SYNBIOS® MEDIA
Safety. Performance. Innovation

SYNBIOS®ASSISTED OOCYTE ACTIVATION KITS

For the activation of oocytes in ICSI fertilization failure

COMPOSITION
Media are **SYNBIOS®**- culture medium based and contains HEPES, with or without human serum albumin (HSA), Calcium ionophore Calcimycin and Calcium chloride. Antibiotics gentamycin is present.

MATERIAL INCLUDED WITH THE KIT ARE INDICATED BELOW:
One kit will provide sufficient medium for approximately 6-7 procedures.

OA10000 OOCYTE ACTIVATION kit contains vials of the following media:

OA10001 Calcium Chloride solution	1.5mL
OA10002 Calcimycin Calcium Ionophore	1.5mL

MATERIAL NOT INCLUDED WITH THE KIT
- Dishes, embryo culture
- Dishes, ICSI
- Dishes oocyte collection
- Embryo handling pipets
- Laminar flow hood
- Zoom Stereo Microscope,
- Lab timer
- Hand-held micro heat sealer
- Embryo culture and gamete handling media

SYNBIOS®ASSISTED OOCYTE ACTIVATION kit is compatible for use with **SYNBIOS** vitrification cooling and warming kits, **SYNBIOS** Hypoosmotic, Culture, Gamete and Flushing media to select viable sperm, culture, wash oocytes and embryos and for the cryopreservation of embryos.

PRODUCT SPECIFICATIONS
•Chemical composition
•pH: 7.20 – 7.40
•Osmolality (mOsm/kg):Pre-incubation med: 270-290
•Sterility: Sterile (SAL 10-3)
•Endotoxins: < 0.25 EU/ml
•Use of Ph Eur or USP grade products if applicable
• Certificate of analysis and MSDS are available upon request

STORAGE INSTRUCTIONS
Product must be stored in original package between 2-8°C. It must not be aliquoted into smaller containers for storage. Once removed from container, discard excess medium. Do not freeze. Aseptic technique is adhered to at all times. Do not use after expiry date.

DISCLAIMER:
Every effort has been taken to ensure quality of the product. We have no control over the products during transport. Shippers are aware of the cold-chain protocol. Nevertheless product could be damaged for any reason during transport (e.g. inspection by Border Security). End users must ensure products received are in good condition by internal QC/QA prior to use.

INSTRUCTIONS FOR USE

Read instructions carefully, prepare the necessary dishes and keep handy all required utility items before commencing.

Note: There are a number of variations in the proptocol for oocyte activation. You may follow your own protocol. The following is one version of the protocol.

1.Oocytes were stripped of its cumulus and corona cells using hyaluronidase or Disperase (SYNBIOS® Media) as per standard methods at 4 hours after oocyte retrieval.
2.Incubate denuded oocytes at 37°C in a 6% CO_2 air atmosphere in equilibrated SYNBIOS® embryo culture medium.
3.Metaphase II oocytes were selected for ICSI at 5-6 hours after retrieval.
4.About 1ul of washed spermatozoa specimen is resuspended into the PVP (SYNBIOS® Media) droplet of the previously prepared ICSI dish. The ICSI dish was prepared as per standard methods with the following additional item, namely a 5ul droplet of $CaCl_2$ solution (provided in the SYNBIOS® kit for Artificial Oocyte Activation)
5.Selected sperm is inflicted with a tail injury using the injection needle as per standard methods.
6. If no motile sperm are available, use the SYNBIOS® Hypoosmotic medium or the SYNBIOS Sperm Activation medium using protocol of the manufacturer to select live or viable sperms.
7. The selected sperm is drawn into an injection pipette.
8. Keeping the sperm head at the very tip of the pipette move the injection needle to the $CaCL_2$ droplet made from the $CaCL_2$ provided in the SYNBIOS® Oocyte Activation kit
9. Very carefully suck in the $CaCL_2$ solution into injection pipet ensuring that the length of Calcium chloride drawn corresponds and does not exceed that of the diameter of the oocyte.
10. Then move and position the pipet towards the oocyte for the ICSI procedure.
11. Inject the oocytes with $CaCl_2$ solution and the spermatozoa into the oocyte. Transfer the injected oocyte into the previously equilibrated SYNBIOS® embryo culture medium and then return dish to the CO_2 incubator for 30 minutes
12. At 30 minutes post incubation the injected oocytes are exposed for 15 minutes to the SYNBIOS®Ca_2+ ionophore medium droplet held in the same dish followed by thorough but gentle intensive washing in 3 droplets of equilibrated SYNBIOS® embryo culture medium held in the same dish and then culture in a fresh droplet of SYNBIOS® embryo culture medium and returned to the CO_2 incubator.
13. The oocytes are then cultured.
14. Some workers re-expose the oocytes to the Ca_2+ ionophore medium after 30 minutes of culture for another 15 minutes exposure to the oocyte activation medium. [This step may be performed at your discretion if it is part of your protocol otherwise one exposure to the oocyte activation medium is sufficient. If oocytes are re-exposed for a second time to oocyte activation medium it must be ensured oocytes are washed thoroughly before overnight culture].
15.Next morning oocytes are examined for fertilization.
16. Oocytes with normal 2 pronuclei are separated for further culture until embryo transfer as per standard procedure.
16.Normal embryos obtained following oocyte activation are examined and selected for embryo transfer and the remaining quality embryos are cryopreserved for the patient's later use.
17. End of instructions

WARNINGS AND PRECAUTIONS
This product must be used only by laboratory personnel competent in laboratory human Assisted Reproduction Technology (ART). All human and organic material is potentially infectious; including this product, if it contains HSA. All specimens must be handled as capable of transmitting harmful viral or prion diseases or hitherto unknown pathogenic agents. Wear protective garments. Strict aseptic techniques must be employed to avoid contamination.
•The product must not be used if any of the media supplied is cloudy and also do not use the product if the seal has been removed or is defective.

IMPORTANT: It is not possible to sterilize HSA with 100% certainty (Truyen et al., 1995) thus HSA must be treated as potentially infectious. ART Lab personnel are urged to wear personnel protective apparel including goggles for their safety. Lab Personnel must adhere to Good Laboratory Practices (GLP) for optimizing outcome and to avoid mishaps.

CAUTION: All media exposed to the elements or above 8ºC for >8 hours may be unfit for use for human ART treatment due to possible formation of toxic free radicals and products of putrefaction.

IMPORTANT: The biosafety of the calcium ionophore remains to be fully elucidated. This technique is still experimental. Workers are cautioned of possible adverse outcome as a direct consequence of this technique although no adverse outcome is known at this moment. Decision to use this technique/product have to considered carefully.

Note: This product is classified as a medical device. US Federal Law restricts its sale by or on order of a physician (Rx only). For intended use only.

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 May 2020

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20 Years of Research

Ensures Optimal Performance

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Safety. Performance. Innovation

GMP-Manufactured

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Ref: OA10000 Oocte Activation Kit with HSA
Ref: OA10000.SYN Oocyte Activation Kit without HSA

SYNBIOS® ASSISTED OOCYTE ACTIVATION KIT is sterilized by sterile filtration. Comes with and without HSA.