IVF consumables and media
IVF consumables and media
About Planer

Planer Ltd is now part of the Hamilton Thorne Group, a leading provider of precision instruments, consumables, software and services to the Assisted Reproductive Technologies (ART) and developmental biology research markets.

In 1973, Planer started out by developing and producing pioneering one-off machines for cryogenic researchers. Now, 45 years on, we are proud to have become the gold-standard supplier of equipment for cell preservation. Over this time, we have also helped scientists around the world achieve many notable breakthroughs, including the first baby born from a frozen embryo in 1984 and the first successful frozen ovary transplant 2014. In August 2019, we were acquired by the Hamilton Thorne Group, a move intended to significantly accelerate the growth of Planer operations around the world.

Our customers include laboratories, hospitals, pharmaceutical companies and the assisted reproduction fields, which we supply with the hardware, software and systems for the safe preservation, storage and monitoring of biological specimens such as embryos, blood products, tissue and biologicals.

In our factories near London’s Heathrow airport, we design and build our controlled rate freezers, incubators, sensors and systems. Around 90% of these products are exported with the help of our 80 or so sales and service distributors around the world. Planer equipment can now be found in most countries and our watchwords are robust design, compliant operation, Just-in-Time manufacture and long-standing relationships with our customers and distributors.

IVF consumables and media

Now, as UK distributor for the Gynemed media and consumables and Kitazato needles and catheters, our offering to the UK IVF market has been considerably strengthened. These carefully selected ranges, which meet our demanding quality standards, aim to help ensure that IVF practitioners achieve the maximum level of success.

Take a look at this catalogue to see the full range of IVF consumables and media products we have available and if you have any questions, please get in touch with the Planer sales team by emailing enquiries@planer.com.

Our product ranges

Cryopreservation

Cryopreservation using controlled rate freezers is used in a variety of applications including IVF; ART and research as well as stem cell, blood and large scale vaccine storage. Our range of freezers comes in a variety of sizes and caters for a broad range of different needs, whether a single straw or 8,000 vials need to be slow frozen.

Incubation

Our space saving, precise benchtop incubators are increasingly used in human and veterinary assisted reproduction applications. They offer the best possible in-vitro environment with rapid gas and temperature recovery times – and all with a battery backup.

Monitoring

To keep laboratories safe, both for samples and for operators, parameters such as temperature, humidity, carbon dioxide, liquid nitrogen level, oxygen, door status etc. need to be monitored around the clock. Our DATAssure™ wireless monitoring system meets the most stringent standards to help our customers to comply with HACCP, BRC, FDA and MRHA legislative requirements.

Cryo Storage

Ultra low cryo storage offers security for biological samples at -190 °C with long holding times and can be used in areas such as assisted reproduction, immunology, gene therapy, tissue banking, stem cells, cord blood, algae, fungi and viruses. The range we offer extends from small dewars to large capacity electronically controlled vessels.

UK Distributor for Hamilton Thorne and Gynemed products

Now, as part of the Hamilton Thorne Group, we are also the UK distributor for Hamilton Thorne clinical lasers and the Gynemed range of media and consumables for the IVF market.

Other products distributed in the UK by Planer

Planer now distributes the range of Kitazato IVF needles and catheters, including their OPU needles, IUI needles and ET catheters.

Visit our website to find out more

www.planer.com
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Gynemed
IVF consumables
and media
Applications for the Gynemed MediaLine

**Applications for Oocytes**
- GM501 Flush
  Aspiration medium

- GM501 Wash or GM501 Wash with Phenolred and Gentamicin
  Washing medium for oocytes

- GM501 Hyaluronidase
  Denudation of oocyte

- GM508 CultActive
  Activation of oocytes (ionophore)

**Applications for Sperm**
- GM501 SpermAir or SpermActive
  Sperm processing

- GM501 SpermStore
  Freezing medium (sperm and testicular tissue)

- GM501 SpermMobil
  Sperm activation (Theophylline)

**Cryopreservation**
- GM501 EmbryoStore
  Slow freezing kit

- GM501 VitriStore
  Vitrification kit

- GM501 Cult
  Culture medium with Gentamicin or Gentamicin and Phenolred optional

- GM501 Mineral Oil
  Cover of culture medium

**Catalysis**
- GM501 PVP
  Immobilisation of sperm cells

- GM501 Collagenase
  Digestion of testicular tissue

- GM501 Mineral Oil
  Cover of culture medium

- GM501 Gi142/V1
  A Hamilton Thorne Company
Gynemed
Oocyte Handling
GM501 Flush

Cell culture medium for human oocyte pick-up, GM501 Flush is a ready-to-use medium for flushing the ovarian follicles during the aspiration and/or oocyte pick-up intended for extra corporeal fertilisation procedures.

Composition
- NaCl, KCl, KH$_2$PO$_4$, MgSO$_4$, CaCl$_2$
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Heparin

Instructions for use
GM501 Flush needs to be warmed at 37 °C over night before use (no CO$_2$, with closed lid). GM501 Flush is HEPES buffered. Incubation in a CO$_2$-incubator will lower the pH.

Tested specifications
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

Order codes
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<tr>
<th>Size</th>
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<tr>
<td>1 x 50 ml</td>
<td>2 - 8 °C</td>
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</tr>
<tr>
<td>1 x 500 ml</td>
<td>2 - 8 °C</td>
<td>6 months</td>
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</table>

On request also available in the sizes 20 ml, 100 ml and 250 ml

" from time of manufacture

Cell culture medium for human oocyte pick-up, GM501 Flush is a ready-to-use medium for flushing the ovarian follicles during the aspiration and/or oocyte pick-up intended for extra corporeal fertilisation procedures.

- Ready-to-use
- HEPES buffered (21 mM)
- CO2-incubation is not required
- Contains Heparin (2.5 IU/ml)
- CE marked class III (0344)
GM501 Wash

GM501 Wash is a ready-to-use medium designed for washing procedures of human oocytes and embryos and any short-term handling procedures outside the incubator like washing after Hyaluronidase treatment (denudation), ICSI, polar body or blastomere biopsy.

Instructions for use
GM501 Wash must be equilibrated over night in a humidified CO₂-incubator (at 5 - 7 % CO₂, 37 °C).

Composition
• NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
• Bicarbonate, HEPES, EDTA
• Glucose, Lactate, Pyruvate
• Non-essential and essential Amino Acids, Alanyl-Glutamine
• Human Serum Albumin

Tested specifications
• pH
• Osmolality
• Sterility
• Endotoxins
• MEA

Order codes
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<td>2 - 8 °C</td>
<td>6 months</td>
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* from time of manufacture

Oocyte Handling
GM501 Wash

GM501 Wash is a ready-to-use medium designed for washing procedures of human oocytes and embryos and any short-term handling procedures outside the incubator like washing after Hyaluronidase treatment (denudation), ICSI, polar body or blastomere biopsy.

• Ready-to-use
• HEPES (15 mM) and bicarbonate buffered
• After CO₂ incubation the medium is stable at room atmosphere for short-term handling procedures
• Contains Human Serum Albumin (5.00 g/litre)
• CE marked class III (0344)
GM501 Wash with Phenolred and Gentamicin is a ready-to-use medium designed for washing procedures of human oocytes and embryos and any short-term handling procedures outside the incubator like washing after Hyaluronidase treatment (denudation), ICSI, polar body or blastomere biopsy and other.

**Composition**
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin, Phenolred

**Instructions for use**
GM501 Wash with Phenolred and Gentamicin must be equilibrated overnight in a humidified CO₂-incubator (at 5 - 7 % CO₂, 37 °C).

**Tested specifications**
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

**Order codes**
- FGY4GM501W+PR-20: 1 x 20 ml
- FGY4GM501W+PR-50: 1 x 50 ml

**Technical Specification**
- 

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**GM501 Wash with Phenolred and Gentamicin**

GM501 Wash with Phenolred and Gentamicin is a ready-to-use medium designed for washing procedures of human oocytes and embryos and any short-term handling procedures outside the incubator like washing after Hyaluronidase treatment (denudation), ICSI, polar body or blastomere biopsy and other.

- **Ready-to-use**
- **HEPES (15 mM) and bicarbonate buffered**
- **After CO₂ incubation the medium is stable at room atmosphere for short-term handling procedures**
- **Contains Human Serum Albumin (5.00 g/litre)**
- **Contains Gentamicin (10 mg/litre)**
- **Contains Phenolred**
- **CE marked class III (0344)**
GM501 Hyaluronidase is a ready-to-use solution designed to facilitate the removal of cumulus cells. Hyaluronidase digests the extracellular matrix in the cumulus-oocyte complex consisting of hyaluronic acid.

- Ready-to-use
- HEPES buffered
- CO₂-incubation is not required
- Contains Human Serum Albumin (4.00 g/litre)
- Contains pharmaceutical grade hyaluronidase (80 IU/ml)
- CE marked Class III (0344)

**Composition**

- NaCl, KCl, NaH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES
- Glucose, Lactate, Pyruvate
- Human Serum Albumin
- Pharmaceutical grade hyaluronidase from bovine origin

**Tested specifications**

- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

**Instructions for use**

GM501 Hyaluronidase contains HEPES; no CO₂-incubation is required, just warm it up to 37 °C.

**Recommended application**

For further information, see page 76.

**Can be used in combination with**

- GM501 Cult media
- GM501 Wash
- GM501 Mineral Oil

**Order codes**

<table>
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* from time of manufacture
GM501 Cult with Gentamicin

GM501 Cult with Gentamicin is a ready-to-use bicarbonate buffered culture medium, designed for fertilisation and for human embryo culture from day 1 to blastocyst stage. It can also be used for embryo transfer.

Composition
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin

Tested specifications
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

Can be used in combination with
- GM501 Mineral Oil

Instructions for use
GM501 Cult with Gentamicin must be equilibrated over night in a humidified CO₂-incubator (at 5 - 7 % CO₂, 37 °C).

References

Order codes
- FGY4GM501H+G-20 1 x 20 ml 2 - 8 °C 6 months
- FGY4GM501H+G-50 1 x 50 ml 2 - 8 °C 6 months

* from time of manufacture
GM501 Cult with Gentamicin and Phenolred

GM501 Cult with Gentamicin and Phenolred is a ready-to-use bicarbonate buffered culture medium, designed for fertilisation and for human embryo culture from day 1 to blastocyst stage. It can also be used for embryo transfer.

- Ready-to-use
- Bicarbonate buffered
- Single step medium - from fertilisation to blastocyst stage
- Can be used with or without medium change at day 3
- Suitable for microdrop (single and group) culture under oil and open culture systems
- Contains Gentamicin (10.00 mg/litre)
- Contains Human Serum Albumin (10.00 g/litre)
- Contains Phenolred
- CE marked class III (0344)

Composition
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin, Phenolred

Instructions for use
GM501 Cult with Gentamicin and Phenolred must be equilibrated over night in a humidified CO₂ - incubator (at 5 - 7 % CO₂, 37 °C).

References

Can be used in combination with
- GM501 Mineral Oil

Order codes
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<tr>
<td>1 x 50 ml</td>
<td>2 - 8 °C</td>
<td>6 months</td>
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* from time of manufacture
GM501 Mineral Oil

GM501 Mineral Oil is a ready-to-use oil for covering the medium during IVF/ICSI treatment. GM501 Mineral Oil protects the medium from evaporation and thereby stabilises pH and temperature.

**Composition**
- Paraffin oil
- Density 0.83 - 0.86
- Viscosity < 30 cP at 30 °C
- Pre-washed twice with ultra-pure water

**Instructions for use**

GM501 Mineral Oil is a pre-washed ready-to-use oil, so no further preparations are necessary. After pre-incubation (5 hours/37 °C) overlay the culture medium with GM501 Mineral Oil until it is completely covered.

**Recommended application**

For further information, see page 77.

**References**


**Composition**

- Paraffin oil
- Density 0.83 - 0.86
- Viscosity < 30 cP at 30 °C
- Pre-washed twice with ultra-pure water

**Tested specifications**

- Density
- Viscosity
- Sterility
- Endotoxins
- MEA
- Peroxide level

**Can be used in combination with**

- GM501 Cult media
- GM501 PVP
- GM501 Hyaluronidase

**Order codes**

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<td>1 x 500 ml</td>
<td>15 - 25 °C</td>
<td>18 months</td>
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* from time of manufacture
Gynemed

Sperm Processing
GM501 SpermAir is a ready-to-use medium designed for all human sperm preparation, sperm washing, swim up techniques and density gradient centrifugation as well as for testicular tissue.

**Composition**
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin, Phenolred

**Tested specifications**
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

**Instructions for use**
Do not equilibrate GM501 SpermAir in a CO₂-incubator, just warm it up to 37 °C. GM501 SpermAir is HEPES buffered. Incubation in a CO₂-incubator will lower the pH.

**Recommended application**
For further information, see pages 78 & 79.

**Order codes**

<table>
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<td>1 x 50 ml</td>
<td>2 - 8 °C</td>
<td>6 months</td>
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* from time of manufacture

GM501 SpermAir is a ready-to-use medium designed for all human sperm preparation, sperm washing, swim up techniques and density gradient centrifugation as well as for testicular tissue.

**Ready-to-use**
**HEPES buffered (21 mM)**
**CO₂-incubation is not required**
**For all human sperm handling and preparation procedures**
**Suitable for washing, swim-up and density gradient centrifugation**
**For handling of testicular tissue**
**Contains Gentamicin (10.00 mg/litre)**
**Contains Phenolred**
**Contains Human Serum Albumin (5.00 g/litre)**
**CE marked class III (0344)**
GM501 SpermActive is a ready-to-use medium designed for all human sperm preparation, sperm washing, swim up techniques and density gradient centrifugation as well as for testicular tissue.

**Composition**
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin, Phenolred

**Instructions for use**
GM501 SpermActive must be equilibrated over night in a humidified CO₂-incubator (at 5 - 7 % CO₂, 37 °C).

**Recommended application**
For further information, see pages 78 & 79.

**Tested specifications**
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

**Order codes**
- **FGY4GM501SA-20**: 1 x 20 ml, 2 - 8 °C, 6 months
- **FGY4GM501SA-50**: 1 x 50 ml, 2 - 8 °C, 6 months

* from time of manufacture

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Sperm Processing

GM501 SpermActive

GM501 SpermActive is a ready-to-use medium designed for all human sperm preparation, sperm washing, swim up techniques and density gradient centrifugation as well as for testicular tissue.

- **Ready-to-use**
- **HEPES** (15 mM) and bicarbonate buffered
- **After CO₂ incubation** the medium is stable at room atmosphere for short-term handling procedures.
- **For all human sperm handling and preparation procedures**
- **Suitable for** washing, swim-up and density gradient centrifugation
- **For handling of testicular tissue**
- **Contains** Gentamicin (10.00 mg/litre)
- **Contains** Phenolred
- **Contains** Human Serum Albumin (5.00 g/litre)
- **CE** marked class III (0344)
GM501 Gradient

GM501 Gradient is an isotonic solution for semen preparation with a density of approximately 1.12 g/ml.

Tested specifications
- pH
- Osmolality
- Density
- Viscosity
- Sterility
- Endotoxins
- Sperm Survival Test

Can be used in combination with
- GM501 SpermAir
- GM501 SpermActive

Order codes

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<td>1 x 250 ml</td>
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* from time of manufacture

Instructions for use
Mix the density gradient bottles by 5 bottle inversions before use. We would advise you to produce a 2-phase system from the 100 % gradient (45 % and 90 %). You may prefer a different mixing ratio (e.g. 40 % and 80 %) or a multi layer gradient (45 %–70 %–90 %). Mix 9 parts of GM501 Gradient 100 % with 1 parts of washing medium to produce the 90 % gradient. Mix 4.5 parts of GM501 Gradient 100 % with 5.5 parts of washing medium to produce the 45 % gradient.

Note: Gradients should be prepared and repacked under sterile conditions (e.g. LAF-bench, ISO Class 5). For optimal results prepare the gradient media a maximum of 24 hours prior to use. Mix well after diluting the GM501 Gradient 100 %.

Recommended application
For further information, see page 80.
GM501 PVP

GM501 PVP is a ready-to-use media to reduce the motility of sperm making it easier to catch them with an ICSI pipette. It is possible to dilute the solution with HEPES buffered sperm processing media.

Composition
- NaCl, KCl, NaH$_2$PO$_4$, MgSO$_4$, CaCl$_2$
- Bicarbonate, HEPES
- Glucose, Lactate, Pyruvate
- Human Serum Albumin, Polyvinylpyrrolidone

Instructions for use
Warm the PVP solution to 37 °C

Standard procedure: Place a small drop of PVP solution (5 µl - 10 µl) in a dish and cover with GM501 Mineral Oil. Add a small volume (1 µl - 2 µl) of washed sperm cells into the centre of the PVP droplet. Wait for a few minutes to allow the sperm cells to migrate to the periphery of the droplet. Select and recover the spermatozoa for injection. Warm the PVP solution and HEPES buffered sperm processing medium to 37 °C.

Alternative procedure (with extra washing step): Place a small drop of PVP solution (5 µl - 10 µl) and 1 or more small drops HEPES buffered sperm processing medium in a dish and cover with GM501 Mineral Oil. Add a small volume (1 µl - 2 µl) of washed sperm cells into the centre of the PVP droplet. Wait for a few minutes to allow the sperm cells to migrate to the periphery of the droplet. Select the spermatozoa for injection and nick (break) the tail of the spermatozoon with the tip of the pipette. Transfer the spermatozoon into one of the HEPES buffered sperm processing medium droplets and wash by transferring the sperm cell in and out of the sperm processing medium several times. Aspirate the sperm cell into the pipette and use for ICSI procedure.

Can be used in combination with
- GM501 Mineral Oil
- GM501 SpermAir
- GM501 SpermActive

Order codes

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<td>9 months</td>
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* from time of manufacture
Bromelain in Dulbecco’s PBS

Bromelain in Dulbecco’s PBS is designed for liquefaction of viscous semen samples prior to semen analysis and preparation for further IVF treatment.

- Ready-to-use
- For liquefaction of viscous semen samples
- Contains 10 IU/ml Bromelain
- Formulated according to WHO laboratory manual for the Examination and processing of human semen - Fifth edition
- CE marked class IIb (0482)

Composition

- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂, Na₂HPO₄
- Glucose
- Bromelain

Tested specifications

- pH
- Osmolality
- Sterility
- Endotoxins
- MEA
- Sperm Survival Test

Instructions for use

Warm the Bromelain in Dulbecco’s PBS to 37 °C.

Recommended application

For further information, see page 86.

References


Order code | Size | Storage | Shelf life* |
---|---|---|---|
FGY4GM501BROM10 | 1 x 10 ml | 2 - 8 °C | 6 months |

* from time of manufacture
GM501 Collagenase

GM501 Collagenase is a reagent for the digestion of human testicular tissue obtained by testicular biopsy (TESE) for in-vitro examination procedures. Using Collagenase, it is possible to degrade testicular tissue in single cells to facilitate the isolation of sperm cells.

**Composition**
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Human Serum Albumin, Gentamicin, Phenolred
- Collagenase (obtained from culture filtrates of Clostridium histolyticum)

**Can be used in combination with**
- GM501 SpermAir
- GM501 SpermActive

**Instructions for use**
Do not equilibrate GM501 Collagenase in a CO₂-incubator, just warm it up to 37 °C. GM501 Collagenase is HEPES buffered. Incubation in a CO₂-incubator will lower the pH.

**Recommended application**
For further information, see page 87.

**References**

**Sperm Processing**

GM501 Collagenase is a reagent for the digestion of human testicular tissue obtained by testicular biopsy (TESE) for in-vitro examination procedures. Using Collagenase, it is possible to degrade testicular tissue in single cells to facilitate the isolation of sperm cells.

- Ready-to-use
- HEPES buffered
- Digestion of human testicular tissue obtained by biopsy
- Facilitates isolation of sperm cells from TESE digestion
- Contains 1000 CDU/ml (Collagen Digestive Units)
- Contains Human Serum Albumin (5.00 g/litre)
- CE marked class IIb (0482)

**Tested specifications**
- pH
- Osmolality
- Sterility
- Sperm Survival Test

Performing LAL-endotoxin- and MEA-tests is not possible with this medium as the activity of the Collagenase inactivates the enzymes (LAL) and damages the mouse embryos (MEA), which are used for these assays, respectively. The basic medium is LAL- and MEA-tested.

<table>
<thead>
<tr>
<th>Order code</th>
<th>Size</th>
<th>Storage</th>
<th>Shelf life*</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGY4COLL-AIR</td>
<td>1 x 3 ml</td>
<td>2 - 8 °C</td>
<td>9 months</td>
</tr>
</tbody>
</table>

* from time of manufacture
Insemination Kit

The Insemination Kit is a complete system for the simple and safe preparation and processing of human spermatozoa out of the ejaculate, for homologous and heterologous intrauterine inseminations (IUI). The Insemination Kit uses the self motility of the male germ cells to isolate motile spermatozoa in high concentrations. We recommend the use if the ejaculate is normozoospermic or slightly oligoand/ or asthenozoospermic.

One kit contains
- 1 x Vial with 2 ml of GM501 SpermAir medium (sterile)
- 2 x 2 ml syringes (sterile)
- 1 x short cannula (sterile)
- 2 x long cannulas (sterile)
- 1 x Insemination catheter standard or memo (sterile)
- 1 x Instructions for use
- 1 x Ampulla rack

Instructions for use

Recommended application
For further information, see page 81.

<table>
<thead>
<tr>
<th>Order code</th>
<th>Size</th>
<th>Storage</th>
<th>Shelf life*</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGY4SA-Kit-002</td>
<td>1 x Standard kit</td>
<td>2 - 8 °C</td>
<td>6 months</td>
</tr>
</tbody>
</table>

* from time of manufacture
Gynemed
Cryopreservation
GM501 EmbryoStore

GM501 EmbryoStore is a set of ready-to-use antibiotic free media for freezing and thawing of human embryos between 2PN and 4-cell stage.

Composition
- NaCl, KCl, KH₂PO₄, Na₂HPO₄
- Sucrose
- Propandiol
- Human Serum Albumin

Tested specifications
- pH
- Osmolality (EmbryoStore Thaw 3)
- Sterility
- Endotoxins
- MEA

Instructions for use
Ensure all media are mixed well and warmed up to room temperature before use.

Recommended application
For further information, see page 82.

Order codes
<table>
<thead>
<tr>
<th>Order codes</th>
<th>Size</th>
<th>Storage</th>
<th>Shelf life*</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGY4EMF01_P_F</td>
<td>1 x 10 ml Freeze</td>
<td>2 - 8 °C</td>
<td>18 months</td>
</tr>
<tr>
<td>FGY4EMF01_P_T1</td>
<td>1 x 10 ml Thaw 1</td>
<td>2 - 8 °C</td>
<td>18 months</td>
</tr>
<tr>
<td>FGY4EMF01_P_T2</td>
<td>1 x 10 ml Thaw 2</td>
<td>2 - 8 °C</td>
<td>18 months</td>
</tr>
<tr>
<td>FGY4EMF01_P_T3</td>
<td>1 x 10 ml Thaw 3</td>
<td>2 - 8 °C</td>
<td>18 months</td>
</tr>
</tbody>
</table>

* from time of manufacture
GM501 VitriStore Freeze - GM501 VitriStore Thaw

GM501 VitriStore Freeze/VitriStore Thaw are a set of ready-to-use antibiotic free media for vitrification and thawing of human embryos.

Composition

**GM501 VitriStore Freeze**
- NaCl, KCl, KH₂PO₄, Na₂HPO₄
- Sucrose
- DMSO, Ethylene Glycol, Ficoll
- Human Serum Albumin

**GM501 VitriStore Thaw**
- NaCl, KCl, KH₂PO₄, Na₂HPO₄
- Sucrose
- Human Serum Albumin

Tested specifications
- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

Recommended application
For further information, see pages 83 & 84.

Instructions for use
Ensure all media are well mixed before use. We strongly recommend that you read through all the steps of the vitrification/thawing procedure before starting the procedure.

Order codes

<table>
<thead>
<tr>
<th>Order codes</th>
<th>Size</th>
<th>Storage</th>
<th>Shelf life*</th>
</tr>
</thead>
</table>
| FGY4VF_KIT1  | 1 x VitriStore Freeze Kit
1 x 5 ml Pre-vitrification Medium
1 x 1 ml Freeze Medium 1
1 x 1 ml Freeze Medium 2 | 2 - 8 °C | 12 months |
| FGY4VT_KIT1  | 1 x VitriStore Thaw Kit
1 x 5 ml Thaw Medium 1
1 x 1 ml Thaw Medium 2
1 x 1 ml Thaw Medium 3
1 x 1 ml Thaw Medium 4 | 2 - 8 °C | 12 months |

* from time of manufacture

References
GM501 SpermStore

GM501 SpermStore is a antibiotic free medium for freezing human spermatozoa including epididymal or testicular sperm.

**Composition**
- NaCl, KCl, MgSO$_4$, NaH$_2$PO$_4$
- Bicarbonate, HEPES
- Glucose, Lactate, Sucrose
- Glycine
- Glycerol
- Human Serum Albumin

**Instructions for use**
Ensure all media are mixed well and warmed up to room temperature before use.

**Recommended application**
For further information, see page 85.

**Tested specifications**
- pH
- Sterility
- Endotoxins
- Sperm Survival Test

**Order code**

<table>
<thead>
<tr>
<th>Order code</th>
<th>Size</th>
<th>Storage</th>
<th>Shelf life*</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGY4 SCP-20</td>
<td>1 x 20 ml</td>
<td>2 - 8 °C</td>
<td>18 months</td>
</tr>
</tbody>
</table>

* from time of manufacture
Gynemed
Miscellaneous &
In Vitro Diagnostics
GM501 HSA

GM501 HSA is intended for use in assisted reproductive procedures which include gamete and embryo manipulation. These procedures include the use of GM501 HSA solution as a supplement for culture medium.

Composition

- Human Serum Albumin in saline buffer (100.00 mg/ml)

Tested specifications

- pH
- Osmolality
- Sterility
- Endotoxins
- MEA

Can be used in combination with

- GM501 Gradient

Instructions for use

To supplement the 50 ml culture medium (e.g. GM501 Basic) with GM501 HSA to a protein end concentration of 1%. Keep 45 ml culture medium in the bottle and add 5 ml of GM501 HSA. Mix well.

In Vitro Diagnostics

GM501 HSA

GM501 HSA is intended for use in assisted reproductive procedures which include gamete and embryo manipulation. These procedures include the use of GM501 HSA solution as a supplement for culture medium.

- For individual supplementation of media used in assisted reproduction
- Contains Human Serum Albumin (100.00 g/litre)

Order code | Size | Storage | Shelf life*  
--- | --- | --- | ---  
FGY4HSA0005 | 1 x 5 ml | 2 - 8 °C | 12 months

* from time of manufacture
GM501 SpermMobil is a HEPES buffered HSA free reagent containing low bicarbonate. It is used for in vitro examination of sperm cells of necrozoospermic ejaculates as well as of immotile sperms isolated from testicular tissue (TESE).

**Composition**
- NaCl, KCl, KH₂PO₄, MgSO₄, CaCl₂
- Bicarbonate, HEPES, EDTA
- Glucose, Lactate, Pyruvate
- Non-essential and essential Amino Acids, Alanyl-Glutamine
- Theophylline, Phenolred

**Instructions for use**
Do not equilibrate GM501 SpermMobil in a CO₂-incubator, just warm it up to 37 °C. GM501 Sperm-Mobil is HEPES buffered. Incubation in a CO₂-incubator will lower the pH.

**Recommended application**
For further information, see page 88.

**References**

**Tested specifications**
- pH
- Osmolality
- Sterility
- Endotoxins
- Sperm Survival Test

**Can be used in combination with**
- GM501 SpermAir
- GM501 SpermActive

**Order codes**
- FGY4GM501 SMOBIL5: 1 x 5 ml, 2 - 8 °C, shelf life 6 months
- FGY4GM501 SMOBIL5-S: 1 x 1 ml, 2 - 8 °C, shelf life 6 months

* from time of manufacture
GM508 CultActive

GM508 CultActive is a bicarbonate buffered HSA free reagent designed to investigate oocytes of patients with failed fertilisation after previous intracytoplasmatic sperm injection cycles.

GM508 CultActive is designed to investigate if fertilisation failure after previous ICSI cycles is due to a deficient oocyte activation.

Composition
• NaCl, KCl, KH$_2$PO$_4$, MgSO$_4$, CaCl$_2$
• Bicarbonate, EDTA
• Glucose, Lactate, Pyruvate
• Non-essential and essential Amino Acids, Alanyl-Glutamine,
• Ca$^{2+}$-Ionophore A23187, DMSO

Instructions for use
GM508 CultActive must be equilibrated 4 hours in vial not firmly closed at 5 - 7 % CO$_2$ and 37 °C prior to use.

Recommended application
For further information, see page 89

References

Can be used in combination with
• GM501 Cult media

Order code | Size | Storage | Shelf life*
--- | --- | --- | ---
FGY4GM508CULT-active 1 | 1 x 1 ml | 2 - 8 °C | 6 months

* from time of manufacture
Gynemed Pipettes
Holding micropipettes

Specifications and quality control

- To meet international standards as well as the requirements of the FDA, the Holding micropipettes are sterilised by gamma radiation.
- The Holding micropipettes are prepared from borosilicate glass tubing.
- Outer diameter 1.00 mm, inner diameter 0.75 mm, total length 5.50 cm, polished opening, length arm 0.9 mm, bending angle 20°-40°.
- The micropipettes are available straight or with bending angle.
- A Mouse Embryo Test (MEA) is available for each batch upon request from our website with respective lot number.

Order codes

<table>
<thead>
<tr>
<th>Order codes</th>
<th>Outer diameter µm</th>
<th>Polished opening µm</th>
<th>Angle</th>
<th>Box (pieces)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Small outer diameter</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FGYHP-80-0°</td>
<td>80 µm</td>
<td>15 µm</td>
<td>Straight</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-80-20°</td>
<td>80 µm</td>
<td>15 µm</td>
<td>20 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-80-30°</td>
<td>80 µm</td>
<td>15 µm</td>
<td>30 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-80-35°</td>
<td>80 µm</td>
<td>15 µm</td>
<td>35 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-80-40°</td>
<td>80 µm</td>
<td>15 µm</td>
<td>40 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>Medium outer diameter</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FGYHP-100-0°</td>
<td>100 µm</td>
<td>20 µm</td>
<td>Straight</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-100-20°</td>
<td>100 µm</td>
<td>20 µm</td>
<td>20 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-100-30°</td>
<td>100 µm</td>
<td>20 µm</td>
<td>30 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-100-35°</td>
<td>100 µm</td>
<td>20 µm</td>
<td>35 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-100-40°</td>
<td>100 µm</td>
<td>20 µm</td>
<td>40 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>Large outer diameter</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FGYHP-120-0°</td>
<td>120 µm</td>
<td>25 µm</td>
<td>Straight</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-120-20°</td>
<td>120 µm</td>
<td>25 µm</td>
<td>20 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-120-30°</td>
<td>120 µm</td>
<td>25 µm</td>
<td>30 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-120-35°</td>
<td>120 µm</td>
<td>25 µm</td>
<td>35 degrees angle</td>
<td>20</td>
</tr>
<tr>
<td>FGYHP-120-40°</td>
<td>120 µm</td>
<td>25 µm</td>
<td>40 degrees angle</td>
<td>20</td>
</tr>
</tbody>
</table>

* from time of manufacture

Holding micropipettes are used for the fixation of oocytes, embryos or blastocysts and are therefore essential for all micromanipulation procedures in ART like ICSI, assisted hatching and polar body or blastomere biopsy.

- Sterile
- 3 years shelf life*
- Individually packed
- Customized production of pipettes available on request
- Mouse Embryo Tested
- CE marked (2265)
**Injection micropipettes**

ICSi (Intracytoplasmic Sperm Injection) micropipettes are used to aspirate and inject the sperm directly into the oocyte. Large ICSI-Spermatid micropipettes are used for aspiration and injecting immature sperm directly into the oocyte. ICSI-Spermatid micropipettes have an inner diameter of 7.00 - 8.00 µm and a tip outer diameter of 9.00 - 10 µm.

- **Sterile**
- **3 years shelf life**
- **Individually packed**
- **Customized production of pipettes available on request**
- **Mouse Embryo Tested**
- **CE marked (2265)**

**Specification and quality control**
- To meet international standards as well as the requirements of the FDA, the ICSI micropipettes are sterilised by gamma radiation.
- The ICSI micropipettes are prepared from borosilicate glass tubing.
- Outer diameter 1.00 mm, inner diameter 0.78 mm, total length 5.50 cm, bending angle 20°-35° with length of arm 0.90 mm, beveled 35°, with inner diameter of the tip 4.50 - 5.00 µm.
- The micropipettes are made with or without spike, straight or with bending angle and beveled 30°-40°.
- A Mouse Embryo Test (MEA) is available for each batch upon request from our website with respective lot number.

**Order codes**

<table>
<thead>
<tr>
<th>Order codes</th>
<th>Inner diameter µm</th>
<th>Angle</th>
<th>Spike(s)</th>
<th>Box (pieces)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGYIC-5-0°-be</td>
<td>5 µm</td>
<td>Straight</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-0°-s</td>
<td>5 µm</td>
<td>Straight</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-20°-be</td>
<td>5 µm</td>
<td>20 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-20°-s</td>
<td>5 µm</td>
<td>20 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-30°-be</td>
<td>5 µm</td>
<td>30 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-30°-s</td>
<td>5 µm</td>
<td>30 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-35°-be</td>
<td>5 µm</td>
<td>35 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-35°-s</td>
<td>5 µm</td>
<td>35 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-40°-be</td>
<td>5 µm</td>
<td>40 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-5-40°-s</td>
<td>5 µm</td>
<td>40 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-0°-be</td>
<td>7 µm</td>
<td>Straight</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-0°-s</td>
<td>7 µm</td>
<td>Straight</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-20°-be</td>
<td>7 µm</td>
<td>20 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-20°-s</td>
<td>7 µm</td>
<td>20 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-30°-be</td>
<td>7 µm</td>
<td>30 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-30°-s</td>
<td>7 µm</td>
<td>30 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-35°-be</td>
<td>7 µm</td>
<td>35 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-35°-s</td>
<td>7 µm</td>
<td>35 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-40°-be</td>
<td>7 µm</td>
<td>40 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-7-40°-s</td>
<td>7 µm</td>
<td>40 degrees angle</td>
<td>With spike</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-SL-4.6-30°-s</td>
<td>4.6 µm</td>
<td>30 degrees angle</td>
<td>Thin line</td>
<td>20</td>
</tr>
<tr>
<td>FGYIC-SL-4.6-35°-s</td>
<td>4.6 µm</td>
<td>35 degrees angle</td>
<td>Thin line</td>
<td>20</td>
</tr>
</tbody>
</table>

* from time of manufacture
Preimplantation Genetic Diagnosis - PGD.

Biopsy micropipettes are used to perform biopsies on the embryo (blastocyst) or the oocyte (polar body) for Preimplantation Genetic Diagnosis - PGD.

- Sterile
- 3 years shelf life
- Individually packed
- Customized production of pipettes available on request
- Mouse Embryo Tested
- CE marked (2265)

**Specification and quality control**

- To meet international standards as well as the requirements of the FDA, the Biopsy micropipettes are sterilised by gamma radiation.
- The Biopsy micropipettes are prepared from borosilicate glass tubing.
- Outer diameter 1.00 mm, inner diameter 0.78 mm, total length 5.50 cm, bending angle 20°- 45°, length of the arm 0.50 mm, with blunt opening (A) or beveled 40° and polished (B) and with inner diameter 10, 15, 20, 30 and 35 µm.
- Biopsy micropipettes may be ordered straight or with bending angle.
- A Mouse Embryo Test (MEA) is available for each batch upon request from our website with respective lot number.

**TECHNICAL SPECIFICATION**

<table>
<thead>
<tr>
<th>Order codes</th>
<th>Inner diameter µm</th>
<th>Angle</th>
<th>Opening</th>
<th>Box (pieces)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGYBP-10-0°-bl</td>
<td>10 µm</td>
<td>Straight</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-10-0°-be</td>
<td>10 µm</td>
<td>Straight</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-10-30°-bl</td>
<td>10 µm</td>
<td>30 degrees angle</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-10-30°-be</td>
<td>10 µm</td>
<td>30 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-10-35°-bl</td>
<td>10 µm</td>
<td>35 degrees angle</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-10-35°-be</td>
<td>10 µm</td>
<td>35 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-0°-bl</td>
<td>15 µm</td>
<td>Straight</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-0°-be</td>
<td>15 µm</td>
<td>Straight</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-30°-bl</td>
<td>15 µm</td>
<td>30 degrees angle</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-30°-be</td>
<td>15 µm</td>
<td>30 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-35°-bl</td>
<td>15 µm</td>
<td>35 degrees angle</td>
<td>Blunt</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-15-35°-be</td>
<td>15 µm</td>
<td>35 degrees angle</td>
<td>Bevelled</td>
<td>20</td>
</tr>
<tr>
<td>FGYBP-20-0°-bl</td>
<td>20 µm</td>
<td>Straight</td>
<td>Blunt</td>
<td>20</td>
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A Mouse Embryo Test (MEA) is available for each batch upon request from our website with respective lot number.
DENU-Tips

The DENU-Tips are used for the manipulation and transfer of oocytes and embryos during IVF/ICSI procedures.

- Made out of Polyamid
- Singly and sterile packed of 20
- Sterilised for 3 years*
- Different sizes available
- Mouse-Embryo-tested
- CE marked (Class IIa)

* from time of manufacture

Specification and quality control

- Considered to be Bisphenol A (BPA) free.
- The DENU-Tips are available in a variety of sizes with an inner diameter from 130 µm to 550 µm. For easy differentiation, each size is color coded.
- A Mouse Embryo Test (MEA) result is available for each lot number upon request from our website.

<table>
<thead>
<tr>
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Preparation for denudation of fresh oocytes

**RECOMMENDED APPLICATION**

GM501 Hyaluronidase

**Preparation of the 4-well dish**

- One 4-well dish (“Hy-dish”) needs to be prepared for 10 oocytes.
- First fill well 2 to 4 with 400 µl GM501 Wash medium and cover the filled wells with GM501 Mineral Oil. Equilibrate the dish overnight in a humidified CO₂ incubator.
- Warm the GM501 Hyaluronidase and fill the first well with 400 µl warmed GM501 Hyaluronidase.
- Preparation of the 4-well dish

**Denudation procedure using the microdrop dish**

1. Pipette up to 10 oocytes into the first drop (“1”) of the dish.
2. Transfer 5 oocytes to the “Hy” drop.
3. Pipette the oocytes immediately up and down (5 to 10 times) using for example a pipette with 100 µl tip (MEA-tested) adjusted to 50 µl. The cumulus cells will detach and the oocytes still surrounded by corona cells will be visible.
   **ATTENTION:** The oocytes should not be in the Hyaluronidase for more than 30 seconds!
4. Pick up the oocytes using the denudation pipette (inner diameter 125-155 µm) and transfer them to the next GM501 Wash containing drop (“2”). Aspirate and blow back the oocytes repeatedly to remove residual hyaluronidase.
5. Transfer the oocytes to the next GM501 Wash containing drop (“3”). Aspirate and blow back the oocytes repeatedly until nearly all corona cells are removed.
6. Transfer the oocytes to the next GM501 Wash containing drop (“4”). Leave the denuded oocytes in this drop.
7. Repeat steps 2 to 6 with the remaining oocytes.
8. When all oocytes are collected in GM501 Wash drop “4” and are free of cumulus and corona cells, wash them in the last two drops (“5” and “6”).
9. The denuded oocytes can then be transferred to a dish containing GM501 Cult for further incubation until ICSI is performed or directly to an ICSI dish.

Microdrop dish

Day -1:
Preparation of a microdrop dish with sufficient 80 µl GM501 Cult. Add up to 6 oocytes per well for fertilisation and the appropriate amount of prepared spermatozoa. Prepare an additional 4-well dish in the same fashion for the next day (“culture” dish).

Day 0:
IVF - Add up to 6 oocytes per well for fertilisation and the appropriate amount of prepared spermatozoa. Prepare an additional 4-well dish in the same fashion for the next day (“culture” dish).

Day 1:
ICSI - Directly after microinjection add up to 6 oocytes per well for culture.

**Simple “one-step” culture**

**Day -1:**
Preparation of dishes for IVF or ICSI using GM501 Cult Media

**Day 0:**
Fertilisation by IVF or ICSI and culture in GM501 Cult Media

**Day 1:**
Ongoing culture in GM501 Cult Media

**Day 2 or 3:**
Embryo transfer

**Extended “one-step” culture**

**Day -1:**
Preparation of dishes for IVF or ICSI using GM501 Cult Media

**Day 0:**
Fertilisation by IVF or ICSI and culture in GM501 Cult Media

**Day 1:**
Ongoing culture in GM501 Cult Media

**Day 2:**
Preparation of new dishes for day 3 if wanted

**Day 3:**
Ongoing culture in GM501 Cult Media

**Day 4:**
Ongoing culture in GM501 Cult Media

**Day 5:**
Embryo transfer

---

**Embryo Culture**

Preparation of dishes for IVF or ICSI using GM501 Cult Media

**Day 0:**
Fertilisation by IVF or ICSI and culture in GM501 Cult Media

**Day 1:**
Ongoing culture in new dishes

**Day 2:**
Preparation of new dishes for day 3 if wanted

**Day 3:**
Ongoing culture in GM501 Cult Media

**Day 4:**
Ongoing culture in GM501 Cult Media

**Day 5:**
Embryo transfer

**Day -1:**
Add the appropriate amount of prepared spermatozoa. Prepare an additional dish in the same fashion for the next day (“culture” dish).

**Day 0:**
If desired, assess pronuclear status.

**Day 1:**
ICSI - No further intervention necessary. If desired, assess pronuclear status.

**Day 2:**
ICSI - No further intervention necessary. If desired, assess pronuclear status.

**Day 3:**
ICSI - No further intervention necessary. If desired, assess pronuclear status.

**Day 4:**
ICSI - No further intervention necessary. If desired, assess pronuclear status.

**Day 5:**
ICSI - No further intervention necessary. If desired, assess pronuclear status.
**Swim-Up**

**Preparation**

We recommend one of the products from the GM501 MediaLine:
- GM501 SpermAir
- GM501 SpermActive

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**Washing 1**

To prepare the swim-up tubes, transfer 4.5 ml medium into a new conical centrifuge tube. Mix well with 1.0-3.0 ml liquefied semen. Centrifuge the tube at 300-400x g for 10 minutes.

**Swim-Up**

Aspirate the supernatant without dispersing the pellet and discard it. Carefully overlay the pellet with equilibrated GM501 SpermActive or warmed GM501 SpermAir medium. Put the tube in the CO₂-incubator (GM501 SpermActive) or heating cabinet (GM501 Sperm-Air) for 1 hour with top not firmly closed.

Aspirate the supernatant containing the motile sperm and pour into a new conical centrifuge tube.

---

**Washing 2**

Resuspend the pellet with 1 ml equilibrated GM501 SpermActive or warmed GM501 SpermAir medium. Centrifuge the tube at 300-400x g for 6 minutes. Aspirate the supernatant without dispersing the pellet and discard it. Repeat the step.

---

**Swim-Up**

To prepare the swim-up tubes, transfer 2 ml medium into a new conical tube. Underlay gently 1 ml of liquefied semen.

Place the tube in the CO₂-incubator (GM501 SpermActive) or heating cabinet (GM501 Sperm-Air) for 1 hour with top not firmly closed.

Aspirate the supernatant and discard it.

For ICSI or Insemination resuspend it in GM501 SpermActive or GM501 SpermAir.

---

**Washing**

Aspirate the upper media layer containing the motile sperms without dispersing the native ejaculate and fill it into a new conical centrifuge tube.

Centrifuge the tube at 300-400 g for 6 minutes.

Aspirate the supernatant without dispersing the pellet and discard it. Resuspend the pellet with 1 ml equilibrated GM501 SpermActive or warmed GM501 SpermAir medium. Centrifuge the tube at 300-400x g for 6 minutes. Repeat the step.

Aspirate the supernatant and discard it.

For ICSI or Insemination resuspend it in GM501 SpermActive or GM501 SpermAir.
Density centrifugation

**Preparation**

Before use warm all components of the system and the samples to 37 °C or to room temperature. Mix the density gradient bottles by a minimum of 5 bottles inversions before use.

Pipette 2.5 ml of the lower density gradient (e.g. 45%) into a sterile disposable centrifuge tube.

Using a 3 ml syringe with a 21 G needle, layer 2.5 ml of the higher density gradient (e.g. 90%) under the lower density gradient (e.g. 45%) free of air bubbles.

Take care that the two layers are distinctly separated. This is done by placing the tip of the needle at the bottom of the centrifuge tube and slowly dispensing the higher density gradient.

These two layers of density are stable for about two hours.

Gently place 2.5 ml of liquefied semen onto the upper layer using a transfer pipette or syringe. Centrifuge at 350-400x g for 15-18 minutes. In case, no pellet is visible after this step, centrifuge for another 3 minutes. Centrifugation force should not be increased over 500x g.

Aspirate the supernatant.

Using a syringe, resuspend the pellet with 2-3 ml of fresh washing medium. Centrifuge at 300x g for 8-10 minutes. In case you want to gain higher sperm concentrations it is advisable to centrifuge for the whole 10 minutes.

Aspirate the supernatant and repeat the last two steps.

Finally remove the remaining liquid to leave the pellet resuspended in the required amount for use with the subsequent procedure of assisted reproductive medicine (e.g. IVF, ICSI, IUI).

Insemination Kit

1. Warm up the GM501 SpermAir vial to 37 °C.
2. Remove metal cap from the stopper and desinfect the stopper’s surface with isopropylalcohol (70%).
3. Insert the enclosed short cannula through the stopper. (It serves as a pressure balance valve).
4. Aspirate liquefied, analysed ejaculate into an enclosed 2 ml syringe and attach a long cannula.
5. Hold the syringe with its tip upwards to collect air in the upper part of the syringe and press it out.
6. Insert the syringe’s cannula (tip downwards) through the vial’s stopper until the tip touches the bottom of the vial.
7. Now release the ejaculate slowly and carefully by depressing the syringe and let it suspend under the preparation medium without mixing the two liquids.
8. Remove the syringe with the cannula carefully while leading the tip along the inner wall of the vial. Discard the syringe and the cannula.
9. Now carefully place the vial’s neck into the rack’s fork and store the vial at 37 °C in an incubator (no CO₂) or in a warming cabinet for least 45 minute and not longer than 3 hours.
10. At the appropriate time carefully take the vial out and turn it upright. Attach a fresh long cannula on the tip of a fresh 2 ml syringe, aspirate 1ml of air and insert it again through the disinfected stopper.
11. Aspirate 0.5 to 1.0 ml of the upper media layer and remove the syringe with the cannula. The syringe now contains the SpermAir fraction with the isolated motile sperms. Until the insemination procedure place the syringe with the cannula with the attached protection cap of the cannula in an incubator (no CO₂) / warming cabinet at 37 °C.

12. To inseminate remove the cannula from the syringe and attach the enclosed IUI-catheter to the tip of the syringe.
13. The position assistance is adjusted corresponding to the anatomical proportions determined before.
14. The catheter is inserted until the assistance is positioned on the outer uterine orifice.
15. As soon as the requested position has been reached, the catheter will be turned, so that the marks on the grip are lying visible on top. In this way both of the lateral openings at the very end of the catheter are lined up towards the applicators orifice.
16. The suspension with the spermatozoa is injected slowly into the cave uteri.
17. Finally the catheter is slowly extracted out of the uterus.

Advice

If the sperm is not liquefied sufficiently 30 minutes after ejaculation, liquefy it by aspirating it into a sterile disposable syringe (2 or 5 ml) and flushing it out several times. Before doing this, let disturbing rude particles sediment and do not aspirate them into the syringe.

It is recommended that the sperm concentration is analysed prior to insemination. At least 2 million grade A spermatozoa should be present. An insemination with lower than 0.5 million/ml motil spermatozoa is not recommended.

If performed optimally, the sperm suspension should contains no or very few immotile spermatozoa.
**Thawing**

1. In a 4-well culture dish place 1 ml of each EmbryoStore Thaw thawing solution (1, 2 & 3). This leaves 1 well empty to retrieve the frozen/thawed embryos.

2. Prepare a water bath at 37 °C to thaw the straws. Prepare a 1 ml tuberculin-syringe by filling it with 0.8 ml of air first followed by 0.2 ml of EmbryoStore Thaw 1 medium.

3. Remove the straws from liquid nitrogen and leave at room temperature for about 5 seconds.

4. Submerge the straw in the water bath at 37 °C for another 5 seconds (ensure no frozen part remains in the straw).

5. Empty the straw by opening both ends of the straw (above the empty well) and blowing the contents of the syringe through the straw.

6. Retrieve the embryos and place them in EmbryoStore Thaw 1 thawing solution.

7. Transfer the embryos to EmbryoStore Thaw 2 thawing solution after 3-5 minutes.

8. After another 3-5 minutes the embryos are transferred to EmbryoStore Thaw 3 thawing solution. Leave for a further 3-5 minutes before proceeding.

9. At the final stage the embryos are transferred in IVF culture medium (e.g. GM501 Cult media) for washing and further culture.

**Freezing**

1. Using a sterile pipette place 1 ml of EmbryoStore Freeze medium in a centre well dish (at room temperature).

2. Add the embryos to the freezing medium and allow them to settle for about 30 seconds.

   **Caution:** Due to density differences, the embryos tend to float upwards and shrink.

3. Load the embryos in straws leaving about 1/5 air in the straw.

4. Seal the straws and label with name, date and number of embryos.

5. Start freezing program within 5-10 minutes. Below is an example of a freezing protocol.

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<td>-2 ºC/min</td>
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<td>Phase 3</td>
<td>-6 ºC (autoseeding)</td>
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<td>Phase 4</td>
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<td>Phase 5</td>
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**Vitrification**

**Preparation**

Ensure all media are warmed up to 37 °C and mixed well before use.

**We would strongly advise you to read through all the steps of the vitrification/thawing procedure before starting the procedure.**

**Preliminary steps**

1. In a 4-well dish fill the first well with 300 µl of VitrStore Pre-vitrification Medium, the second with VitrStore Freeze 1 and the third with Vitr-Store Freeze 2 solution.

2. Make sure that the liquid nitrogen is available to ensure fast work flow.

3. Next open as many packs of vitrification devices as will be required for the vitrification step. Place the separate parts of the vitrification device on the workbench for easy access later in the procedure.

**Vitrification**

1. Using an attenuated pipette or an equally suitable device, place a maximum of 2 blastocysts in a volume of approximately 0.3 µl of VitrStore Freeze 2 on the tip of your vitrification straw.

2. Place the vitrification straw in the outer sheath and seal it as indicated in the instructions for use of the vitrification device.

3. Plunge the sealed device into the liquid nitrogen.

* Before starting the vitrification procedure, in order to reduce the negative effect of the blastocoel, expanded blastocysts should be collapsed by reducing the volume of the blastocoel artificially with a glass pipette. (Vanderzwalmen et al., 2002; Sonet al., 2003; Hiraoka, 2004)
**Vitrification**

**Preparation**
Ensure all media are warmed up to 37 °C and mixed well before use.
We would strongly advise you to read through all the steps of the vitrification/thawing procedure before starting the procedure.

**Thawing**
1. In a 4-well dish fill the first well with 300 µl of VitriStore Thaw Medium 1, the second with VitriStore Thaw Medium 2, the third with VitriStore Thaw Medium 3 and the 4th with VitriStore Thaw Medium 4.
2. Remove the vitrification straw from the outer sheath as indicated in the instructions for use of the vitrification device.
3. Immediately plunge the vitrification straw into pre-heated VitriStore Thaw Medium 1 (37 °C) and leave in Thawing 1 medium for 3 minutes.
4. Transfer into VitriStore Thaw Medium 2 (37 °C) and leave in this medium for 2 minutes.
5. Transfer into VitriStore Thaw Medium 3 (37 °C) and leave in this medium for 2 minutes.
6. Finally transfer into VitriStore Thaw Medium 4 (37 °C) and wash for at least 1 minute.
7. Transfer into blastocyst culture medium for continued cell culture (e.g. GM501 Cult media).

**Sperm freezing**

**Preparation**
Ensure all media are well mixed before use.

**Before freezing**
In case of very low sperm concentrations it is advisable to concentrate the sperm before freezing. GM501 Gradient can be applied before freezing to remove debris and to enrich the concentration of motile cells in a sample. This may increase sperm quality after thawing and will reduce the number of straws to be frozen. GM501 SpermStore needs to be warmed to room temperature before use to avoid cold-shock.

**Freezing**
1. Allow the semen to liquefy at room temperature for 30 minutes.
2. Mix 1.00 ml of sperm with 0.70 ml GM501 SpermStore. Add the medium in drops while gently swirling.
3. Leave the mixture for 10 minutes at room temperature for equilibration.
4. Aspirate the sample/medium mixture into the freezing straws, leaving approximately 1.5 cm of air at the end of the straws.
5. Seal the straws.
6. Dry off individually with a lint-free wipe.
7. Shake to move the air-bubble to the centre of the straw.
8. Place the straw horizontally on a styrofoam board in a liquid nitrogen bath to allow for freezing in vapour phase. Leave for (at least) 15 minutes.
9. Transfer straws quickly into liquid nitrogen and store at -196 °C.

**After thawing**
If necessary, use sperm preparation techniques after thawing the semen to eliminate dead sperm cells and debris.
1. Warm the Bromelain in Dulbecco’s PBS to 37 °C.
2. Dilute the semen sample with the same volume of Bromelain (for highly viscous, tenacious ejaculates we recommend to grind the ejaculate roughly beforehand.
3. Swirl the semen solution carefully.
4. Incubate the ejaculate for approx. 10 minutes at 37 °C.
5. Use the liquefied semen sample for evaluation.
6. Continue IVF treatment according to internal standard procedures.

Attention: To calculate the sperm cells concentration the dilution with factor of semen with Bromelain must be accounted.

WHO laboratory manual-quote: These treatments may affect seminal plasma biochemistry, sperm motility and sperm morphology, and their use must be recorded.

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Collagenase treatment

1. Transfer 1.5 ml GM501 Collagenase into a 5 ml round-bottom centrifugation tube.
2. Warm the GM501 Collagenase at 37 °C. GM501 Collagenase is HEPES-buffered. Incubation in a CO₂ incubator will lower the pH.
3. For digestion of testicular tissue carefully pick up the chosen tissue pieces with a fine syringe cannula. For easier handling, if necessary, fill the tissue suspension into a 60 mm petri dish. Let adhesive transport or cryo medium drop off occur as much as possible and transfer into Collagenase tubes.
4. Close the tube completely and place in the incubator (or ideally in a heat cabinet for digestion of the tissue) for 60 minutes. Slight agitation every 20-30 min will facilitate the formation of a single cell suspension.
5. Suspend the digested tissue by carefully pipetting up and down. Under ideal conditions a suspension of single testicular tissue cells and free semen cells has been formed. If coarse tissue pieces are still visible, repeat step 4 for a further 20 to 30 minutes.
6. Now centrifuge the tissue cell suspension and wash twice with 1–2 ml HEPES-buffered sperm processing medium (e.g. GM501 SpermAir). Discard the obtained supernatant. Alternatively, the cell suspension can be processed using a density gradient system (e.g. GM501 Gradient).
7. Resuspend the pellet in a small volume of 30–80 µl HEPES-buffered sperm processing medium. Add a few µl of this suspension into a dish.
8. Continue IVF treatment according to internal standard procedures.
9. If no motile sperms can be found Gynemed recommends the application of GM501 Sperm-Mobil.
**GM501 SpermMobil**

1. Do not equilibrate GM501 SpermMobil in a CO₂ incubator, just warm up to 37 °C.

2. To facilitate sperm activation add 1.50 - 2.00 µl GM501 Sperm Mobil to the sperm cells containing drop (approx. 30.00 - 40.00 µl/dilution 1:20) of processing media inside the Petri dish.

3. GM501 SpermMobil should be added to the opposing side from where the sperm cells are to be aspirated.

4. Wait for 10 minutes. The activating effect initiates after a few minutes and lasts approximately for one hour.

5. The dish should be placed on a heating plate at 37 °C during the diagnostic evaluation.

**GM508 CultActive**

1. GM508 CultActive must be shaken directly before use for approximately 30 sec.

2. GM508 CultActive must be equilibrated 4 hours in a vial not firmly closed at 5 - 7% CO₂ and 37 °C prior to use.

3. Equilibrate culture medium for washing (e.g. GM501 Cult) for 4 hours in a vial not firmly closed at 5 - 7% CO₂ and 37 °C prior to use.

4. Prepare for each oocyte 1 drop (30.00 µl) GM508 CultActive and 2 drops (30.00 - 50.00 µl) culture medium MOPS and HEPES free, (e.g. GM501 Cult). An oil overlay of the drops using suitable oil (e.g. GM501 Mineral Oil) is recommended. Please be aware that protein-free media drops (e.g. GM508 CultActive) can exhibit slightly different dynamic properties compared to other media.

5. Immediately after the ICSI procedure incubate the oocytes for 15 minutes in the pre-equilibrated Ca²⁺-ionophore GM508 CultActive drops. (See picture below - Step 1 - Activation)

6. Remove the oocytes from the GM508 CultActive drop and wash twice in culture media. This has to be done in a HEPES or MOPS free media, e.g. GM501 Cult media. (See picture below - step 1 and 2 - washing)

7. Put the oocytes in your culture medium for further culture.

8. Assess the development on select time points.
Kitazato IVF catheters and needles
NEEDLES & CATHETERS

The Kitazato range of oocyte retrieval needles, with their steel triple cut blades, are designed to provide a fast puncture, minimising damage to the ovarian tissue. Good control of the puncture, allows a reduced operation time helping oocytes to be retrieved in the best possible condition. Kitazato OPU needles offer the widest range of diameters on the market – from 16 G to 21 G. This provides ideal options for working with patients with low response in natural cycles and allows follicular aspiration to be undertaken, without an anaesthetic, using the smallest diameters.

**Oocyte Retrieval Needles**

Needles designed to maximise control and patient comfort.

*Improved patient comfort*
*Maximum control*
*Reduced procedure time*
*Larg range of sizes*
*Single or dual lumen*

### Kitazato OPU Needles

Kitazato OPU needles offer the widest range of diameters on the market from 16 G to 21 G

<table>
<thead>
<tr>
<th>Order Number</th>
<th>Description</th>
<th>Gauge</th>
<th>Length</th>
<th>Aspiration Line</th>
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<tbody>
<tr>
<td>FDM326350</td>
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<table>
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**Kozato OPU Needle - Double Lumen**

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**Other**

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### Technical Specification

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</tr>
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</table>

### Quality Control

- **MEA (Mouse Embryo Assay):** One cell assay > 80% after 96 hours
- **Endotoxin:** < 20EU/device
- **SAL 10**
- **Cytotoxicity Test**
- **Intracutaneous Reactivity Test**
- **Sensitisation Test**

### Composition

- Stainless Steel SUS304
Embryo transfer catheters

ET catheters are designed to improve ease of insertion, control and patient comfort to maximise implantation.

The Kitazato ET catheters, with their smooth malleable tip and guide with widened entrance, are designed for easy atraumatic insertion whilst maintaining their shape.

The superior smoothness and friction-free materials provide improved comfort during transfer while the ergonomic handle allows for safe and precise handling. Stylets are available which moulds and provides rigidity to the guide during the most difficult transfers.

- **Maximum control**
- **Improved patient comfort**
- **Easy Insertion**
- **Large range of sizes & types**
- **Available straight or precurved**

**EC-PRO Routine catheter:** The guide shapes to comfortably and quickly access uterine cavity with the precision, provided by its ergonomic handle. Available supported or unsupported and with or without guide.

**EC-PRO Trial catheter:** Closed tip catheter to examine cervix before transfer and to determine its difficulty. Avoids entry of cervical mucus in guide during channeling and guarantees an easy and clean insertion of the transfer catheter.

### TECHNICAL SPECIFICATION

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<tr>
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<td>20 cm</td>
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*Kitazato Precurved ET Catheters are sold in boxes of 10 units. **EC-PRO Diameter: 4.7 Fr.*

**Quality Control**

- **MEA (Mouse Embryo Assay):** One cell assay ≥ 80% after 96 hours
- **Endotoxin:** < 20EU/device
- **Sterility (Bacteria, Fungi):**
- **Intracutaneous Reactivity Test:**
- **Sensitisation Test:**

**Composition**

- **EC-PRO ET catheters**
  - Catheter: Polyurethane
  - Guide: Fluorocarbon Resin
  - Sterilisation: Ethylene Oxide Gas Irradiation
- **EC-PRO Routine ET catheters**
  - Catheter: Silicone, Stainless Steel SUS304
  - Guide: 12 Nylon
  - Sterilisation: Gamma Irradiation
To allow for different users’ preferences, the Kitazato IUI catheter range is available in two different lengths (10 & 18 cm), two different gauges (5.2 and 6 Fr) and four different levels of rigidity (hard, ultimate, intermediate and soft).

The smooth and rounded tip allows for easy insertion and the double lateral opening tip improves the dispersion of the semen.

- Improved semen dispersion
- Easy insertion
- Large range of sizes
- Variety of catheter rigidity options

Intrauterin Insemination catheters (IUI)

Improve your artificial insemination success rates with the Kitazato range of IUI catheters, which are available in 4 levels of rigidity and two different IUI catheter lengths.

### TECHNICAL SPECIFICATION

<table>
<thead>
<tr>
<th>Order Number</th>
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*Kitazato IUI Catheters are sold in boxes of 10 units.

**Quality Control**
- MEA (Mouse Embryo Assay): One cell assay > 80% after 96 hours
- Endotoxin: < 20EU/device
- SAL 10^{-6}
- Sterility Test (Bacteria, Fungi)
- Cytotoxicity Test
- Intracutaneous Reactivity Test
- Sensitisation Test

**Composition**
- Polyvinylchloride DEHP free
- Stainless Steel SUS304

*Kitazato IUI Catheters are sold in boxes of 10 units.*
Visit our website, to find out more about our range of products.
www.planer.com