Some Effects of 2,4,5-Trichlorophenoxyacetic Acid on the Mitotic Cycle of Lateral Root Apical Meristems of *Vicia faba*

R. D. MacLeod

Department of Biology, University of Missouri, St. Louis Missouri

Received April 21, 1969 / Accepted April 28, 1969

Abstract. Roots of *Vicia faba* were given a one hour pulse label with $^3$H-TdR (1 $\mu$C/ml), either before or after a three hour treatment with a $10^{-5}$ M solution of 2,4,5-trichlorophenoxyacetic acid (TCPA). The durations of the various phases of the mitotic cycle were derived from labeled prophase curves, prepared from autoradiographs of lateral root apical meristems. — TCPA was found to lengthen the duration of the mitotic cycle, primarily because it extended the duration of the period of DNA synthesis (S), though post-synthetic interphase (G2) was also longer. No measurements could be made with respect to the duration of presynthetic interphase (G1), because of rapid changes in the lengths of the G1 and S periods following treatment. — As well as extending the duration of S, TCPA treatment also resulted in at least an initial increase in the rate of DNA synthesis and a decrease in the actual number of cells in S. These results have been discussed with respect to the control of the organization of the root apical meristem.

Introduction

Extensive investigations carried out in the past few years have revealed the complex nature of the apical meristem of many Angiosperm roots. Structurally, this meristem consists of a group of quiescent cells surrounded by proliferating cells (Clowes, 1961), which are heterogeneous with respect to cell cycle time (Clowes and Hall, 1963; Webster and Davidson, 1968). Dividing cells with different cycle times are not distributed at random throughout the root apical meristem however, as the duration of the mitotic cycle of any cell, or group of cells, seems to be a property of the position of that cell in the root (Clowes and Hall, 1963).

Variation in cycle time among the cells of the root apical meristem is paralleled, not unexpectedly, by physiological differences between these cells. It is known, for example, that following X-ray treatment, division of the proliferating cells is inhibited, but the cells of the quiescent center come into division (Clowes, 1959). Similar results have been found

* Supported by a grant from the Assistant Professor Research Fund of the University of Missouri.
following cold treatment (Clowes and Stewart, 1967). If roots are treated with colchicine, the meristematic cells stop dividing (MacLeod and Davidson, 1966) and a new meristem is formed at the root apex from both cells of the quiescent center and former proliferating cells (Davidson, 1961).

Control of the organization of this physiologically heterogeneous mass of cells, which makes up the root apical meristem, must be very exact, and it is not surprising that it is very sensitive to disruption by various treatments. There is some evidence which suggests that control of the organization of the root apex is mediated by one or more growth factors. X-irradiation, for example, is known to upset the biosynthesis of indole acetic acid (IAA) (Gordon, 1956) and colchicine treatment appears to lower the endogenous level of an auxin in the root apex (Davidson and MacLeod, 1966, 1968; Webster, 1967). It has been proposed, in fact, that IAA synthesis takes place in the root apical meristem (Davidson, 1960; Torrey, 1963) and that one of the events in the recovery of roots from irradiation (Davidson, 1961) or colchicine treatment (Davidson, MacLeod and Taylor, 1965) is the re-establishment of the normal endogenous auxin level.

The effects of two growth hormones, namely kinetin and the auxin 2, 4, 5-trichlorophenoxyacetic acid (TCPA), on the duration of the mitotic cycle and its various phases have been determined in apical meristems of lateral roots of *Vicia faba*. The results obtained following kinetin treatment have been reported (MacLeod, 1968); those found after exposure to TCPA will be presented in this paper.

**Materials and Methods**

Beans (*Vicia faba*) were grown in the usual way (Davidson and MacLeod, 1966). They were divided into three groups; one batch represented the controls; the second group were treated with kinetin (see MacLeod, 1968;) and the remaining beans were treated with TCPA. All of the beans were exposed to $^3$H-Thymidine ($^3$H-TdR) for one hour (1 μC/ml; Specific activity 6.7 C/mM). One group of beans was treated with a $10^{-5}$ M solution of TCPA for three hours before being exposed to $^3$H-TdR, while a second batch was treated in the same way following exposure to $^3$H-TdR. Following treatment, the beans were washed and grown on at 20°C.

Five lateral roots were fixed immediately after exposure to $^3$H-TdR in both treated and control roots, and at intervals thereafter for 27 hours. Autoradiographs were made of Feulgen stained squash preparation.

Measurements: The following factors were determined:

1. Mitotic Index (MI), the percentage frequency of cells in mitosis:
2. Labeling Index (LI), the percentage frequency of labeled cells.
3. The durations of the mitotic cycle (C) and its various phases, which are $G_1$, or pre-synthetic interphase, S, or the period of deoxyribonucleic acid (DNA) synthesis, $G_2$, or post-synthetic interphase, and mitosis. Details of the techniques used here may be found in the literature (Quastler and Sherman, 1959; van't Hof, 1965).