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A COMPARATIVE STUDY OF SUPEROXIDE DISMUTASE ACTIVITY OF DIFFERENT YAM SPECIES

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ABSTRACT

The activity of superoxide dismutase (SOD) in three species of yam: *Dioscorea cayenensis* poir cv okpai, *Dioscorea alata* poir cv Chembeli and *Dioscorea alata* poir cv Kurudu was studied. The study demonstrated a significant variation of superoxide dismutase activity in the three varieties of Dioscorea species. The highest activity was found in *Dioscorea cayenensis* and the lowest one in *Dioscorea alata* poir cv Kurudu. The order of activity of the enzyme in the tuber was head > middle > tail physiological region. Inhibition studies with cyanide showed that the enzyme in the yam cell of all the varieties was inhibited by cyanide. These observations indicated that identical SODs are present in the cytosol and mitochondria of the yam cells of the different varieties studied. Thus different yam species vcxz\have different SOD activities that confer peculiar protective role against superoxide '' anion.

Keywords: *Dioscorea alata, Dioscorea cayenensis,* superoxide dismutase.

INTRODUCTION

Oxygen is important in respiration, and plays crucial roles in various metabolic activities occurring in living cells. Moleculer oxygen can be innocuous to aerobic cells when it undergoes successive univalent reductions to produce highly reactive free radical called superoxide anion (Hassan, 1984). This highly reactive species can accumulate, producing deleterious and destructive effect to cells. These effects include lipid peroxidation of polyunsaturated fatty acids in all membranes, which results in oxidative denaturation of proteins and nucleic acids (Robbins and Cotran, 1994).

Nature, however has devised an elegant system of defence against the effect of superoxide anion. This includes the presence of antioxidant enzymes, which are superoxide dismutase (SOD), catalase and glutathione peroxidase (Harris, 1992). SOD converts superoxide anion to oxygen and hydrogen peroxide (Fridovich, 1995) while catalase converts hydrogen peroxide to oxygen and Water (Voet and Voet, 1990).

The involvement of free radicals in the spoilage of stored *Dioscorea rotundata* and protection by superoxide dismutase has been established (Isamah et al, 2000). The present work deals with a comparative study of SOD activity in protecting different varieties of stored yam from deteriorative effect of superoxide anion.

MATERIALS AND METHOD

MATERIALS

The tubers of *Dioscorea alata* Poir cv Kurudu, *Dioscorea alata* Poir cv chembeli and *Dioscorea Cayenensis* Poir cv Okpai were obtained from a local farm in Obiaruku, Delta State of Nigeria. The tubers selected for this study were in storage for eleven months and were physically intact and free from any disease. All reagents used were of analytical grade.

METHOD

Preparation of extract and the determination of superoxide dismutase (SOD) activity.

The yam tuber pieces (50g) from each physiological region(head, middle and tail) of *Dioscorea cayenensis* were separated, washed with ice-cold water and homogenized with 100cm of ice-cold 0.05M phosphate buffer pH 7.0 containing 1%(w/v) Triton X-100 (to disrupt the mitochondrial membrane) using Potter-Elvehjem homogenizer. The extract obtained was clarified by centrifugation for 20 minutes, at 7000g at 4°C

The supernatant (S_1) was precipitated on ice with 0.30 volumes of Chloroform-methanol (3:5v/v) according to the method of (Aksnes and Njaa 1981). Homogenization was performed on ice using Potter-Elvehjem homogenizer and clarified by centrifugation for 20 minutes at 7000g at 4°C. The supernatant (S_2) obtained was used for the assay of SOD activity, which is based on its ability to inhibit the oxidation of epinephrine by superoxide anion (Misra and Fridovich 1972) One unit of SOD activity is the amount of the enzyme required for 50% inhibition of the oxidation of epinephrine to adrenochrome per minute. The amount of MnSOD was analysed in the presence of 1mM NaCN to suppress Cu-ZnSOD activity and the cytosolic Cu-ZnSOD activity was determined as the difference between

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total and cyanide-sensitive enzyme activity (Crapo,et al,1978).The enzyme activities were assayed with an SP 1800 UV/VISS Spectrophotometer.

The method described for *Dioscorea cayenensis* was then repeated for *Dioscorea alata* (Kurudu and Chembeli).

Statistical analysis

The data are presented as mean ± SEM. The values were compared using the Student T-test. The significant level was set at $P < 0.05$.

RESULTS AND DISCUSSION

The activities of superoxide dismutase (SOD) in different yam species are given in Table 1. SOD activities were found in all varieties of yam tested, but actual levels varied somewhat between varieties.

The levels of SOD activities in the three varieties of yam significantly differ ($P < 0.05$) from one another with *Dioscorea Cayenensis* being higher than *Dioscorea alata* Poir cv chembeli while *Dioscorea alata* Poir cv Kurudu has the lowest value. It is apparent that in yam tubers the activity of SOD is much higher in the head than in the middle and the tail physiological regions. The order of activity of the enzyme in the tubers were head > middle > tail. The relative amount of SOD activity in the yam tubers is generally similar to previous report (Isamah et al, 2000).

The activities of the enzyme were inhibited when the homogenates from the different varieties of yam were incubated with 1mM sodium cyanide (final reaction volume for 20 minutes. Eukaryotic cells have different forms of SOD (Weisiger and Fridovich 1973_a; Weisiger and Fridovich 1973_b) the cytoplasmic containing copper and Zinc (CuZnSOD) and the mitochondrial containing manganese (Mn SOD). Cyanide is an inhibitor of CuZnSOD while the MnSOD is insensitive to cyanide (Chantal et al, 1975). It appears, thus that identical SODs, (CuZnSOD and MnSOD) are present in the cytosol and mitochondria of the yam cells of the different varieties studied.

SOD which is inducible (Fridovich,1974) is one of the endogenous scavengers that plays an essential role in cellular defence against reactive oxygen radicals under physiological conditions (Coudray et al,1995). Increased SOD activity may be caused by a higher metabolism and a higher oxygen consumption (Aksnes and Njaa, 1981). The higher levels in *Dioscorea cayenensis* relative to *Dioscorea alata* obtained in this study may potend natural adaptations to protect the yam from enhanced oxygen radical toxicity and serves to extend the shelf- lives of the corm.

Thus, different yam species have different SOD activities that confer peculiar protective role against superoxide anion.

Table 1: Superoxide dismutase activity in different varieties of yam tuber.

Varieties of yam	Superoxide dismutase activity (Unit/g wet tissue)		
	Head	Middle	Tail
Dioscorea cayenensis poir cv Okpai	29.72 ± 0.31^a	25.60 ± 0.22^a	22.00 ± 0.84^b
Dioscorea alata Poir cv Kurudu	16.00 ± 0.80^b	15.10 ± 0.89^b	7.24 ± 0.67^a
Dioscorea alata Poir cv Chembeli	25.30 ± 0.77^b	23.52 ± 0.24^b	22.00 ± 0.12^b

Results are expressed as mean ± SEM of five determinations. Values with different superscripts in a row are significantly different ($P<0.05$).

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