

## 負電荷密度の異なる粘土ナノシート上でのタンパク質の吸着挙動と酵素活性の評価

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Research on adsorption behavior and enzyme activity of HRP on clay nanosheets with different negative charge densities (1 Tokyo Metropolitan Univ. Faculty of Urban Environmental Sciences, 2 Tokyo Metropolitan Univ. Graduate school of Urban Environmental Sciences) ○ Katsuya Sato<sup>1</sup>, Yuta Oiwake<sup>2</sup>, Kyosuke Arakawa<sup>2</sup>, Tetsuya Shimada<sup>2</sup>, Tamao Ishida<sup>2</sup>, Shinsuke Takagi<sup>2</sup>

Horseradish peroxidase (HRP) is an enzyme that accelerates a decomposition of hydrogen peroxide. In our previous research, it was turned out that HRP absorbed on clay nanosheet (Sumecton SA (Sap 1.2)) which has negative charges on the surface, was able to work for *tert*-BuOOH as a substrate, while HRP in solution did not show any activity. In this research, Sap 1.0, Sap 1.2, Sap1.4 that has same layered structure as Sumecton SA with different negative charge densities on the surface were used as an adsorbent and the enzyme activity of HRP adsorbed on each nanosheet was examined. The numbers after 'Sap' mean that the average distance (nm) between negative charges when each negative charge array is a hexagonal array. As a result, the activity was enhanced when nanosheets with higher charge densities were used. The results indicate that the structures of HRP were different when it is adsorbed on different nanosheets. The HRP structure will be examined by using the CD and fluorescence measurement.

**Keywords :** Clay Nanosheet; Horseradish Peroxidase(HRP); Adsorption; Enzyme Activity; Protein

西洋わさびペルオキシダーゼ (HRP) は過酸化水素を基質特異的に分解するタンパク質である。当研究室では HRP を表面がアニオン性を帯びている粘土ナノシート (スメクトン SA (Sap1.2)) に吸着させることで、本来は反応性の殆ど無い *tert*-BuOOH を基質に用いた場合の活性が向上することを報告した<sup>1)</sup>。本研究では、粘土ナノシートとして電荷密度の異なる Sap1.0、Sap1.2、Sap1.4 (数字はそれぞれの負電荷の配列がヘキサゴナル配列である仮定したときの平均負電荷間距離 (nm) を表す) を用いた。それぞれに HRP を吸着させたとき、酵素活性がどのように変化するかを検討した。

その結果、電荷密度がより大きいナノシートに吸着した場合に活性の向上が確認された。この理由は電荷密度の異なるナノシートに吸着したときに HRP の構造が異なるためであると考えられる。今後 CD、蛍光スペクトルを用いて詳細な検討を行う。

1) T. Arai, M. Tabuchi, Y. Sato, T. Ishida, T. Shimada, S. Takagi, *Langmuir*. **2020**, 36, 8384-8388.

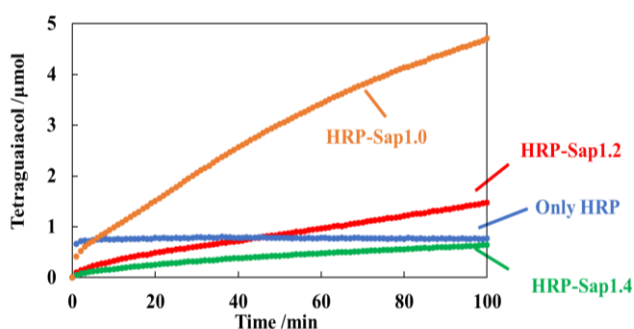


Figure 1. Enzyme activity of HRP for *tert*-BuOOH as a substrate on the nanosheets in water. (orange) Sap1.0, (red) Sap1.2, (green) Sap1.4, (blue) without clay nanosheet. Vertical axis indicates reaction activity.