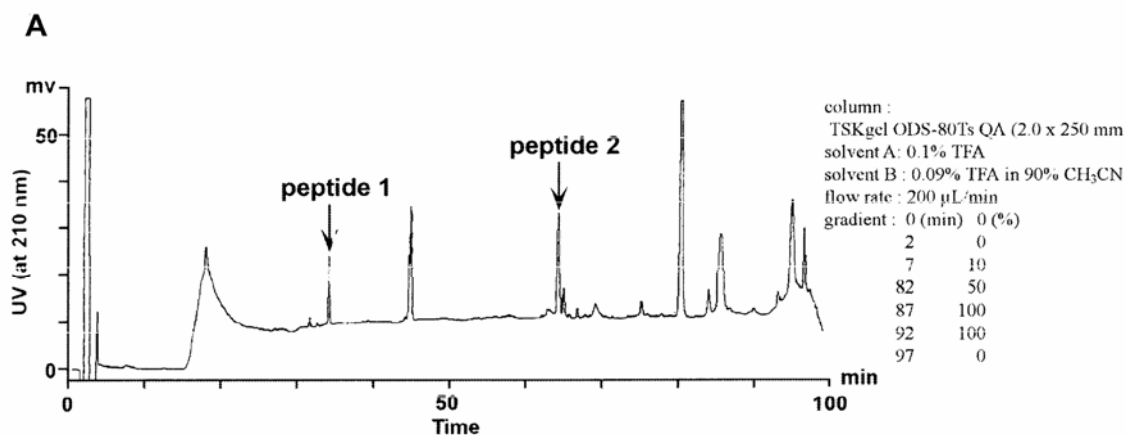
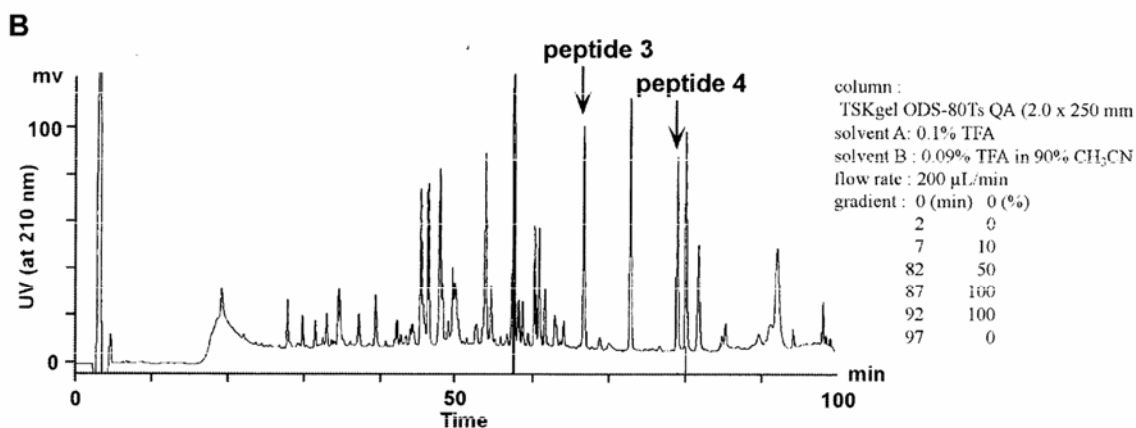


## SUPPORTING INFORMATION



**Peptide 1: WVEQLK**  
**Peptide 2: YTFLQFDPAP...**

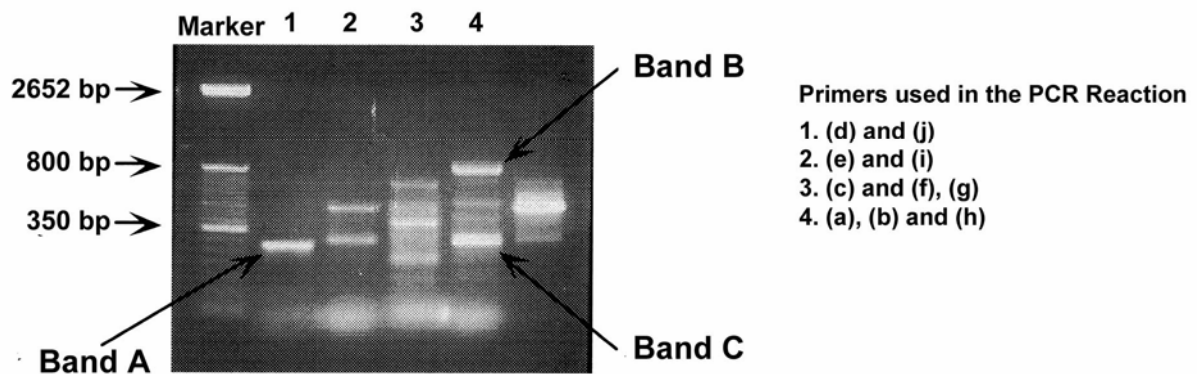


**Peptide 3: FFFTDQDNTT....**  
**Peptide 4: LTESQYVQNA**

**Figure S1.** HPLC analysis of endopeptidase Lys-C digest for Edman degradation. The purified proteins (**A**) OABP1 and (**B**) OABP2 were digested with endopeptidase Lys-C, and separated by TSKgel ODS-80Ts QA ( $\phi$  2.0 x 250 mm). The column was equilibrated with Solvent A of 0.1% TFA at a flow rate of 0.2 mL/min, and loaded with the samples. The Solvent B of 0.09% TFA in 90% CH<sub>3</sub>CN was linearly raised from 0 to 10% in the period of 2-7 min, from 10 to 50% in the period of 7-82 min and from 50 to 100% in the period of 82-87 min. The Solvent B was kept running for another 5 min, and then the solvent was replaced with Solvent A for conditioning. Amino acid sequences of two peptides in each analysis were determined by Edman degradation as listed below the HPLC charts.

**A**

	amino acid sequence	sense strand	antisense strand
OABP1	<u>WVEQLK</u>	(a) 5'TGGGTNGARCARCTNAA3'	(f) 5'TTNAGYTGTCNACCCA3'
		(b) 5'TGGGTNGARCARTTAA3'	(g) 5'TTYAAYTGTCNACCCA3'
OABP2	<u>YTFLQFDPAP</u>	(c) 5'CARTTYGAYCCNGCNCC3'	(h) 5'GGNGCNGGRTCRAAYTG3'
	<u>LTESQYVQNA</u>	(d) 5'CARTAYGTNCARAAAYGC3'	(i) 5'GCRTTYTGNACRTAYTG3'
	<u>FFFTDQDNNTT</u>	(e) 5'TTYTTYACNGAYCARGAYAA3'	(j) 5'TTRTCYTGRTCNGTRAARAA3'

**B**

**Figure S2.** Amplification of cDNA coding OABP1 and OABP2. **(A)** The degenerated forward and reverse primers based on the amino acid residues of OABP1 and OABP2 determined by Edman degradation (14). The letters Y, N and R represents C/T, A/C/G/T and A/G, respectively. **(B)** Agarose gel electrophoresis of PCR products. Lanes 1 and 2 (both for OABP2), and lanes 3 and 4 (both for OABP1) represent the results from individual pairs of primers shown in the right. Bands A, B and C were subjected to the sequence analysis.

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OABP1           WVEQLKQCKQLEEASVKLLCEKAREVLS
P11611    1  MDDKTFTKELDQWVEQLNECKQLNENQVRTLCEKAKEILT
                *****:*****:* .*: *****:***:

OABP1           QESNVQKVKSPVTVCGDVHGGQFHDLMELFRIGGDSPTDNY
P11611    41  KESNVQEVRCPVTVCGDVHGGQFHDLMELFRIGGKSPDNTY
                :*****:*.*****:*****:*****

OABP1           LFMGDYVDRGYYSVETVTLVVALKVRYPNRITLLRGNHES
P11611    81  LFMGDYVDRGYYSVETVTLVVALKVRYPERITILRGNHES
                *****:***:*****

OABP1           RQITQITQVYDECLRKYGNPNVWKF'FDLFDYLP'LTALVD
P11611   121  RQITQVYGFYDECLRKYGNANVWKYF'FDLFDYLP'LTALVD
                *****: .*****.*****:*****:*****

OABP1           DQIFCLHGGLSPSIDTLDHIRSLDRLQ'VEPHEGPMCDLLW
P11611   161  GQIFCLHGGLSPSIDTLDHIRALDRLQ'VEPHEGPMCDLLW
                .*****:*****:*****

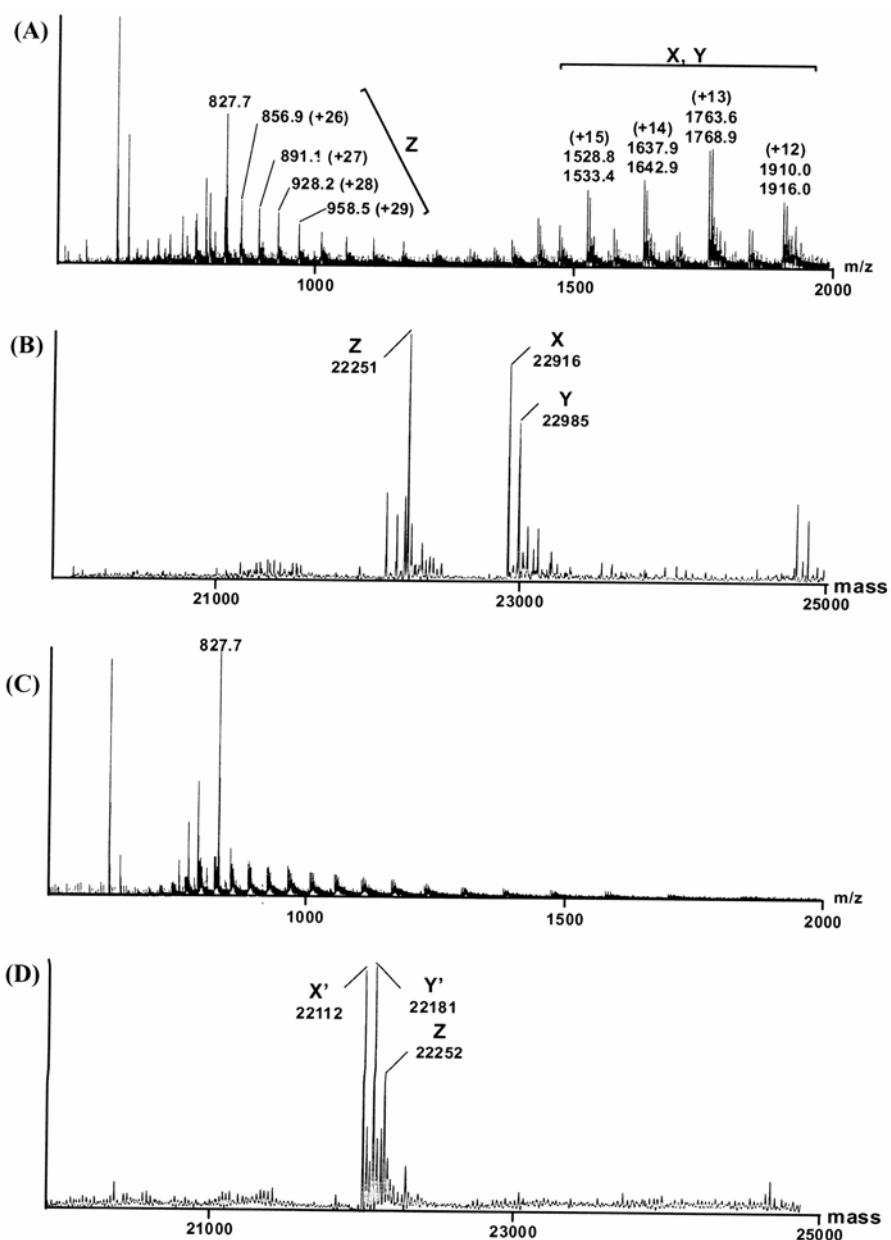
OABP1           SDPDDRGGWGISPRGADYTF'GMDISENFNHNNGLT'LVSRA
P11611   201  SDPDDRGGWGISPRGAGYTF'GQDISETFNHANGLT'LVSRA
                *****.******.******:***

OABP1           HQLVMEGYNWCHERNVVTIF'SAPNYCYRCGNQAAIMELDD
P11611   241  HQLVMEGYNWCHDRNVVTIF'SAPNYCYRCGNQAAIMELDD
                *****:*****:*****

OABP1           GLKYTF'FLQFDPA
P11611   281  TLKYSF'FLQFDPAPRRGEPHVTRRTPDYFL
                ***:*****

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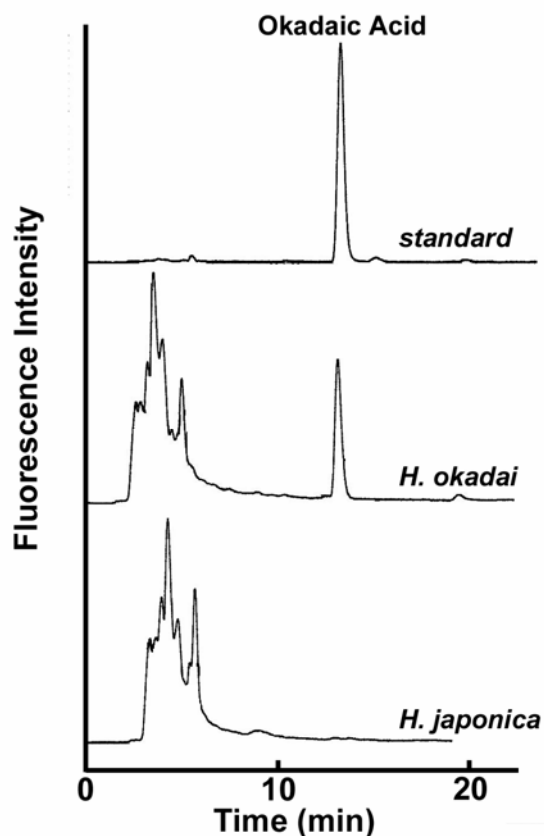
**Figure S3.** Alignment of protein sequences for OABP1 and a rabbit PP2A $\beta$  catalytic subunit (GenBank accession number: P11611). They are 88% homologous (16). OABP1 catalyzed hydrolysis of *p*NPP with 710 pmol/min/ $\mu$ g (13).



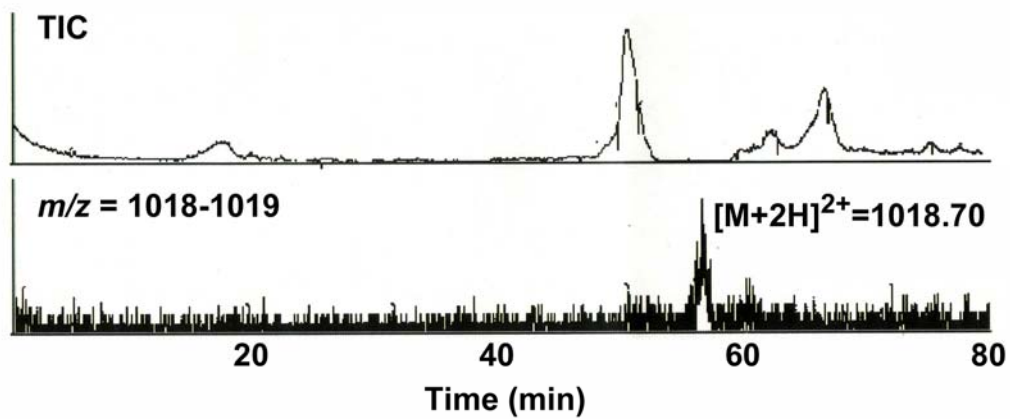
**Figure S4.** ESI-MS spectra of OABP2. **(A, C)** Raw and **(B, D)** deconvoluted mass spectrum for OABP2 measured in **(A)** 5% MeOH and **(C)** 50% CH<sub>3</sub>CN. The X, Y and Z are attributed to OABP2.1, OABP2.2 and OABP2.3, respectively. The peak at 827.7 corresponding to [M+Na]<sup>+</sup> for okadaic acid was present in both of the conditions. The peaks X and Y in 5% MeOH gave larger *m/z* than those observed in 50% CH<sub>3</sub>CN by 804, indicating that OABP2.1 and OABP2.2 possibly form a complex with okadaic acid. The deconvoluted peak assigned to OABP2.3 was independent of the measurement condition.

AB078740	1		A	ACA	ATT	CGA	CTG	TGA	CTT	ATT	TTG	GAA					
AB078740	29	ATG	GCT	AAT	TTA	AAG	GAG	CCA	TCA	GCT	CAC	TGG	TGC	AGG	AAG	ATG	
OABP2.1	1	(M <sup>a</sup> )	A <sup>b</sup>	N	L	K	E	P	S	A	H	W	C	R	K	M	
AB078740	74	CGG	ACA	GTT	TTT	CGG	CCA	TGG	GAT	GTG	GAA	GGA	GGT	TCA	AAA	GGC	
OABP2.1	15	R	T	V	F	R	P	W	D	V	E	G	G	S	K	G	
AB078740	119	TAT	GTC	ACG	GAA	GAG	GTC	TTT	AAA	GAC	GGC	GTT	CAA	AGA	AGG	CTG	
OABP2.1	30	Y	V	T	E	E	V	F	K	D	G	V	Q	R	R	L	
AB078740	164	GAG	AAA	TTT	CCC	GAG	CTT	GCC	CCT	ACG	AAA	GAC	AAA	ATG	TAC	GAG	
OABP2.1	45	E	K	F	P	E	L	A	P	T	K	D	K	M	Y	E	
AB078740	209	CGA	TCA	CAT	CGT	CAC	TGG	GTC	AAC	CAC	TGC	AAC	CTG	GGG	GTC	AAG	
OABP2.1	60	R	S	H	R	H	W	V	N	H	C	N	L	G	V	K	
AB078740	254	ATG	CCT	GAA	GGT	TAC	CGA	CTA	ACC	GAA	TCG	CAG	TAT	GTC	CAG	AAT	
OABP2.1	75	M	P	E	G	Y	R	<u>L</u>	<u>T</u>	<u>E</u>	<u>S</u>	<u>Q</u>	<u>Y</u>	<u>V</u>	<u>Q</u>	<u>N</u>	
AB078740	299	GCC	TGG	CTG	CTC	ATT	CAC	TCG	CCT	GAT	TTT	GAA	GCA	AGT	TTG	AAA	
OABP2.1	90	<u>A</u>	W	L	L	I	H	S	P	D	F	E	A	S	L	K	
AB078740	344	GAA	TCC	AGC	CAA	ACT	TTT	TGG	GAA	GGA	ATC	GAT	AGA	GAA	AAG	AAG	
OABP2.1	105	E	S	S	Q	T	F	W	E	G	I	D	R	E	K	K	
AB078740	389	GGA	TAC	ATC	ACA	AAG	GAG	GAA	GCT	ACT	AAA	CTC	GGT	ATC	CGA	GTC	
OABP2.1	120	G	Y	I	T	K	E	E	A	T	K	L	G	I	R	V	
AB078740	434	ACA	AAA	GAC	CCG	AAT	CTC	AAG	AGC	ACT	GGC	ATC	TTT	GAG	GCT	ATG	
OABP2.1	135	T	K	D	P	N	L	K	S	T	G	I	F	E	A	M	
AB078740	479	GAT	GAA	AAA	AAT	ACT	GGG	AGG	ATC	ACG	TTT	GAG	GAT	ACG	CTC	AAG	
OABP2.1	150	D	E	K	N	T	G	R	I	T	F	E	D	T	L	K	
AB078740	524	GCA	CAA	CTT	TTC	TTC	TTC	ACA	GAC	CAG	GAT	AAC	ACA	ACT	CAT	CCT	
OABP2.1	165	A	Q	L	<u>F</u>	<u>F</u>	<u>F</u>	<u>T</u>	<u>D</u>	<u>Q</u>	<u>D</u>	<u>N</u>	<u>T</u>	<u>T</u>	H	P	
AB078740	569	TTC	AAC	TAT	GTG	AGG	GGA	GCA	CTT	GTG	GAT	TAG					
OABP2.1	180	F	N	Y	V	R	G	A	L	V	D	stop					
AB078740	614	GAA	ATT	CAA	CTG	AAC	AGT	TGC	AGG	GGT	CAA	AAA	GGA				

**Figure S5.** The complete cDNA sequence of OABP2.1 and its amino acid sequence. The peptide sequences determined by Edman degradation were underlined. The MS mapping demonstrated that the posttranslational modification occurs at *N*-terminus. The superscripts <sup>a</sup> and <sup>b</sup> represent (a) deletion of the methionine and (b) acetylation on the alanine in *N*-terminus.



**Figure S6.** Quantification of okadaic acid in the sponges *H. okadai* and *H. japonica*. To 300  $\mu\text{L}$  of the crude extract from each sponge was added 200  $\mu\text{L}$  of 1 mg/mL 9-anthryldiazomethan (ADAM) in MeOH (28). After incubation at room temperature for 60 min in the dark, the mixture was evaporated, loaded to a silica gel column, and eluted with  $\text{CHCl}_3$  followed by 5% MeOH/ $\text{CHCl}_3$ . The latter fraction was collected, and analyzed with HPLC on an ODS column Mightysil RP-18 GP 4.6 x 250 mm using 8:1:1  $\text{CH}_3\text{CN}$ -MeOH- $\text{H}_2\text{O}$  at flow rate of 1.1 mL/min. The wavelength of excitation and emission were set at 365 nm and 412 nm, respectively. HPLC chromatogram for 50 ng of 9-ADMOA (top), 32 mg of *H. okadai* extract (middle) and 52 mg of *H. japonica* extract (bottom) provided evidence that okadaic acid is not contained in *H. japonica*.



$$(1018.7 \times 2 - 2) - 1450 = 585.4$$

According to Fig. 4B, this peptide is deduced to be D<sup>139</sup>PNLK.

**Figure S7.** Identification of the photoaffinity labeled OABP2 fragment by ESIMS. After photolabeling of OABP2, the protein was precipitated in acetone, and resuspended in 10 mM Tris/HCl (pH 9.0). The mixture was digested with endopeptidase Lys-C for 12 h, and then purified with Immunopure Immobilized Monomeric Avidin (Pierce, Illinois, USA). ESIMS analysis was carried out as described in *Experimental Procedures*.