Germicidal Effect of Ultraviolet Irradiation on Paper Contaminated with Mycobacteria

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An ultraviolet chamber for decontaminating single sheets of paper seeded with mycobacteria was investigated. The ultraviolet killing effect ranged from 50% for Mycobacterium kansasii to more than 99% for M. tuberculosis.

Contaminated laboratory data sheets are a potential source of infection for laboratory personnel. An effective, simple, and rapid method for disinfecting such contaminated material is, therefore, desirable. Phillips and Novak (1) described an ultraviolet (UV) pass-through chamber for decontaminating single sheets of paper. The chamber was shown to reduce microbial populations of Bacillus subtilis spores, Serratia marcescens, Escherichia coli B/r, or colophage T-3 by 99.9 to 100% after irradiation.

The work of Riley and associates (2-4) attests to the effectiveness of UV irradiation on airborne tubercle bacilli; however, little information is available concerning the sensitivity to UV irradiation of mycobacteria adsorbed to paper. This study reports our experience with the paper pass-through chamber in disinfecting paper contaminated with species of mycobacteria capable of causing disease in man.

One strain each of Mycobacterium tuberculosis, M. intracellulare, M. fortuitum, M. scrofulaceum, and M. kansasii was grown in Middlebrook 7H-9 broth (Difco) for 1 week at 35 C under 6 to 10% carbon dioxide. Duplicate strips of white bond paper (10 by 15 mm), previously sterilized by autoclaving, were inoculated with 0.01 ml of either undiluted or 1,000-fold diluted broth cultures of the test organisms. The strips were air-dried and passed through the UV chamber. Paper strips, protected by aluminum foil, were passed through the chamber as specific organism controls. As they passed through the apparatus, the strips were subjected for 10 sec on each side to two 15-w UV lamps, each registering a light intensity of 23 µw/cm² at one meter. Uninoculated paper strips served as sterility checks on contamination from the external environment.

After exposure to the UV light source, the test and control paper strips were aseptically placed in screw-cap tubes containing 7 ml of 7H-9 broth. The contents of the tubes were mixed on a Vortex Junior mixer to facilitate elution of the organisms from the paper. Numbers of viable organisms removed from the paper by this mixing were determined by the immediate dilution-plating technique. Triplicate 0.04-ml samples of each dilution were inoculated to Dubos Middlebrook oleic acid-albumin agar plates supplemented with OADC enrichment (Difco). Plates were spread to dryness and incubated under 6 to 10% CO₂ at 35 C. After 30 days of incubation, colonies were enumerated with the aid of a lighted colony counter. Results of the viable plate counts are shown in Table 1.

From these data it is evident that the UV pass-through chamber is effective in reducing the number of viable mycobacteria on contaminated paper. The UV killing effect varied with the species tested and was inoculum-dependent. M. tuberculosis appeared to be the most UV-sensitive of the species tested. The UV sensitivity of M. intracellulare was similar to that of M. tuberculosis. M. scrofulaceum and M. fortuitum were markedly reduced in number by UV irradiation but were less affected than M. intracellulare and M. tuberculosis. M. kansasii was the least affected by germicidal UV, showing only about a 50% reduction in colony-forming units after irradiation. The less marked effect of UV on M. scrofulaceum, M. fortuitum, and M. kansasii suggests that a more detailed investigation of photoreactivation and dark reactivation should be undertaken.

The UV chamber did not provide complete sterilization of paper heavily contaminated with any of the species tested; however, this method of disinfection would probably be effective against
# Table 1. Bactericidal effect of an ultraviolet pass-through chamber on species of mycobacteria

| Test organism           | Initial inoculum per strip | Avg no. of organisms recovered per strip | Inactivation of test organism |
|-------------------------|----------------------------|-----------------------------------------|-------------------------------|
|                         |                            | Control       | Irradiated        | %                          |
| *M. tuberculosis*       | $2.2 \times 10^3$          | $5.3 \times 10^3$ | $28$              | 99.5                       |
|                         | $2.2^a$                    | $56$          | $0$               |                            |
| *M. kansasii*           | $4.0 \times 10^3$          | $6.3 \times 10^4$ | $2.8 \times 10^3$ | 55.5                       |
|                         | $0.4^a$                    | $56$          | $28$              |                            |
| *M. intracellulare*     | $2.4 \times 10^5$          | $3.0 \times 10^4$ | $2.1 \times 10^4$ | 99.3                       |
|                         | $2.4 \times 10^6$          | $1.4 \times 10^4$ | $2.5 \times 10^3$ | 98.2                       |
| *M. scrofulaceum*       | $4.8 \times 10^5$          | $3.5 \times 10^4$ | $1.0 \times 10^5$ | 97.1                       |
|                         | $4.8 \times 10^6$          | $9.1 \times 10^3$ | $1.7 \times 10^4$ | 81.3                       |
| *M. fortuitum*          | $8.9 \times 10^5$          | $5.6 \times 10^4$ | $1.6 \times 10^4$ | 97.1                       |
|                         | $8.9 \times 10^6$          | $2.1 \times 10^4$ | $1.3 \times 10^4$ | 93.8                       |

* Colony counts for these 1,000-fold dilutions were calculated.

* Not determinable.

The small numbers of mycobacteria with which paper might inadvertently be contaminated under normal laboratory working conditions.

**Literature Cited**

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