# STUDIES ON THE SITE OF ACTION OF SAN 9785 [4-CHLORO-5-DIMETHYLAMINO-2-PHENYL-3(2H) PYRIDAZINONE] ON CELL DIVISION USING ONION ROOT TIPS

## KARUNYAL SAMUEL and SALIL BOSE

School of Biological Sciences, Madurai Kamaraj University, Madurai 625 021, India.

## **ABSTRACT**

The effect of SAN 9785 [4-chloro-5-dimethylamino-2-phenyl-3-(2H) pyridazinone] on mitotic division was examined using onion root tips. When onion root bulbs were placed in the presence of 0.5 and 1 mM SAN 9785, there was a significant decrease in the number and length of roots as compared to the control. Mitotic studies using onion root tip squashes showed that the cell division was inhibited at the prophase stage by the herbicide (SAN 9785) treatment.

#### INTRODUCTION

Pyridazinone herbicides are widely used in studying the structure and function of photosynthetic apparatus. The effects of substituted pyridazinones on plants have been reviewed<sup>1,2</sup>. SAN 9785 is one of the pyridazinones which affects the fatty acid content maximally without much affecting the pigment content<sup>3</sup>. Recently it has been shown that the growth of algae and higher plants was affected by pyridazinone herbicides<sup>4,5</sup>. To check whether the growth of the non-photosynthetic system is also affected by SAN 9785 and to study the site of action of the herbicide, onion roots were used for the present study.

#### MATERIALS AND METHODS

Smaller variety of the onion bulbs was used for mitotic studies. The outer dry scales were peeled off and the bases scrapped to expose the eyes. The bulbs were placed in injection vials containing various concentrations of SAN 9785 (0.5, 1 mM), touching the surface of the solution. After 3 days, the roots were removed from the base with a sharp blade. The number and the length of the roots were measured. The roots from the bulbs per treatment were excised 2 mm above the tip and fixed with acetic acid: ethanol (1:3) for a day. The root tips were washed with water, hydrolysed in 6 NHCl for 5 min rinsed with water and stained with Feulgen stain<sup>6</sup>. Squashes were made from 20 different bulbs per treatment. The number of cells was counted at four foci per slide and the sum total per slide ranged from 60 to 300 cells. The cells showing different phases were classified using a compound microscope. The percentage of cells in different phases

was calculated for each slide with respect to the total number of cells for each treatment. Mean and SD values were calculated for different phases of control and treated sample.

### RESULTS

Table 1 shows the number and length of roots, formed from onion bulbs, which were placed on water with and without SAN 9785. Compared to the number of roots, the growth of roots as measured by length was affected more by the herbicide treatment (table 1). There was a 12–18% decrease in the number of roots at 0.5 and 1 mM SAN 9785 as compared to control. The decrease in the length of roots at 0.5 and 1 mM SAN 9785 was 44% and 70% respectively (table 1).

Mitosis in onion root tips was inhibited by SAN 9785 treatment as seen from tables 2 and 3. In the control, compared to the total number of cells, the cells in the prophase, metaphase, anaphase and telophase were 9, 8.5, 4 and 2% respectively. The percentage of cells with respect to the total number of cells in the different phases of the control was

Table 1 Effect of SAN 9785 on emergence and growth of roots in onion root tips

SAN 9785 (mM)	No. of roots	Length of roots (mm)		
Control	156 (100%)	6.9 + 5.7 (100%)		
		(n=101)		
0.5	128 (82%)	$4.4 \pm 3.0 (64\%)$		
		(n = 92)		
1.0	138 (88.5%)	$2.6 \pm 1.5 (37\%)$		
	'	(n=114)		

<del></del>					0.5 mM 1.0 mM						<del></del>	
		Coi	atrol			0.5	mM			1.0	mM	
	Pro- phase	Meta- phase	Ana- phase	Telo- phase		Meta- phase		Telo- phase		Meta- phase		_
Highest value	52.5	19.1	14.7	1.3	27.6	16.3	8.0	0	11.4	5.3	3.3	0
Mean Standard	10.9	8.5	3.6	0.2	10.1	7.5	3.5	0	3.7	1.0	0.3	0
deviation	10.8	4.4	3.5	0.4	8.0	4.2	1.8	0	3.6	1.7	0.7	0

Table 2 Effect of SAN 9785 on mitosis of onion root tips\*

similar to that at 0.5 mM. However, in the root tips treated with 1 mM SAN 9785, the number of cells in the prophase was 66% lesser than that of the control. The decrease in the number of cells in the metaphase and anaphase was 88% and 91% compared to their respective controls.

#### DISCUSSION

The herbicide (SAN 9785) inhibited the growth of roots (table 1). Onion root tips offer a good classical system to study mitosis. The results (tables 2 and 3) indicate that the inhibition is at the prophase. The chromosome condensation and subsequent transport to the central plane might have been affected by SAN 9785 treatment. Similar results were observed in soybean roots treated with a different herbicide (Chloropropham)<sup>7</sup>. Certain fungicides and toxins from bacteria are known to inhibit mitosis<sup>8,9</sup>. The effects of a number of herbicides such as N-phenylcarbamates, MH, dinitroanilines, phenoxyalkane, carboxylic acid, pronamide, diallate and CDAA on cell division and mitotic activity have

been summarized by Linck<sup>10</sup>. The present results (tables 2 and 3) provide an example of a pyridazinone herbicide (SAN 9785) which acts as an inhibitor of the mitotic division at the prophase. This inhibition could be at a critical stage i.e. when the chromosome duplicates. It is also known that the uncoupling of oxidative phosphorylation causes aberrations<sup>11</sup>. Attempts are being made in this line to examine these possibilities.

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**Table 3** Statistical analysis of the effect of SAN 9785 on the number of cells at various stages during mitosis in onion root tips

SAN 9875 (mM)	Prophase	Metaphase	Anaphase	Telophase	
Control	10.9 + 10.8	8.57 + 4.4	3.61 + 3.5	0.2 + 0.4	
0.5	10.1 + 8.0	7.53 + 4.2	3.57 + 1.8	0	
1.0	3.7 + 3.6	1.03 + 1.7	0.31 + 0.7	0	
<del></del> -		T values	······································		
Control vs 0.5 mM	2.48*	1.47**	0.03**		
Control vs 1.0 mM	18.82*	8.31*	2.78*		
0.5 mM vs 1.0 mM	3.6*	6.89*	1.47**		

<sup>\*</sup> Significant; \*\* Not significant.

<sup>\*</sup> Condensed form of the percentage of cell in different phases of mitosis from 20 squashes of control and SAN 9785-treated onion root tips. The least value in all the cases was zero.

<sup>1.</sup> Eder, F. A., Z. Naturforsch., 1979, C34, 1052.

<sup>2.</sup> Rittig, F. R., St. John, J. B., Becker, R., Bleiholder, H., Feichtmayr, F. and Haedicke,

- E., Pestic. Sci., 1983, 14, 299.
- 3. St. John, J. B., Plant Physiol., 1976, 57, 38.
- 4. Karapetyan, N. V., Strasser, R. and Boger, P., Z. Naturforsch., 1983, C38, 556.
- 5. Mannan, R. M., Ph.D. thesis, Madurai Kamaraj University, Madurai, India, 1983.
- 6. Jensen, W. A., In: Botanical histochemistry: principles and practice, W. H. Freeman, San Francisco, 1962, p. 408.
- 7. Davis, D. G., Hoerauf, R. A., Dusbabek, K. E. and Dougall, D. K., Physiol. Plant., 1977, 40,

- 15.
- 8. Sahu, R. K., Behera, B. N. and Sharma, C. B. S. R., The Nucleus, 1981, 24, 60.
- 9. Sharma, C. B. S. R., Sahu, R. K. and Panigrahi, S., Caryologia, 1978, 31, 89.
- 10. Linck, A. J., In: Herbicides: physiology, biochemistry, ecology, (ed.) L. J. Audus, Academic Press, London, 1976, Vol. 1, p. 83.
- 11. Kihlman, B. A., In: Actions of chemicals on dividing cells, Prentice-Hall, New Jersey, 1966, p. 260.

# **NEWS**

### THE JACK-OF-ALL-TRADES OFFICE COMPUTER

A workstation management system devised by a UK company enables a single computer to be used as a stand-alone computer, applications processor, networked file-server and front-end processor linking users with a mainframe computer.

Centred on the concept of open systems interconnection (OSI) and the Unix operating system, the WMS/X from ROCC Computers of Crawley in south-east England embodies Motorola 68000 and RISC (reduced instruction set computer) chip technology. It is claimed to be one of the fastest Unix systems available for eight to 32 users, and has integrated software for most general office purposes.

One of a series of three minicomputer product ranges called the TRIPOS workstation management systems, WMS/X is specially designed to support information processing by handling data, text and graphics. It provides an overall system interface manager for Unix, relational databases and the C-Check II fourth-generation programming language developed by ROCC. Offering full networking capability; it is compatible with Ethernet and

X25 standards and functions in conjunction with the ROCC 28X8 range of computers.

Other TRIPOS software operating systems include the basic WMS which supports various types of workstation terminal, including synchronous, asynchronous and videotex units. This allows real-time data processing with up to 96 concurrent users, in conjunction with the ROCC 28X5 computer range. The WMS/V system is designed to support up to 1000 videotex terminals in typical usage.

ROCC Computers, formed in 1984, has its origins in the Redifon group of British companies and later in Rediffusion Computers Ltd. Claimed to be the only fully integrated supplier of complete videotex systems (both hardware and software), it announced sales of £16 million in the year ending 31 March 1988.

There are more than 1800 ROCC computers installed worldwide. (ROCC Computers Ltd, Kelvin Way, Crawley, West Sussex, RH10 2LY, England; British Information Services, British High Commission, Chanakyapuri, New Delhi 110 021).