

Anoxic Microenvironments: A Treatment For Pest Control

Anoxic (without oxygen) conditions may be used to treat insect-infested objects in museums. The procedures employed are relatively simple, and are safe for both people and collections. This makes anoxia an attractive alternative to toxic fumigants. However, the oxygen levels needed to kill insects are so low that an anoxic microenvironment must be carefully designed and constructed to be effective. This leaflet describes how to create anoxic microenvironments using heat-sealable barrier films and inexpensive oxygen absorbers, sometimes called "oxygen scavengers." For a discussion of the various non-toxic alternatives to pesticides see *Conserve O Gram* 3/8.

Materials Needed

Barrier Films: Anoxic microenvironments for killing pests can be made from flexible plastic films heat-sealed into a pillow-shaped bag. A highly efficient plastic, called a barrier film, must be used to maintain the critical conditions.

Barrier films are usually laminates that combine the benefits of several layers; one may be a gas or moisture barrier, while another provides puncture resistance or the capability to heat-seal the film together. The efficiency of the final laminate film can be rated in terms of its water vapor transmission rate (WVTR) or its oxygen transmission rate (OTR), often expressed as the amount of material (grams of water or cc of gas) that can pass through one square meter of the film in 24 hours.

For anoxia, the OTR of the film must be lower than the rate at which the oxygen scavenger can absorb oxygen. For pest control treatments that typically take 10 days or longer, barrier films with oxygen transmission rates under 100 cm/m^2 are suitable. The most efficient laminates contain a thin layer of aluminum sandwiched between layers of nylon or polyester (e.g., MarvelSeal 360[™]). The transmission rates of such films approach zero, and are thus excellent choices for microclimate construction. These films are among the least expensive barrier films available, but you cannot see through them. More expensive transparent barrier films based on polvester (PET, trade name Mylar[®]). polyvinylidene chloride (PVDC, trade name Saran[®]), or ethylene vinyl alcohol (EVOH, trade name OxyShield[®]), are useful for the techniques described in this leaflet.

Most barrier films include a polyethylene layer on the inner surface that enables them to be heatsealed at temperatures around $117^{\circ}C$ ($350^{\circ}F$). Because polyethylene is such a poor oxygen barrier, a thick seam is necessary to match the transmission characteristics of the rest of the film. Therefore, well fused heat-seals wider than 3/8" (1 cm) are essential. Most anoxia failures are due to faulty seams.

A variety of heat-sealers are available, from hand-held models costing a few hundred dollars, to much more expensive tabletop models. It is possible to use a tacking iron as long as it gets hot enough to fuse the plastic, but if you plan to use this technique regularly, a more expensive dedicated model with long 3/8" wide elements is well worth the price.

Oxygen Absorbers: The brand of oxygen absorber commonly used in anoxic microenvironments is called Ageless[™] (Mitsubishi Gas Chemical Company, Inc.), although other brands are becoming available. Ageless is composed of finely divided ferrous oxide, a chloride salt, and a humectant sealed inside a permeable packet. A number of packets come vacuum-sealed inside a transparent oxygen barrier pouch along with a single in-package oxygen monitor called an Ageless Eye[™].

Ageless is available in several types and capacities. The type normally used for pest control, Ageless Z, is effective at relative humidities between 35% and 85%. The absorbing capacity is rated by the amount of oxygen that a single packet can absorb. For example, Ageless Z-1000 will absorb one liter (1000 ml) of oxygen before it is exhausted, and Z-500 half that. Since the atmosphere is about one-fifth oxygen, this means that one packet of Ageless Z-1000 can remove all the oxygen from five liters of air. It's helpful to remember that it takes about six packets of Ageless Z-1000 to remove the oxygen from one cubic foot of air.

Every pouch of Ageless packets includes an Ageless Eye. This is a small tablet containing a chemical that changes from purple to pink when the oxygen concentration falls below 0.5%. At concentrations above 0.5% oxygen the pill quickly changes back to purple. The tablet can be reused several times, but after a while may become slow to respond, or may not change color at all, rendering it useless. Extended exposure to oxygen will deteriorate the eye, as will storage at higher temperatures. The eye will also fade after long exposure to light.

Each Ageless Eye is enclosed in its own tiny cellophane envelope, which is perforated to allow air circulation. The envelope should not be opened, but should be used to tape the Eye to a readily visible location.

Nitrogen Flushing: This is an optional practice that will help flush out most of the oxygen at the beginning of treatment. Nitrogen flushing is recommended because less oxygen will be left for the Ageless to absorb and there will be less tendency for the Ageless to heat up and produce humidity. There will be less collapse in the bag because the nitrogen will remain in the bag while the oxygen is absorbed.

Constructing an Anoxic Microenvironment

Follow this six-step guide to create a useful microenvironment.

1. Making the bag: First, estimate the size of the bag needed. Make the bag as small as practical. A larger bag will have longer seals, increasing the likelihood of leakage. It will also require more Ageless. Cut the plastic carefully so that the opposite sides are the same size. When heat-sealing a bag, it is easier to avoid leaky crimps and wrinkles if the sides match. Cutting twice the length of the bag from a roll and folding it over avoids an unnecessary seam.

Practice seals on scrap film before beginning the final product. To test your process, heat-seal two pieces of barrier film together, let them cool for several seconds, and then tear the seal apart. If the heat is too low, the pressure not great enough, or the heating time too short, the seal will peel apart with little or no damage to the film. If done correctly, the layers of the laminate will separate or tear before the seal lets go. A few quick tests will make this clear.

If the barrier film is opaque, it is necessary to make a small window in the bag so the Ageless Eye can be observed. This is a difficult operation, and often results in leaks that render the bag useless. Carefully cut a 1" square hole in the barrier film, and a 3" square piece of clear plastic from a discarded Ageless pouch. Using a small tacking iron, carefully heat-seal the inside surfaces of both films together to make a window, keeping in mind that the clear plastic is more sensitive than barrier film. Practice with scrap pieces first. Always make the window before making the bag, since a failure here means starting completely over.

2. Preparing the Object: Before the object or specimen is placed inside the barrier bag, determine whether it needs any special

preparation. Will the object be harmed by the weight of the plastic or any collapse in the volume of the bag? Remember that if the bag is not flushed with nitrogen, the volume of the bag will collapse by 20%. Are there any sharp points on the object that can puncture the bag itself? To protect an object you can wrap it in acid-free paper or place it in a cardboard box, since oxygen will move freely through these materials.

Always test the closure of the bag to make sure that the final side can be easily sealed shut before proceeding.

3. Calculating the Amount of Oxygen

Absorber: Six Z-1000 packets per cubic foot is a handy rule of thumb to use when making an enclosure. A more accurate calculation is:

volume of bag in cm (LxWxH) – weight of object in grams / 5 = ml of oxygen in bag

In practice, it is common to use two or three times more Ageless than is actually needed. This may seem inefficient, but is far better than a complete failure of the treatment due to incorrect calculations or a faulty seal. If just the minimum amount of Ageless is used, absorption may be so slow that several days can pass before problems are discovered. If, however, an excess of Ageless is used, any problems will typically be evident within 24 hours, and there will be a sufficient reserve of Ageless available after unexpected leaks are located and resealed.

After the treatment, partially used Ageless packets may be sealed up in a barrier bag with an Eye and refrigerated for another use until they are exhausted. With every new microenvironment, use fresh Ageless as the primary absorber, and the secondhand packets as reserve.

4. Employing the Oxygen Absorber: Cut open the Ageless pouch and insert the calculated number of packets. There's no need to hurry. The oxygen absorber reacts slowly at first, especially if it has been refrigerated. Distribute the packets around the interior of the bag so that there is a maximum amount of Ageless packet surfaces exposed. Avoid clumping the packets together, and try to prevent the Ageless packets from directly contacting the object or specimen.

As an Ageless packet reacts with oxygen, some heat and moisture is generated. In normal air, packets of Ageless have been observed to get as hot as 46° C (115° F), especially if clumped together. Anoxia is less effective in humid conditions. To remedy this, flush the bag with nitrogen to remove most of the oxygen before sealing. This will dramatically reduce heat and moisture. If moisture and heat could adversely affect the object, buffer the environment by wrapping objects in paper or cloth.

Using small pieces of adhesive tape, secure an Ageless Eye in its cellophane envelope inside the bag where it will remain visible. Be careful not to cover the perforations in the envelope with tape. Since the Ageless Eye is so prone to failure, it is safer to use two or three. As long as one turns pink, the microenvironment is a success. Place an Ageless Eye on the outside of the bag for comparison, as the beginning shades of color change are subtle. The comparison will be helpful.

5. Sealing the Bag and Flushing with

Nitrogen: Seal the bag using the heat-seal technique. Take your time and be careful; this is the most critical step. If you are going to flush the bag with compressed nitrogen from a cylinder, you will need a tank of compressed nitrogen, a pressure regulator, and a hose and nozzle. Seal the final side of the bag completely except for a 2" section. Use the rubber hose and nozzle to inject lots of nitrogen and displace the air inside the bag before final sealing. Leaving the bag slightly inflated helps protect the object and also makes finding leaks easier.

6. Checking for Leaks: If you plan to make anoxic bags frequently, a flammable gas detector that gives an audible signal in the presence of certain gases is essential. Although gasses like butane, propane, and freon are very effective, an alternative is ozone-safe canned air. Test each brand before use, since some of the newer formulas give very weak signals. To test, inject a small amount of the gas into the bag just before it is sealed up. You can also inject a sealed bag with a large hypodermic needle, and patch the hole with high efficiency pressure-sensitive barrier tape (e.g., 3M aluminized polyester tape #850). Let freshly heat-sealed areas cool before using a detector, since vapors from the melting plastic can give false readings. Move the detector head slowly around the outside of the bag very close to potential sources of leaks, such as seams, folds, windows, or areas that may have been creased or punctured. Loose seams can often be fixed with a heat sealer, and small pinholes sealed by covering with a broad piece of barrier tape. Unfortunately, large holes or crimps are difficult, if not impossible, to seal effectively.

Once the bag is sealed, the Ageless Eye may take up to several days to turn pink, depending on the bag size and amount of Ageless. If, after a day or two, the eye shows no sign of change, there may be a leak, a depleted Ageless Eye, or an insufficient quantity of Ageless. Larger bags and colder temperatures can both result in slow response times.

When the eye is definitely pink, mark the date of this change on the outside of the bag. Check again for the first few days to make sure the seals are holding. If there is a slow leak, the Ageless may be consumed before the completion of the treatment, and the Eye will revert to purple.

Length of Anoxic Exposure

A two-week exposure at room temperature is sufficient for insect eradication. Tests have shown that most insects and their eggs will be killed effectively in less than five days at concentrations of no more than 0.5% oxygen, but a few resistant species, like the cigarette beetle,

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may require 10-12 days for 100% elimination. Oxygen concentrations greater than 1% are not effective, even for longer exposures. Anoxia kills insects more by dehydration than suffocation. At very low oxygen concentrations the spiracles of the insect open so wide that the insect essentially desiccates. Dry and warm conditions can accelerate this effect. If conditions are humid or cool, however, the insect may retain sufficient moisture or slow its metabolism enough to render the anoxia treatment unsuccessful. Fungi are not killed by anoxia.

When a microenvironment is ready to be opened, cut it near one of the seals, remove and prepare the Ageless and Ageless Eye for refrigeration as described above. If the empty bag is stored carefully, with minimum folding and creasing, it may be reused several times.

For current sources of materials and supplies contact the NPS Museum Supply and Equipment Program at (304) 535-6072. This *Conserve O Gram* is adapted from "Anoxic Environments: A Simple Guide," SPNHC Leaflet, Vol. 1, No. 1, Spring 1996. It is available on the web at < http://www.geo.ucalgary.ca/spnhc/>.

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John Burke, Chief Conservator The Oakland Museum of California 1000 Oak Street Oakland, CA 94607

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