic procedures have been observed by the user.

- Remove desired volume of product using aseptic procedures.
- Once product has been removed from the original container, reseal the container to ensure a tight seal. Write the date the product was first opened on the product label. Do not use product longer than 30 days after opening the container.
- Once removed, do not return any volume of product to the original container.
- Once the product has been opened, store the sealed container at 2°C to 8°C.
- Do not use if the product becomes discolored, cloudy, turbid, or shows any evidence of microbial contamination.

One-cell MEA tested and passed with 80% or greater blastocyst. USP Endotoxin gel clot tested and passed with <1 EU/mL.

A Certificate of Analysis is available for this product.

USER QUALITY CONTROL

- Examine the PureCep™ solutions. Do not use any medium that appears discolored. The PureCep™ gradient should have normal opalescent appearance.

2. To avoid contamination:

- Enter all bottles using sterile technique within a suitable sterile environment.
- Do not use the same sterile pipette or needle to reenter a bottle of medium.
- When entering a bottle through the stopper via a needle, use a sterile needle. Wipe the stopper with alcohol and air dry. Use a new sterile needle for each gradient.

MATERIALS REQUIRED BUT NOT PROVIDED

- Sterile disposable polystyrene 15-mL conical centrifuge tube.
- Sterile 5-mL disposable pipettes (graduated/transfer) or 3-cc syringes with 1.5"/21g needles.
- Centrifuge/fixer or horizontal. Must be able to operate for up to 30 minutes at 250g to 750g.
- 37°C incubator or water bath.
- Counting chamber.
- Microscope with x10 and x20 objectives.

CONTINUOUS ONE-STEP PURECEPTION™ GRADIENT

- Bring all reagents to 37°C before use. The PureCep™ 80% (REF #LPHTF-B) solution and PureCep™ Sperm Washing Medium (REF #SWHTF-A) should be stored at 2°C to 8°C until the expiration date shown on the box and container labels.
- Add 1.0 mL of PureCep™ 80% to a 15-mL conical centrifuge tube.
- Gently layer 1.5 to 2.0 mL of fresh liquefied semen on top of the PureCep™ 80% using a transfer pipette or syringe. Frozen-thawed samples should be thoroughly warmed to 37°C to maximize sperm

motility and diluted with at least x10 volumes of PureCep™ Sperm Washing Medium (REF #SWHTF-A) to dilute the cryoprotectant used in the freezing process. This diluted sperm mixture is then centrifuged at 300g for 5 minutes and the pellet resuspended in 1.0 to 2.0 mL of PureCep™ Sperm Washing Medium (REF #SWHTF-A), which is then layered on top of the PureCep™ 80%. There should be no mixing of the sample and the PureCep™ 80%. If the semen volume is more than 2.0 mL, use more than one tube of PureCep™ 80%.

- Centrifuge at 300g for 20 minutes. If the sample is viscous or has a low count, centrifugation for an extra 10 to 20 minutes may help to retrieve more sperm in individual cases.
- Using a pipette or syringe, carefully remove the PureCep™ 80% and seminal fluid without disturbing the sperm pellet, leaving a small amount of PureCep™ 80% over the sperm pellet. Aspirate from the top downward by always keeping the pipette tip just below the fluid surface. If no sperm pellet is clearly visible, remove all but 0.4 mL of the PureCep™ 80% layer. This will allow for the collection of sperm suspended in the PureCep™ 80%.
- Using a clean syringe or pipette, remove the sperm suspension from the bottom of the original tube and transfer it to a clean centrifuge tube; combine the contents of several tubes from the same specimen into this one clean tube. Add 3 to 5 mL of PureCep™ Sperm Washing Medium (REF #SWHTF-A) and resuspend the pellet by gently flicking the tube with your fingers.
- Centrifuge at 300g for 5 minutes to wash away residual PureCep™ 80% solution.
- Carefully remove the supernatant and resuspend the sperm pellet in a suitable volume of appropriate

medium (eg, use 0.4 mL of PureCep™ Sperm Washing Medium [REF #SWHTF-A] for IUI; use bicarbonate-buffered medium such as Quinn's Advantage™ Fertilization [HTF] Medium [REF #1020/1021] for IVF and dilute the sperm to an appropriate volume).

Each laboratory should make its own determination of which medium and protocol to use for each particular procedure.

Information on specific aspects of IVF, embryo culture, and cryopreservation is available in our Product Catalog (REF #80572).

TROUBLESHOOTING

1. Occasionally, samples that do not liquefy properly and remain too viscous to pass through the gradient will be encountered. Increasing the centrifugal force up to but no more than 750g will aid in separating the sperm in these cases. Pelleting motile sperm from highly viscous semen does not usually present a problem using the PureCep™ system.
2. The most important semen parameter contributing to a pellet of highly motile sperm is sperm progression. The higher the number of progressively motile sperm in the initial sample, the greater the number of sperm you will have in the final pellet. If sperm percent motility or progression is below WHO guidelines for normal semen parameters, you may be able to compensate for shortcomings in the initial semen analysis by using 1.0 mL of PureCep™ instead of 2.0 mL. You may also want to leave the last 0.5 to 0.7 mL of the postcentrifugation solution above the pellet and wash the sperm caught in transit through this portion. If this is done, be sure to

double the volume of PureCep™ Sperm Washing Medium (REF #SWHTF-A) in order to dilute out this remaining material.

QUALITY CONTROL

All PureCep™ solutions are tested using one-cell mouse embryo culture and endotoxin assays to ensure quality and safety. However, it is recommended that in-house quality control be performed with each lot.

PRECAUTIONS AND WARNINGS

PureCep™ comes packaged with tamper-proof seals and caps. If the seal is broken or the cap loose, do not use product.

PureCep™ 80% has a naturally opalescent appearance. Do not use product if it shows any evidence of particulate matter or contamination. This may be evident by extreme cloudiness or discoloration.

PureCep™ should remain tightly capped when placed in a CO₂ incubator to avoid pH changes.

When using this product you must use aseptic techniques to avoid contamination.

This product contains albumin, a derivative of human blood. All donors used in its manufacture were individually tested and found to be nonreactive for hepatitis B surface antigen (HBsAg) and antibodies to hepatitis C virus (HCV) and human immunodeficiency virus (HIV) by approved testing methods. Donors of the source material have been screened for Creutzfeldt-Jakob disease (CJD). Based on effective donor screening and product manufacturing processes, it carries an extremely remote risk for transmission of viral diseases. A theoretical risk for transmission of CJD is also considered extremely remote. No cases of transmission of viral diseases or CJD have ever been identified for albumin.

Caution: Federal law restricts this device to sale by or on the order of a physician (or properly licensed practitioner).

RELATED PRODUCTS

SAGE Assisted Reproduction Products™ has a full line of products for the Reproductive Medicine Specialist. Please call or write for specific information or to receive a copy of our current catalog. For technical questions, or to reach our Customer Service Department, call the SAGE Support Line at the number below.

PureCep™ is a trademark of CooperSurgical, Inc.

Quinn's Advantage® is a registered trademark of CooperSurgical, Inc.

Patrick Quinn, PhD, HCLD, SAGE IVF, Inc.

**Call the SAGE SUPPORT LINE at:
1-800-243-2974 or 1-203-601-5200**



SAGE In-Vitro Fertilization, Inc. A CooperSurgical Company
95 Corporate Drive, Trumbull, CT 06611
www.coopersurgical.com