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Instructions for VitroTemp

Temperature measurements of media on heated surfaces



Background

VitroTemp™ is a thermometer specifically designed for temperature measurements of media in Vitrolife's square dishes while handled outside the incubator.

The temperature of the medium in a dish will depend on the type of dish used and the dish set-up, such as medium volume, if an oil overlay is used, or if the lid is on or off during the measurement. For this reason, VitroTemp comes with two different types of dishes/probes (Culture dish 40 mm and 5 Well culture dish). With these two types of dishes/probes, you will cover your need for accurate temperature measurements.

With VitroTemp, the measurement is through temperature dependent resistance in a small platinum plate (Pt100 probe), that is attached to the bottom of a Vitrolife dish. The high quality of the measurements performed with this custom-made device depends on:

- the sensitivity and accuracy of the actual Pt100 sensor
- the fact that the device measures temperature exclusively at the small platinum plate without any thermal conductivity disturbance
- the device design allowing temperature measurement of the medium at the bottom of the dish (i.e., mimics oocyte/embryo position)

Note, this custom-made temperature probe, shall never be in contact with gametes and embryos, it can exclusively be used for temperature measurements and never for holding or culturing gametes and embryos.

Temperature measurement

- 1** To measure the temperature of a medium in a dish during manipulations outside the incubator is very simple and straight forward when using this custom-made device.
- 2** If the measurements are performed in a correct way (the method and the timing), the measurements will be stable and repeatable. Note, if the temperature fluctuates during your measurements or varies between measurements, it is likely caused by surrounding factors rather than by the probe itself.
- 3** To prepare a VitroTemp probe/dish for temperature measurement, dispense medium and/or oil into the dish according to your normal procedures during routine operations in the lab.
- 4** Place the probe/dish on a heated surface and allow it to equilibrate fully before you take the reading.
- 5** Note, it will take 10-20 minutes for a medium in a dish to reach full temperature equilibration when placed on a heated surface, depending on the dish set-up.
- 6** It is very important to make sure that the dish stands flat on the heated surface during equilibration and temperature measurements. If not, the temperature reading will be incorrect. If this is difficult, mainly due to the cable of the probe, one can facilitate the attachment of the dish to the surface with a non-sticky tape. This will also minimise the spilling oil or media during the measurements.
- 7** When measuring the medium temperature using micro-droplets under oil, there is no need to add any medium since you will get the same temperature reading by taking oil alone. This is not the case when working with larger volumes of medium and oil, such as in the 5 Well culture dish.

- 8** The equilibration times mentioned above, as well as the equilibrated temperature, are valid for medium at the bottom of the dish where the sensor is located (as well as the gametes and embryos). Be aware that there will be a gradient between the medium temperature at the bottom of the dish and the surface. This temperature difference can be significant when working with larger medium volumes like during denudation and vitrification.
- 9** Remember, that the temperature of the medium in a dish will depend on the dish type and the dish set-up. Therefore, we recommend that you use a dish type and a dish set-up mimicking that used during routine work on each specific heated surface. In short, mimic your clinical practice.
- 10** If you use several different types of dishes or dish set-ups on the same heated surface, we also recommend that you set the temperature for the “warmest case scenario” (dish type and dish set-up that gives the highest temperature) to avoid that you expose gametes and embryos to temperatures above 37.0°C.
- 11** If a heated surface will be used for any dish with the lid on, that specific heated surface should be calibrated by a probe/dish with the lid on. Medium temperature in a dish with the lid on will be significantly higher than in a dish without a lid. This is true even when an oil overlay is used.
- 12** The same applies for the use of an oil overlay, i.e., the medium temperature in a dish will differ with or without the use of oil. Note, an oil overlay will for sure stabilise the temperature during handling outside the incubator.
- 13** Note, the temperature in dishes placed on heated surfaces is also dependent on surrounding factors, such as the laboratory room temperature or whether working in a ventilated laminar air flow bench or not. If for example the laboratory temperature goes up a few degrees, the medium temperature will increase as well.

- 14 When working on heated surfaces, it is important to “know” your heated surfaces. Therefore, validate the temperature of any heated surface over time. Also map the temperature of your heated surface to verify that the temperature is consistent and stable on its entire surface.
- 15 When working with mammalian gametes and embryos, make sure that the temperature in the media never exceeds 37.0°C. Therefore, be aware of the inaccuracy of your thermometer and your measurement method and use appropriate safety margins to not exceed 37.0°C.
- 16 Always use the thermometer accurately calibrated (to use the accurate correction factor). You can find the current and latest Calibration Certificate automatically by just scanning the QR-code on the probe. If the calibration is on its way to be outdated, you have to send it away for an annual calibration.
- 17 One final thing that is important to highlight is that Vitrolife square dishes, including the dishes of VitroTemp, create direct contact with the heated surface. This means that heated surface temperatures set for Vitrolife square dishes will not be accurate for dishes without direct contact to the heated surface!

Cleaning after measurements

- 1 After measurements, we recommend that you aspirate as much of the medium and oil as possible by using a pipette. You can then rinse the dish with distilled water or 70% ethanol solution a couple of times and then finally wipe the dish clean with a lint free cloth.
- 2 The accuracy and the function of the probe will not be affected by how well you have cleaned the probe. The cleaning process is more for visual and practical reasons (to avoid soiling various surfaces with oil).

Calibration

- 1** The VitroTemp probe needs to be calibrated on a regular basis. Regulatory authorities recommend that you calibrate thermometers for IVF annually.
- 2** We recommend you calibrate the thermometer at 35, 37 and 39°C in order to cover measurements at physiological temperature, as well as temperatures below and above this temperature.
- 3** Send the thermometer and the probe to an organisation certified for this type of calibration. Calibrate according to standard procedures for this type of probe. Note, the custom-made probe should be calibrated exactly as the procedure for an insertion probe. In short, that means that during the calibration the whole probe, i.e., the sensor together with the dish is submerged in the calibration liquid.
- 4** You can find the current and latest Calibration Certificate automatically by just scanning the QR-code on the probe.

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