## Hydroxyproline-containing Citrus Esterase (第1報) 精製および性質

久保田幸穂, 庄司省三, 船越崇行, 塩永一寛, 植木 寛 薬学誌雑 (YAKUGAKU ZASSHI), 103 (6), 655-661 (1983)

## Hydroxyproline-containing Citrus Esterase. I. Purification and Properties of the Enzyme from Citrus natsudaidai HAYATA

Yukiho Kubota\*, Shozo Shoji\*, Takayuki Funakoshi\*, Kazuhiro Shionaga\*, and Hiroshi Ueki

ABSTRACT An esterase was separated from the exocarp of *Citrus natsudaidai* HAYATA in a highly purified form. The enzyme hydrolyzes p-nitrophenyl esters of fatty acids, phenyl acetate, and tosylarginine methyl ester at varying rates. Of the substrates tested, p-nitrophenyl acetate, for which the specific activity was determined to be 18.60 units per mg of protein, was most rapidly hydrolyzed. The esterase was free from carboxypeptidase and other proteolytic activities. The esterase had a pH optimum at pH 5.5 for p-nitrophenyl acetate. It was strongly inhibited by HgCl<sub>2</sub>. The other metal ions or inorganic anions tested show no significant effect on the enzymatic activity. The values of  $s_{20,w}$  and  $D_{20,w}$  were 1.8S and  $4.5 \times 10^{-7}$  cm<sup>2</sup>/s, respectively, and the molecular weight was calculated to be 40000 from these values. The enzyme was composed of 323 amino acid residues: Trp<sub>2</sub>, Lys<sub>21</sub>, His<sub>9</sub>, Arg<sub>10</sub>, Hyp<sub>23</sub>, Asp<sub>24</sub>, Thr<sub>18</sub>, Ser<sub>35</sub>, Glu<sub>22</sub>, Pro<sub>20</sub>, Gly<sub>40</sub>, Ala<sub>22</sub>, Cys(half)<sub>2</sub>, Val<sub>18</sub>, Met<sub>3</sub>, Ile<sub>12</sub>, Leu<sub>17</sub>, Tyr<sub>14</sub>, Phe<sub>11</sub>.

抄録 ナツミカンの外果皮から単離したエステラーゼ中に従来の酵素タンパク質中には検出されないヒドロオキシプロリンが酵素 1 モル当り 2 3 モル含まれることを見出した。本酵素は脂肪酸のp-ニトロフェニルエステル,フェニルアセテートや TAME などを種々の程度に水解した。このうちp-ニトロフェニルアセテートを最も良く水解した。本酵素中には,カルボキシペプチダーゼやタンパク分解酵素活性は見出されなかった。本酵素は pH5.5 の最適 pH を有し, HgCl2 によって強く阻害されたが,他の金属イオンや無機イオンによっては阻害されなかった。 s20,w は 1.8S, D20,w は  $4.5 \times 10^{-7}$  cm²/s で,これから算出した分子量は 40,000 であった。 3 2 3 個のアミノ酸残基の組成は, Tr2, Ly21, Hi39, Ar310, Hy323, As324, Th718, Se735, Gl1022, Pr7020, Gl340, Al322, Cy36(h316)2, V3118, M613, Il312, Il312, Il313, Il314, Il314, Il315, Il315, Il316, Il316, Il317, Il317,

\* Faculty of Pharmaceutical Sciences, Kumamoto University 熊本大学薬学部