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Missing your Cy5 signal? Tired of drying your slides at dawn? Kludge up a good, cheap ozone-free room for your microarray slide spin drying.



A Good, Cheap Kludge build an Ozone-free room

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Don't lose your microarray Cy5 dye signal to ambient room ozone. Cy5 destruction has been reported after as little as 10-30 seconds exposure to ozone levels above five to ten parts per billion (ppb), especially during the drying and scanning steps.

For additional background information and references, please see our report *The Ozone Annihilator* available at http://cmgm.stanford.edu/pbrown/protocols/Ozone_Prevention.pdf where you will find instructions for building inexpensive ozone-free equipment enclosures.

Here we provide simple plans for a good, cheap kludge. If you can install an industrial quality HVAC ozone filtering system that would be best. Your goal is a final ozone level of one or two ppb, or less.

For a do-it-yourself solution, keep reading. You will need a smallish room, with enough space for a slide washing table and your slide drying centrifuge. You will have to be able to limit the airflow through the room, as small ozone catalyst systems will not be able to clear all the ozone from your entire building. In many laboratories, there will be building HVAC airflow through ducting. You might have to have the vent airflow into your ozone-free room reduced or turned off. Check places where building air can enter the room such as doors etc.

Next, build your recirculating ozone filter. Start with one or more 26"x47" sheets of aluminum honeycomb-like material with an ozone catalyst coating from BASF Catalysts LLC (formerly Engelhard) of Iselin, NJ. You will cut these sheets into pieces to fit a household room air filter. We

used a BlueAir 501, as it has a large filter volume relative to the size of the unit:

A common home air cleaner can be used to recirculate room air through ozone catalyst sheets



The Blueair 501 holds three filter cartridges, each is about 33 x 23.5 x 6.7 cm



and four layers of the honeycomb ozone catalyst sheet just happens to match the thickness of each cartridge. Cut your ozone catalyst honeycomb into enough pieces to entirely replace all three air filter cartridges:

Cut the ozone catalyst to fit your air cleaner. Pick a unit that lets you use multiple layers of catalyst.



You will find that it is easy to slide the twelve pieces of cut catalyst into the room air filter, leaving you with a self-contained rolling ozone recirculating catalyst unit.

IMPORTANT NOTE: The BlueAir 501 contains a high voltage circuit that appears to be used to charge the air passing through it. We think this is probably intended to cause dust to stick to the inside of the unit. Unfortunately, this high voltage also generates some amount of ozone, which is not good.

We measured the ozone level in our room both with and without the circuit working, and found that the level was several parts per billion higher with the high voltage connected. It was not possible to get down to 1 ppb with the circuit operating, so we disabled the high voltage circuit.

You can of course substitute any kind of fan or blower that will recirculate the room air through your catalyst. The catalyst needs some contact time to work, so you might want to experiment with different air velocities and find the most effective setup for your room.

Naturally it is important to verify that your ozone-free room is functioning. We borrowed a Teledyne Photometric O3 Analyzer, Model 400E to measure our ozone level. You might be able to find one by contacting local air pollution or environment offices, as this instrument is approved by the EPA for ambient air monitoring.

<http://www.epa.gov/fedrgstr/EPA-AIR/2002/September/Day-12/a23261.htm>



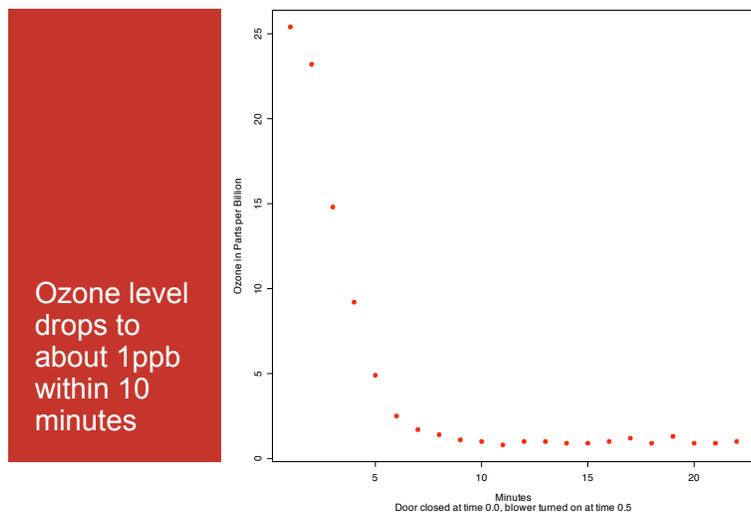
The manufacturer is Teledyne Instruments, Advanced Pollution Instrumentation Division, 6565 Nancy Ridge Drive, San Diego, California. <http://www.teledyne-api.com/products/400e.asp>

To test, we leave the door to our ozone free room open for a few hours and check the ambient reading which matches building air. Then we close the door and turn on the air cleaner / catalyst to "MAX"



Today the afternoon ozone level was around 25 ppb in our building. Here are measured results for

ozone in ppb vs time in minutes for our 10' x 9' by 8.5' closed room, with the door closed and air cleaner / catalyst turned on:



NOTES ON DRYING MICROARRAY SLIDES

As previously mentioned, ozone levels of even five or ten ppb may destroy Cy5 dyes. The most sensitive time appears to be while transitioning from wet to fully dry.

One method that has worked well for us is to perform the final wash in an ozone free room. Prepare a matching rack of centrifuge balance slides if needed. Then, remove your slide rack from the final wash and gently but firmly place onto a stack of dry absorbent material, in order to remove any large drops of solution from the bottom of the slide rack.

Quickly place the slide rack into your centrifuge. You want to have the slides dry by centrifuge moving the liquid off the slide, and not by evaporation. Evaporation could result in salts or hybridization solution drying onto the slide. We use a plate type rotor and run at about 200g for four or five minutes.

Avoid letting your slides touch anything with fluorescence compounds, most common paper towels do contain 'whiteners' that fluoresce brightly in the green channel. Filter paper appears to be better in this respect.

Once your slides are dry, keep them in an ozone free environment before and during scanning. See *The Ozone Annihilator* for do-it-yourself instructions to build enclosures for storing and scanning microarray slides.