BENEFITS OF NDIR OVER TC CO2 SENSORS INSIDE HIGH HUMIDITY CO2 INCUBATORS



Inside cell culture laboratories CO₂ incubators are one of the most important tools to grow living cells. These cells can vary from simple cell lines to human embryos grown for IVF purposes. The incubators mimic the human body as closely as possible to make sure the cells feel comfortable and grow and multiply successfully. Alongside a stable temperature, pH level is very important to make cells grow well. CO₂ sensors in combination with CO₂ gas are used to get the pH value to a correct level. This means it is very important to make sure the CO₂ reading inside incubators is as accurate as possible.

The CO₂ incubator as a black box

XiltriX International has been installing Temperature & CO_2 monitoring systems inside incubators for more than 30 years. Over time a lot has changed, especially

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 CO_2 sensor technology has evolved tremendously. Laboratory staffwho have witnessed this evolution can still remember the difficulty of measuring CO_2 by manually using a chemical based <u>Fyrite meter</u>. When used correctly an accuracy of roughly ± 0.5% could be achieved. One of the first measuring principles to supersede the Fyrite meter was the widely used TC (<u>Thermal Conductivity</u>) CO_2 sensor. This sensor used the electrical resistance through the air to measure how much CO_2 was present inside the incubators. When it detected too little CO_2 , it would let in more gas. When all parameters in the incubators were correct, the accuracy of the CO_2 sensor was acceptable, but it had one fatal flaw; it was terribly susceptible to changes in humidity.



The next evolution of the CO₂ sensor was the NDIR (<u>Non Dispersive Infrared</u>) CO₂ sensor. After development the NDIR sensor had one downside to the TC sensor, it was a lot more expensive. It also suffered from drift. After many years of development though the current generations of NDIR sensors are mostly very stable and will run for prolonged periods of time without the need for frequent manual calibrations.



Now back on topic: why incubators are considered a black box. When asking a lab tech what the CO₂ percentage in their incubator is, they will probably look at the display and tell you the value they see. They usually do not have an idea what sensor technology is involved in deriving this value and therefore cannot judge if this value is correct. Most incubators in the market today are using NDIR sensor, but not all. I will explain the risks of using a TC sensor inside an incubator using a real live example.

Risks of using a TC CO_2 sensor inside a CO_2 incubator

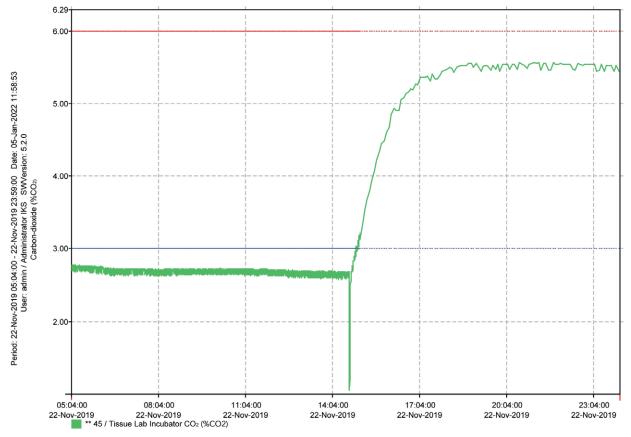
XiltriX solely relies on using NDIR sensors for monitoring purposes inside CO_2 incubators. Depending on the brand and type of incubator multiple form factors are being used. Some sensors can withstand high temperature decontamination, others fit inside a smaller access port, but all are very accurate. One of our customers contacted us with a question. They mentioned the XiltriX sensor was consistently measuring 3% lower than the 5.5% display value on their CO_2 incubator. The customer told us the incubator had not been opened for several days not disturbing the culture and they did not own a handheld CO_2 sensor to check whether the value of the incubators was correct, or the value shown on XiltriX.



Not having an accurate handheld CO₂ analyzer in these kind of situations can be really tricky. Without the ability to measure CO₂ or pH, it is nearly impossible to gauge which of the devices is showing the correct value. This customer did not have any reason to suspect the incubator, since it was very recently serviced & calibrated by the manufacturer. The device also did not sound any alarms or show any signs of fault externally.

After remote consultation by the XiltriX technical team, the customer was asked to check whether the sensor inside the incubator was positioned correctly and if any signs of condensation were present. Condensation could influence the reading of the NDIR sensor if it the condensate would have dripped inside the sensor tip and might block the light source or detector. After checking the incubator the CO₂ values started to slowly recover. There was no indication of anything wrong aside from a fleeting comment of the customer that he had refilled the water tray of the incubator.





CO2 level recovered to normal value after refilling the water tray

This fleeting comment triggered alarm bells in our tech team. Our engineers started inquiring about the CO₂ sensor and found out it was of the TC type. This is not always clearly marked on the outside of the incubator and sometimes one type of incubator is supplied with multiple types of sensors. The customer was definitely not aware of the type of sensor used. Without the knowledge of what sensor was being used the graph above did not make any sense. Knowing now that a TC sensor was involved made everything very clear.

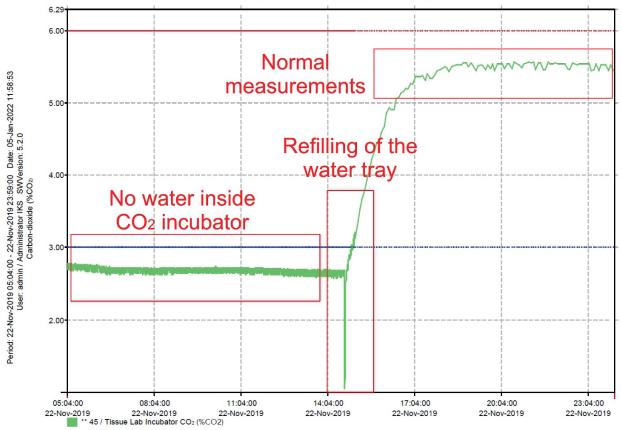
The incubator had slowly run out of water. The water had naturally evaporated and had not been refilled in a timely manner. Because the cells inside the incubator were not so susceptible to excess evaporation of culture media, the lab techs did not think anything of it. What they failed to recognize though is the fact the lack of humidity also changed the Thermal Conductivity of the air, making the incubator "Think" it was running at 5.5% CO₂, but actually only operating at 2.5% CO₂ concentration.

Why the water tray had run out of water is not clear. It might have had something to do with the amount filled in the first place (too little water), human error (forgetting to refill the tray) or simply more frequent door openings causing more humidity to evaporate than was normal for this device. Looking at the graph though you can see that it takes multiple hours for the CO_2 values to return to their normal setpoint, because the evaporation of the humidity is so slow. Needless to say, without an NDIR CO_2 sensor as an independent monitoring device, this failure would have probably gone unnoticed. Running the incubator at such an incorrect CO_2 value though will have had a profound effect on the pH levels of the growth media with the cells growing inside.

Why XiltriX uses independent sensors

In my 20 years of experience in the field of laboratory monitoring I have seen many devices and technologies fail. Some because of unfortunate events, others due to human error human error. One thing did stand out every time: **ONE = NONE**. To rely on a single sensor, one is bound to run into problems at some point. In 2012 the Harvard Brain Bank lost a -80°C freezer with priceless donated brain samples (Harvard Brain Freezer Failure). This incident threw back the research into autism many years. Harvard did invest in monitoring technology. The available information describes separately failed circuits. It is not mentioned however whether these circuits were driven by a single temperature sensors/controller or independent ones.





CO2 level recovered to normal value after refilling the water tray

When only monitoring the onboard controller and sensors, in other words the (small) computer that operates the say freezer or incubator, one is led to believe monitoring is in place, but If the controller or circuitry itself fails, no alarm will be sounded. There is no independent referee in place nor in control.

The device will assume all parameters are running at optimum settings. The empty water pan in the incubator never caused an alarm, and the TC CO₂ sensor cannot detect the difference, it was properly calibrated for use at high humidity. As shown in our example.

The Results

Running a lab without monitoring is an unmitigated risk. Running a lab with a monitoring system built with dependent sensors is equally risky as well. Operating a monitoring system relying on dependent sensors will reduce risks on average, but serious damage is very likely. A critical incident would lead to reputation damage and the very real risk of a liability case. Not mentioning direct losses in labor, investments and Science. On top of that, precious time will be lost in Science because a lot of experiments will have to be redone since the results will not be reproducible.

In our conversations with customers on multiple occasions they ask about the possibility of digitally connecting devices via available communication ports. This in essence is not a bad idea, because it will give monitoring systems more meta data to provide more powerful insights into event happening. This should be seen as an extra option though and not a replacement of independent monitoring sensors. XiltriX will continue to develop and evolve partnership with our customer to provide the most reliable and accurate monitoring solution on the market today.



Do you want help or advice with monitoring of incubators or other devices? Please contact XiltriX at <u>sales@xiltrix.com</u>, or visit our website <u>www.xiltrix.com</u>.