

ลักษณะสมบัติทางชีวเคมีของซูเปอร์ออกไซด์ดีสมิวเทสจากรากหนอนตายหยาก *Stemona  
tuberosa* Lour



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BIOCHEMICAL CHARACTERIZATION OF SUPEROXIDE DISMUTASE FROM THE ROOTS  
OF *Stemona tuberosa*

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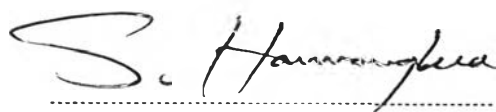
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พลอยพัฒน์ นิยมพลอย : ลักษณะสมบัติทางชีวเคมีของซูเปอร์ออกไซด์ดิสมิวเทสจากราก  
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เอนไซม์ซูเปอร์ออกไซด์ดิสมิวเทสและไซโคลไทด์ซึ่งเป็นเปปไทด์ที่มีฤทธิ์ทางชีวภาพถูก  
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 การต้านเซลล์มะเร็งสูงที่สุดในขณะที่ไซโคลไทด์ชื่อว่า kalata S ไม่พบรายงานโครงสร้างแบบทุติยภูมิจึง  
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 คล้ายคลึงกับ kalata B1 ซึ่งเป็นไซโคลไทด์ที่ในปัจจุบันนำมาประยุกต์ใช้ในการออกแบบยา



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PLOYPAT NIYOMPLOY: BIOCHEMICAL CHARACTERIZATION OF SUPEROXIDE  
DISMUTASE FROM THE ROOTS OF *Stemona tuberosa*. ADVISOR: ASSOC. PROF.  
POLKIT SANGVANICH, Ph.D., CO-ADVISOR: ASST. PROF. APHICHART  
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Superoxide dismutase (SOD) and bioactive cyclotide were firstly discovered from the root of *Stemona tuberosa* and the leaves of *Viola sumatrana*, respectively. Crude protein from *S. tuberosa* was determined SOD activity and showed the highest SOD activity compared to other ten plants. The crude protein was then performed in two experimental developments. The first experiment is the development of non-denaturing two dimensional gel electrophoresis coupled with SOD staining activity using bovine erythrocyte as a positive control. This technique can be used for the separation of different SOD isozymes. The second experiment was a purification and characterization of SOD from *S. tuberosa*. The purified SOD (ST-1) which has the highest SOD activity compared to other SODs in this plant was characterized as a purified Mn-SOD. In addition, crude protein from leaves of *V. sumatrana* was also purified and yielded four bioactive cyclotides. The cyclotides were determined their cytotoxicity on four different human cancer cell lines. The result showed that cyclotide called cycloviolacin O2 showed the most potency on cell cytotoxicity. While, cyclotide called kalata S which lack of secondary structure data then it was identified using NMR experiment. The NMR results showed that the secondary structure of kalata S is similar to kalata B1 which is a well-known cyclotide using as a drug design application.



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## LIST OF ABBREVIATIONS

$\mu\text{g}$	microgram
$\mu\text{l}$	microliter
$\mu\text{g} / \mu\text{l}$	microgram per microliter
$\mu\text{M}$	microMolar
$^{\circ}\text{C}$	Degree Celsius
cm	centimeters
cDNA	Recombinant DNA
DNA	Deoxyribonucleic acid
Da	Dalton
DEAE	Diethylaminoethyl
DMEM	Delbecco Modified Eagle's Medium
DMSO	Dimethylsulfoxide
g	gram
h	hour
HPLC	High performance liquid chromatography
mg	milligram
mg/ml	milligram per milliliter
min	minute
mm	millimeter
mM	milimolar
MTT	(3,(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide

nm	nanometer
RNA	Ribonucleic acid
rpm	round per minute
TFA	trifluoroacetic acid
w/w	weight by weight

