Alternative Methods for Sterilization and Cutting Disinfestation

M. T. McClelland

McHenry County Nursery, Inc., 8501 White Oaks Rd., Harvard, IL 60033

M. A. L. Smith

Department of Horticulture, University of Illinois,1201 Dorner Dr., Plant Sciences Lab, Urbana, IL 61801

THE PROPAGATOR'S ROLE AS PART OF THE GREEN INDUSTRY

In recent years, increasing evidence has been reported on the adverse effects of chlorine on our environment. Most of the controversy over chlorine pollution has been centered around two sources: (1) chlorinated hydrocarbons, which deplete the atmospheric ozone layer protecting us from damaging ultraviolet rays, and (2) the manufacturing processes using chlorine (e.g., plastic polyvinyl chlorides and paper bleaching) which produce a multitude of toxic chlorine compounds called organochlorines.

Health effects linked to the bioaccumulation of organochlorines in humans and wildlife include breast cancer and reproductive problems. Many of the compounds are carcinogens and mutagens. While working on a community education project on the dangers of chlorine pollution, we became concerned about what chlorinated products we might routinely use as part of our nursery operations.

At McHenry County Nursery, we have made every effort to use sustainable nursery practices in our field production including contour-tillage for soil and water conservation, and integrated pest management to reduce the need for chemical pest control. But what about other departments? What about the bleach used as part of the cutting and pot sanitation procedures? What are we using and where does it go? What are the environmental risks we are taking in some of our routine nursery practices? And if they are damaging, can we establish some cost-effective alternatives that are also environmentally responsible? As stewards of the land—propagators, producers and promoters of living plants—shouldn't we be concerned about the long-term effects of our practices on our environment?

DISINFECTANTS USED IN CUTTING PROPAGATION

The range of chemicals used in nursery production is quite broad, and includes fungicides, insecticides, herbicides, fumigants, nematicides, fertilizers, and disinfectants. A disinfectant is any chemical used to surface disinfest plant material, or to sterilize tools, work surfaces, or other materials. This paper reviews some of the common disinfectants used for material sterilization and plant surface disinfestation in the cutting propagation phase of nursery production. Many of these disinfectants are also used in micropropagation. Although cuttings are not always surface disinfested before sticking, it is a recommended practice because it reduces the need for chemical fungicide and bactericide treatment later (MacDonald, 1986).

A list of common disinfectants is shown in Table 1. Of these, bleach (sodium hypochlorite) has traditionally been the most effective, least expensive and most available. It is used to surface disinfest plant material and to sterilize materials and tools.

Table 1. Common disinfectants used in cutting propagation.

Disinfectant 1	Chemical formula	
bleach (sodium hypochlorite)	NaClO	
bleach (calcium hypochlorite)	$Ca(ClO)_2$	
isopropyl alcohol	$CH_3(CH-OH)CH_3$	
Physan, Triathalon, Green-Shield	benzalkonium chlorides	
hydrogen peroxide	НООН	
calcium peroxide	Ca(OOH) ₂	
mercuric chloride	HgCl ₂	
ethylene oxide	$(CH_2)_2^2O$	

¹ Italicized disinfectants are oxidizing chemicals.

The most effective disinfectants are oxidizing chemicals (Table 1.). There are always by-products after oxidation. These by-products are what make their way into our soils, surface water, and ground water systems (Table 2.). They can combine with other organic compounds in the environment forming compounds that are often more harmful than the original formula.

Table 2. Common disinfectants and their by-products.

Disinfectant	Principle oxidation reaction by-products
bleach (sodium hypochlorite)	sodium, hypochlorite, and water
bleach (calcium hypochlorite)	calcium, hypochlorite, and water
isopropyl alcohol	isopropyl and other minor by-products
benzalkonium chlorides	chloride and other minor by-products
hydrogen peroxide	oxygen and water
calcium peroxide	oxygen and calcium hydroxide
mercuric chloride	mercuric chloride and other minor by-products
ethylene oxide	ethylene glycol and other minor by-products

Each of these disinfectants has advantages and disadvantages to the plant (effectiveness, toxicity); to production (availability, worker safety); to the budget (long- and short-term costs); and to the environment (consequences of use to plants, soil, and water) (Table 3.).

Table 3. Three common sterilants and their effects on the plant, the budget and the environment.

Sterilant	Plant	Budget	Environment
Isopropyl alcohol	At high concentrations dehydrates plant tissue, at lower concentrations is not strong enough to disinfest plants of microorganisms or work surfaces of bacteria	Inexpensive	Toxic to skin and dangerous to breathe
Benzalkonium chlorides	Must use at commercially recommended rates for each formulation	Economical	Basically inert by-products if not used in excess of what is needed
Bleach, hypochlorite solutions	Can be toxic to plant tissue on contact, if concentration is more than 10%; for work surface sterilization usually 1 part bleach to 10 parts water is recommended.	Inexpensive	The chloride ion (OCl) by- products combine with organic compounds in soils and become toxic in all forms—taken up through roots in plants, ingested by animals; excess bleach that enters environment will also oxidize any living tissue it comes in contact with
Hydrogen peroxide	Must use higher concentrations than hypochlorites (30%)	More expensive than bleach; but has many more uses—kills fungus and bacteria used in soil, water, and on plants	Totally non-toxic byproducts of oxygen and water; if used in excess, evaporates before oxidation of other living tissue can occur

A NEED FOR CHANGE

Isopropyl alcohol is commonly used as a disinfectant for several propagation procedures (most commonly sterilization of work surfaces and tools) but is not a reliable bactericide.

Benzalkonium chlorides have been used in similar ways as both fungicides and bactericides for over 50 years, but have some phytotoxic effects on certain plants (Dirr and Heuser, 1987; Kyte, 1987; MacDonald, 1986; Pierik, 1987; Torres, 1989). Many commercial formulations are available. The naturally occurring chloride ion (Cl, part of what makes up sea water) is one of the oxidation by-products of benzalkonium chloride disinfectants. These by-products do not pose serious environmental hazards when used at recommended rates.

Bleachs (hypochlorite solutions) are inexpensive and effective. Research does indicate that in some uses, however, hypochlorite cannot be completely washed off of plant material. It leaves enough residue to interfere with plant metabolism (Abdul-Baki, 1974a, 1974b). It is also a very uniquely harmful disinfectant. The byproduct of bleach oxidation is the hypochlorite ion (OCl). It is the hypochlorite ion (not the chloride ion) that is harmful when it makes its way into our natural water systems. The hypochlorite ion attaches to organic compounds in the soil and forms very stable chlorinated organic compounds. These compounds are then taken up through roots and ingested by humans, aquatic species, and animals. Neither the hypochlorite ion nor its organic compounds occur in nature—plants and animals, therefore, have no enzymes to metabolize or detoxify them. These chlorinated organic compounds bioaccumulate in body fat (Thornton, 1991). Recent studies in the Great Lakes region of the U.S. indicate that some health epidemics can be attributed in part to organochlorine compounds. Increases in human breast and prostate cancers as well as other health problems in birds, fish, and other aquatic wildlife have been connected to the high levels of organochlorines in tissue (DeCrosta, 1978; Hileman, 1993; Swanson, 1994; Thornton, 1991, 1992).

A promising disinfectant for propagation and other industries is hydrogen peroxide ($\rm H_2O_2$). It can be used as a sterilant for both fungi and bacteria. It has absolutely no toxic by-products (it breaks down to water and oxygen) and it has no residual effects in water or soil. It can be used as a soil, water, and plant disinfectant and is even used as a seed coating to enhance germination. Hydrogen peroxide is also used in cleaning up water polluted with toxic chemicals (Environmental Services Company, Carus Chemical Co., Ottawa).

Hydrogen peroxide can be purchased in bulk at the 35% concentration rate. It can be used to control fungi and bacteria. A recommended concentration for surface disinfestation of plant material is 1 part $\rm H_2O_2$ (35%) to 100 parts water (Environmental Services Company, 1993). It is twice as expensive as bleach, but is currently being considered as an effective alternative for industrial chlorine bleaching used in paper and other manufacturing processes. It may soon be more economical and more available (Kroesa, 1990).

THE FUTURE

The extensive use of elemental chlorine in other industries (e.g., pulp and paper manufacturing, manufacturing and use of organochlorine pesticides, water treatment, metallurgical processes, and plastics manufacturing) pose much greater risks to animals, humans and the environment than our slight uses in propagation. Yet, even something as common as everyday bleach has hidden dangers. We do have a responsibility to become informed about the chemicals we use. In doing so, we can promote safety and health in our work place and our environment.

The important thing is for all industries to start looking for alternatives. By reducing the amount of organochlorine-causing chemicals we use, we begin to reduce the amount found in the environment and reduce the potential health hazards related to them. Other industries have begun exploring alternative methods of purifying, disinfecting, and bleaching products. They include innovative processes of ozonation (to replace chlorination) of drinking water, and using hydrogen peroxide to bleach paper and clothing (Kroesa, 1990; Richardson 1993). From Canada to Switzerland to Sweden, other countries are taking more serious steps in phasing out the use of organochlorines (Hileman, 1993; Palter, 1989; Vallentyne, 1992).

A complete list of commercially available chemicals, and their chemical formulas, by-products, and toxicity to humans and the environment can be acquired by your cooperative extension agent through a computer toxicology network (EXTONET) and from an organization called NCAMP (National Coalition Against the Misuse of Pesticides) that publishes *Pesticide Reviews* .

Being informed makes sense. It can be a bonus for our business and our public relations, our production practices and our plants, our employees and our environment.

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LITERATURE CITED

Abdul-Baki, A.A. 1974a. Hypochlorite and tissue sterilization. Planta 115:373-376.

Abdul-Baki, A.A. 1974b. Pitfalls in using sodium hypochlorite as a seed disinfectant in 14C incorporation studies. Plant Physiol. 53:768-771.

DeCrosta, A. December 1978. Learn to live without chlorine. Organic Gardening p. 77-92.

Dirr, M. and Heuser Jr., C.W. 1987. The reference manual of woody plant propagation. Varsity Press, Athens, Georgia.

Hileman, B. 1993. Concerns broaden over chlorine and chlorinated hydrocarbons. Chemical and Engineering News April 19, p.11-20.

Kroesa, R. 1990. The Greenpeace guide to paper. A Greenpeace Publication, Amsterdam.

Kyte, L. 1987. Plants from test tubes, revised edition. Timber Press, Beaverton, Oregon.

MacDonald, B. 1986 Practical woody plant propagation for nursery growers. Vol. 1, Timber Press, Portland, Oregon.

Palter, J. 1992. Chlorine bans and phase-outs: Reversing the mistake of chlorine chemistry. Chlorine Free. Vol.1, No. 2.

Pierik, R.L.M. 1987. In vitro culture of higher plants. Martinus Nijhoff Publ, Dordrecht.

Richardson, B. 1993. Why Clinton should take chlorine out of paper 'pool'. Christian Science Monitor, October 10. p. 19.

Swanson, S. 1994. Chlorine: Its time may be up. Chicago Tribune, February 27.

Thornton, J. 1991. The product is the poison: the case for a chlorine phase-out, Greenpeace Great Lakes Project. Chicago, IL.

Thornton, J. 1992. Breast cancer and the environment: The chlorine connection, A Greenpeace Report, Great Lakes Office, Chicago, IL.

Torres, K.C. 1989. Tissue culture techniques for horticultural crops, VanNostRand Reinhold Publ., NY.

Vallentyne, J. 1989. The case for phasing out organohalogens. In: J. Palter, (ed.). A Greenpeace Report. Toronto, Ontario Canada.