

hastening of the terminal growth after the flower buds were formed.⁷

The initiation of flowering by these substances does not necessarily imply that they are "florigens"⁸ since they have other effects on plant growth. Furthermore, acetylene and ethylene, compounds chemically

quite unrelated to these phytohormones, also induce premature flowering in *Ananas*.^{9,10}

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

SELF-STERILIZING SURFACES

It is known that extremely small quantities of ionized silver can have a remarkable germicidal effect;¹ it has also been found that one must distinguish between effects produced by small silver ion concentrations in water (volume effect) and those occurring when microorganisms are brought into wetting contact with surfaces upon which Ag is absorbed (surface effect). In the former case the destruction of the organisms is mostly a matter of hours and a variation of the Ag concentration up to 2 parts per million produces no remarkable change in the rate of sterilization. Furthermore, it is difficult, if not impossible, to attack spores of bacteria, molds and yeasts. This behavior is quite at variance with the very rapid destruction of cells which come into close contact with the extreme Ag concentrations existing on surfaces with absorptive capacity for Ag. Here the destruction of large numbers of organisms is reduced to minutes.

This phenomenon suggests a practical application in the form of self-sterilizing surfaces with lasting activity, if the incorporation of an adequate supply of atomic silver which replenishes the surface continuously after a wetting contact can be realized. As many organic colloids, in particular the proteins, bind and thus remove the silver from the surface after contact with it, a disactivation results unless a process of replacement can be provided for.²

The conditions for the permanency of the self-sterilizing qualities is thus the use of a material which (a) exposes at the surface only a small fraction of its total silver content, (b) holds this fraction in a form almost insoluble in water but available to proteins, (c) protects the unexposed supply against chemical attack, (d) permits replacement by diffusion.

⁷ A. E. Hitchcock and P. W. Zimmerman, *Contrib. Boyce Thompson Inst.*, 7: 447-476, 1935.

⁸ M. Kh. Cajlachjan, *Compt. Rend. Acad. Sci., U. S. S. E. (N.S.)*, 4: 79-83, 1936.

¹ Other metals, like gold and copper, share this property with silver at least to some extent. The reason for the particular focus on silver is due to its lack of toxicity compared with copper and its economic advantage over gold. It appears certain that similar materials can be developed with the incorporation of, e.g., copper.

² The use of metallic silver surfaces may appear obvious because of the infinite supply of atomic silver. It is, however, easily demonstrated that metallic silver, even if very clean, is soon disactivated due to the formation of germicidally inert compounds.

Numerous organic liquids could fulfil these conditions, but most practical applications of such surfaces require rigidity. The only rigid substances with adequate properties are vitreous materials of anorganic (glasses) or organic (plastics) nature. The diffusion rate appears to be too small for the former (unless in colloidal form), and even of the plastics only certain types have so far been found to provide for sufficiently fast exchanges.

The compounding of silver with the resins can occur in various ways, either by the dissolution or the colloidal dispersion of silver compounds into the monomers or half-polymers of a resin or by their dissolution or dispersion in solvents of the plastic.

The additional incorporation of stabilizing as well as plasticizing substances is important, also intransparent neutral filling materials are required where a protection of the interior of the material against photochemical effects on the silver content is needed.

The resulting compounded substance represents then a varnish-like viscous fluid which can be applied by brush, spray or impregnation to various bases like plastics, glass, wood, paper, cloth, etc. It is hardened *in situ* either by polymerization or evaporation of the solvents. These surfaces are tasteless and odorless, resist mechanical wear and chemical attack by weak acids and alkali solutions as well as boiling water. They are, however, sensitive to certain organic solvents. The amount of silver removed from the surface by, e.g., touch with the lips is of the order of micrograms, i.e., negligible from the toxic angle. The total quantity of silver which the surface material must contain varies widely with the intended use of the surface and with its intended degree of permanency, it amounts to approximately one gram of silver for 1,000 cm² of exposed surface for the heaviest type of duty so far developed.

By the choice of the proper resin, its degree of polymerization, quantity and type of filling materials, etc., it is possible to vary the rate of Ag replacement, the absorptive capacity for water as well as the hygroscopic qualities of the surface. Hence surfaces which will be wet most of the time and come in frequent touch with large quantities of protein-like substances must have a high replacement rate

⁹ A. G. Rodriguez, *Jour. Dept. Agric. Porto Rico*, 16: 5-18, 1932.

¹⁰ K. R. Kerns, "U. S. Patent No. 2,047,874," 1936.

but a low water permeability in order to prevent a premature exhaustion of the incorporated silver supply, while surfaces mostly dry require certain hygroscopic properties and an appreciable water permeability. Consequently the performance for which a particular surface material is designed represents by necessity a compromise between the rate of sterilization per unit area, the rate of replacement and the total "life time" required for the surface.

The method used for testing the germicidal activity of these surfaces was the following: Samples of the surface material (about 6 cm²) on various bases were placed in humidified containers (for preventing bacterial destruction by drying). The test microorganisms suspended in the desired medium were pipetted onto the surface in volumes of 0.05–0.1 cc, spreading the liquid into a film. Analogously the controls were obtained on neutral surfaces. At definite time intervals this film or part of it was removed by a sterile cotton swab, and was immediately introduced into 9 cc of lactose-beef or thio-glycollate broth. After incubation at 37° C. for 1 to 5 days the growth was determined. A similar technique was applied for the quantitative determination of cell reduction by titration: the entire film was absorbed by the swab, the latter then soaked for 30 minutes in nutrient broth with frequent shaking before 1 cc was serially diluted and plated in nutrient agar. Colony counts were made after three to five days.

The test microorganisms used so far have been *E. coli*, *Staph. aureus*, *B. proteus*, *B. subtilis* Cl. *pasteurianum*, *Penicillium*, *Rhizopus* and *Sacch. cerevisiae*.

Distilled tap and peptone water, nutrient broth, 5 per cent. sucrose and dextrose solutions, cider and milk have been used as suspending media.

The germicidal action obtainable with various surface materials according to extended tests with the above methods are briefly this:

The rate of sterilization varies with the composition of the surface, the highest rate measured sterilizes *E. coli* at 10⁸ cells/cc in less than one minute. Materials requiring rates of more than 5 minutes for *E. coli* at at least 10⁵ cells/cc were discarded. The bacterial concentration does not influence in general the rate of sterilization.

For a given surface this rate does not vary appreciably with different types of organisms (except for spores). Mold suspensions containing high concentrations of spores were readily sterilized in all suspending media except nutrient broth and milk. This was demonstrated by exposing heavy mold suspensions in cider and sugar-peptone solutions for 1 to 5 minutes to surfaces applied within standard bottle

caps, before applying them to 12-ounce bottles containing sterile cider or broth. After sealing, the nutrient was kept in permanent contact with the cap. Subsequent incubation (30 to 60 days) did not produce growth in any bottle, while control bottles with untreated caps showed heavy growth. For bacterial spores (*B. subtilis*, 10 days old, washed and heated to 100° C. for 5 minutes) reduction up to 97 per cent. has been obtained after 15 to 30 minutes exposure.

In general the rate of disinfection depends upon the concentration of protein-like matter in the suspending medium. In this respect milk is most severe, and a surface which destroys *E. coli* in water in about 2 minutes requires 15 to 30 minutes for the sterilization of non-sporulating bacteria in milk.

Endurance tests for various surface materials were made on a special testing machine, which dipped each sample every fifth minute for about 30 seconds into H₂O. At arbitrary intervals the above test was performed and it was found that the activity remained practically unimpaired for up to 30,000 infections over a period of 2 months. The final failure coincided in general with the destruction of the plastic surface by mechanical wear.

Without such treatment good surfaces have not shown disactivation during storage. *

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