Supplementary Material

Selective Hydrolysis of Ovalbumin Promoted by Hf-substituted Wells-Dawson-Type Polyoxometalate

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1 Supplementary Figures

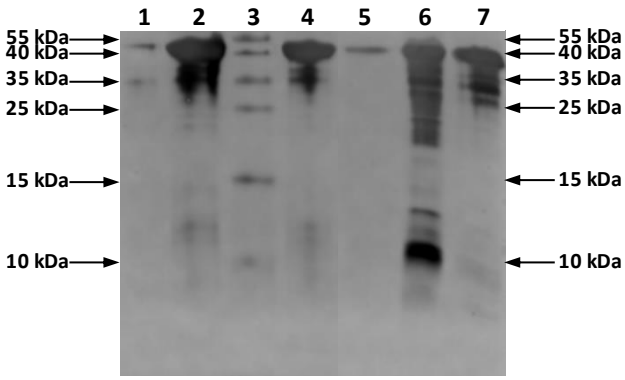

Figure *S1*. Silver-stained SDS-PAGE gel of OVA in the presence of Hf1-WD2.
OVA (0.02 mM) was incubated with 100 equivalents of Hf1-WD2 for 2 days at 60 °C in phosphate buffer (10.0 mM, pH 7.4), acetate buffer (10.0 mM, pH 4.4) or Tris-Cl buffer (10.0 mM, pH 9.0). Lanes 1-10 from left to right: (1) OVA only at pH 4.4, (2) OVA only at pH 7.4, (3) protein ladder, (4) OVA only at pH 9.0, (5) OVA + Hf1-WD2 at pH .4.4, (6) OVA + Hf1-WD2 at pH 7.4, (7) OVA + Hf-1WD2 at pH 9.0.

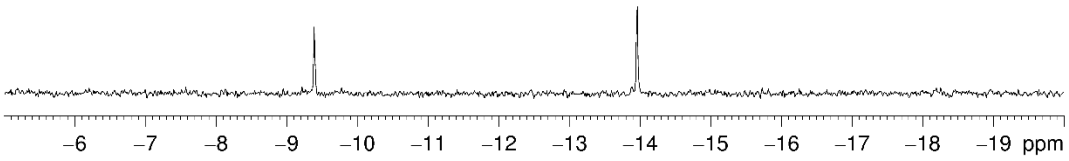

Figure *S2*. ^{31}P NMR spectrum of Hf1-WD2 POM in 100 % D_2O at RT.

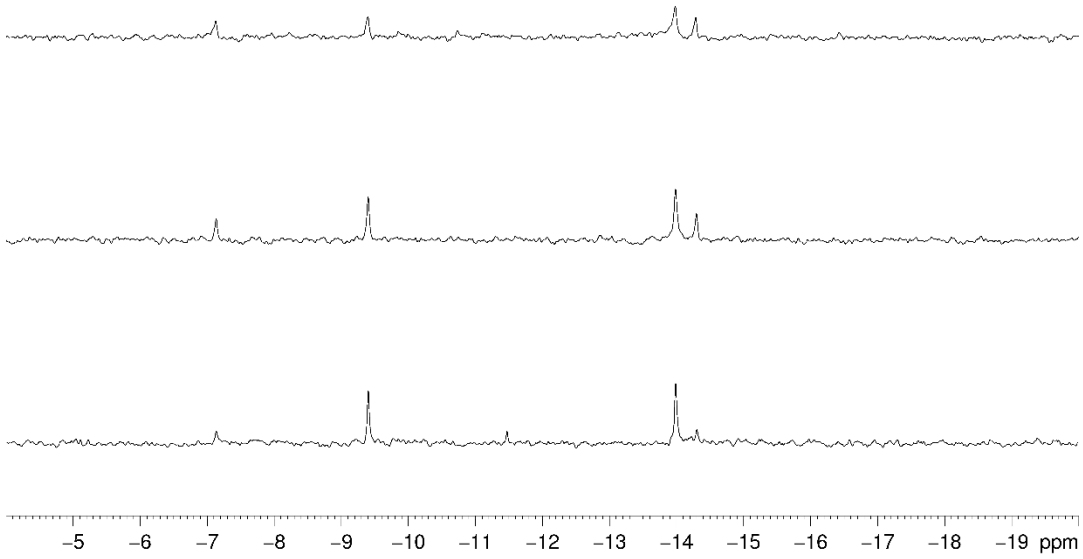

Figure *S3*. ^{31}P NMR spectra of Hf1-WD2 in phosphate buffer.
The ^{31}P NMR spectra of Hf1-WD2 (2.0 mM) is shown in phosphate buffer (10.0 mM, pH 7.4, 10 % D_2O) without OVA at RT (bottom), with 0.4 mM OVA at RT (middle), and with 0.4 mM OVA after 7 days at 60 °C (top).

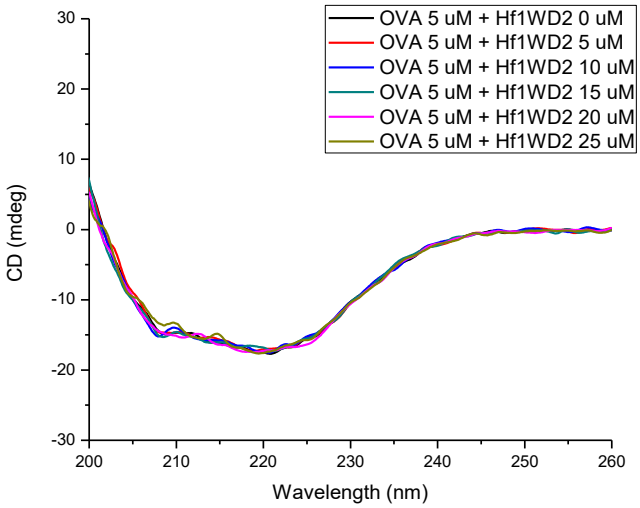

Figure *S4*. CD plot of OVA (5.0 uM) with increasing concentrations of Hf1-WD2 in phosphate buffer (10.0 mM, pH 7.4) after mixing.

Hf1-WD2 POM

XGSIGAASMEFCFDVFKELKVHHANENIFYCPIAIMSALAMVYLGAKD
STRTQINKVVRFDKLPGFGDSIEAQCGTSVNVHSSLRDILNQITKPNDV
YSFSLASRLYAEERYPILPEYLQCVKELYRGGLEPINFQTAADQARELINS
WVESQTNGIIRNVLQPSSVDSQTAMVLVNAIVFKGLWEKAFKDEDTQ
AMPFRVTEQESKPVQMMYQIGLFRVASMASEKMKILELPFASGTMS
MLVLLPDEVSGLEQLESIINFEKLTEWTSSNVMEERKIKVYLPRMKMEE
KYNLTSVLMAMGITDVFSSSANLSGISSAESLKISQAVHAAHAEINEAG
REVVGSAEAGVDAASVSEEFRADDHPFLFCIKHIATNAVLFFGRCVSP

Figure *S5*. Primary amino acid sequence of OVA with the hydrolysis sites induced by Hf1-WD2 POM indicated in purple.

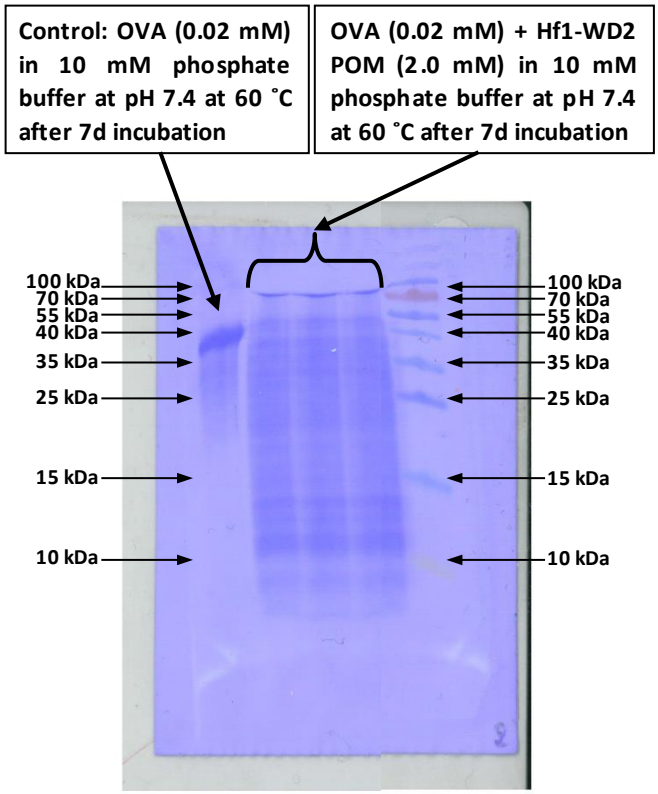

Figure *S6*. Coomassie stained blot of OVA hydrolysis by Zr1-WD2 POM or Hf1-WD2 POM.
An SDS-PAGE gel of OVA (0.02 mM) hydrolyzed by Zr1-WD2 POM or Hf1-WD2 POM (2.0 mM) after incubation in phosphate buffer (10.0 mM, pH 7.4) for 7d at 60 °C was blotted on a PVDF membrane and Coomassie stained.

2 Supplementary Tables

Table S*1*. Overview of N-terminal amino acid sequences determined by Edman Degradation and the corresponding peptide bonds hydrolyzed by Hf1-WD2 POM.

Sequence determined by Edman degradation	**Hydrolyzed peptide bond**
DHPFLFXIXH	Ala361-Asp362
HPFLFXI	Asp362-His363
FLFXIXXIAT	Pro364-Phe365
DVFXELXV	Phe13-Asp14
XVESQTN	Ser148-Trp149
DQAXELINS	Ala139-Asp140
DVYSFSLAS	Asn95-Asp96
DILNQ	Arg85-Asp86