Short Communication

DETECTION OF POTATO VIRUSES Y AND S IN TUBERS
BY ELISA AFTER BREAKING OF DORMANCY WITH
BROMOETHANE OR RINDITE

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Introduction

In the development of enzyme-linked immunosorbent assay (ELISA) for post-harvest detection of virus in seed tubers, artificial breaking of dormancy with rindite to increase the virus concentration in the tuber, has been found to be essential for the reliable detection of such viruses as potato Y (PVY) (5, 7), A (PVA), (7), and leaf roll (PLRV) (4). Reduction in tuber dormancy, however, may be achieved through a variety of treatments (3). One treatment recently reported (3) is the use of bromoethane which is potentially useful due to its lower mammalian toxicity but whether such alternate methods also stimulate virus synthesis is unknown. The present study was therefore undertaken to determine the potential of this alternative means of dormancy breaking with bromoethane as a means of stimulating virus synthesis for post-harvest virus detection.

In addition, it has been suggested that the very low concentrations of PVY and PVA in dormant tubers and their drastic multiplication after breaking dormancy with rindite is a characteristic of potyviruses and is unique to this group among the potato viruses (7). Therefore, the response of potato virus S (PVS) (a member of the carlavirus group) to the breaking of dormancy with rindite was examined in this study.

Materials and Methods

Tubers of the Russet Burbank cultivar, ca. 95% infected with PVY and 100% infected with PVS (on the basis of growing a sample of 100 tubers in the greenhouse and testing the foliage by ELISA), were harvested October 7, 1983, divided into three lots of 300 tubers, and held at room temperature. On October 26, one lot of 300 tubers was treated with rindite (a mixture of ethylene chlorohydrin, dichloroethane, and carbon tetrachloride, 7:3:1 by volume) in a commercial fumigation chamber, a second lot of 300 tubers was

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treated with bromoethane 0.2 ml/L in a plastic container, and the third lot of 300 tubers was left untreated. After aeration, the three lots were stored at 18°C in the dark.

Virus testing was commenced 27 days after treatment and was repeated on days 34, 46, 54, 67, 74, and 81. The general ELISA methods of Clark and Adams (2) were used as previously modified (6). Coating globulin and globulin conjugated to alkaline phosphatase were prepared from locally produced antisera to PVY and PVS. On each testing date, 30 tubers from each lot were processed by taking slices of tuber tissue from the rose end of each tuber. The epidermal slice was discarded and sap was extracted from subsequent slices with a mechanical sap extractor. Six drops of sap from each tuber were diluted in sample buffer to a final volume of ca. 1 ml. Three untreated healthy tuber controls were used in each plate. After adding enzyme, the microtitre plates were kept at room temperature for 1 h, and the absorption at 405 nm was recorded with a Multiskan colorimeter.

The breaking of dormancy after tuber treatment was monitored by taking 10 tubers at random from each of the lots on days 45, 52, 61, and 67. Measurements were made on the number of active eyes and the length of the longest sprout on each tuber.

Data were analyzed by Duncan’s multiple-range test.

**Results and Discussion**

Artificial breaking of dormancy was achieved with both the rindite and bromoethane treatments (Fig. 1), and differences in number of active eyes and rate of sprout elongation were not statistically significant. As had been reported by others (5, 7), treatment of tubers with rindite significantly in-

![Graph A](image1)

**FIG. 1.** Breaking of tuber dormancy after treatment with rindite (■), bromoethane (●), and untreated (▲); A, the mean number of active eyes; and B, mean length of longest sprout.