gel slice #	1 <sup>a</sup>	2	3	4	5	6	7	8	9	1-9
apparent Mr (kDa)	>201	121-200	81-120	71-80	51-70	46-50	25-45	20-24	6-19	10->201
total spectra	144,558	165,475	188,699	184,148	210,706	177,400	231,982	119,325	172,220	1,594,513
matching spectra <sup>b</sup>	13,360	26,213	30,688	20,428	39,675	21,885	36,241	10,133	19,168	217,791
# of protein IDs	39	103	142	100	240	155	303	121	246	654
protein FDR <sup>c</sup>	1.7%	0.8%	0.6%	0.9%	0.4%	0.6%	0.0%	0.0%	0.4%	0.5%
peptide FDR <sup>c</sup>	0.1%	0.1%	0.1%	0.0%	0.0%	0.0%	0.0%	0.0%	0.1%	0.0%

Table S1. Summary statistics for mass spectra and protein identifications

<sup>a</sup> 1–9: protein samples from gel slices 1–9; <sup>b</sup> matching spectra: number of spectra that match certain proteins in the database; <sup>c</sup> FDR: false discovery rate.

Table 52. A list of 702 proteins identified in the gel-fractionated and ACN-treated plasma sample	Table S2. A	list of 702	proteins identified	in the gel-f	ractionated and	d ACN-treated	plasma sam	ples
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#	name	Sea	N/N/		IH apl	<i>p</i> -value	IH/CH	IF/CF	IH/CH	IH/CH	<i>p</i> -value
#	name	Sey.		Cirgei	in gei	gel	gel	RNA	RNA	ACN	ACN
670	4Tox	MQFS	23	0, 0, 0	0, 0, 9	0.4	5.8	344.4	77.5	N.A.	N.A.
298	AMP	MSKIF	11	19, 26, 23	26, 18, 22	0.5	0.9	N.A.	N.A.	0.7	0.3

#: protein identification numbers with the intracellular ones shaded green; protein name: proteins related to immunity are shaded pink; sequence: predicted signal peptide is in green font; MW: theoretical molecular mass in kDa; CH or IH gel: raw spectral counts for each identified protein in three biological replicates of the control (C) and induced (I) plasma samples separated by SDS-PAGE; *p*-value gel: results of the Student's t-tests conducted using normalized spectral counts (NSCs) of proteins identified after gel separation (p < 0.05: green); IH/CH gel: ratios of average NSCs of gel-separated proteins (I/C >1.67: red; I/C <0.60: blue); IF/CF or IH/CH RNA: relative abundances (RAs) or adjusted read numbers (ARNs) of mRNAs in fat body and hemocytes (RA or ARN >5, red; RA or ARN <0.2: blue; N.A.: not available) (22); IH/CH ACN: recalculated I/C ratios of NSCs of proteins identified in the ACN-treated samples (24) (I/C >1.67: red; I/C <0.60: blue); *p*-value ACN: results of the Student's t-tests conducted using NSCs of proteins identified in the supernatants after ACN treatment (p < 0.05: green).

See the attached Excel file for the entire Table S2. A portion of it is shown above.

	CH1	CH2	CH3	IH1	IH2	IH3
CH1	1	0.98	0.99	0.95	0.95	0.94
CH2		1	0.97	0.95	0.94	0.94
CH3			1	0.94	0.94	0.93
IH1				1	0.99	0.99
IH2					1	0.99
IH3						1

#### Table S3. Pearson pairwise correlation among biological replicates

#	Name	MW	IH/CH	D	IF/CF	ІН/СН	ІН/СН	p
		(kDa)	gel	٢	RNA	RNA	ACN	P
395	diapausin5	7	N.A	N.A.	N.A.	N.A.	0.00	0.00
385	ELRRP8	64	0.03	0.02	N.A.	N.A.	N.A.	N.A.
209	HP6	39	0.92	0.55	1.57	1.56	0.00	0.00
62	HP8	41	0.94	0.73	1.88	5.94	0.52	0.00
153	HP21	46	0.69	0.21	0.77	3.09	0.44	0.02
468	IML-9	33	0.13	0.00	1.94	1.48	N.A.	N.A.
325	inducible metallo-PI	18	6.46	0.37	0.84	N.A.	0.54	0.03
155	peroxidasin	155	0.56	0.00	1.02	0.89	N.A.	N.A.
351	ADP/ATP translocase	33	0.09	0.00	0.71	1.19	N.A.	N.A.
220	aldo-keto reductase 2E	36	0.67	0.16	N.A.	N.A.	0.22	0.01
71	aldo-keto reductase-2	41	0.76	0.13	0.82	N.A.	0.28	0.01
108	aminoacylase	44	0.72	0.18	0.00	0.57	0.56	0.00
355	antennal BP 7b	15	1.00	0.96	N.A.	N.A.	0.00	0.01
21	apolipophorin-like	475	0.48	0.02	0.71	0.34	0.00	0.37
318	BCP inhibitor precursor	12	0.36	0.09	48.33	1.70	0.60	0.01
103	cathepsin L1	38	0.92	0.66	0.81	1.33	0.40	0.02
584	chaperonin 1y	59	0.18	0.00	0.57	1.36	N.A.	N.A.
329	chemosensory Pr2	13	1.36	0.43	N.A.	N.A.	0.15	0.01
585	Cl- intracellular channel isoform 1	30	0.38	0.04	0.49	1.06	N.A.	N.A.
282	collagen and Ca binding EGF domains 1	49	0.47	0.01	0.34	5.15	0.59	0.00
231	collagen α-1(IV) chain-like	116	0.17	0.00	0.67	0.80	N.A.	N.A.
129	collagen α-2(IV) chain pr.	183	0.34	0.01	0.70	0.72	0.21	0.29
192	CXE	43	0.49	0.02	1.36	N.A.	N.A.	N.A.
194	dipeptidase 2	46	0.78	0.00	2.96	0.24	0.31	0.02
494	FK506-BP1, FKBP59 like	53	0.13	0.05	0.45	0.94	0.00	0.37
171	flavin-dependent monooxygenase FMO2	52	0.68	0.05	1.19	1.19	0.00	0.00
238	furin	144	0.85	0.69	0.51	1.00	0.00	0.00
589	GST o1	24	0.11	0.02	0.33	0.53	N.A.	N.A.
434	GST σ2	24	0.55	0.01	1.27	0.40	N.A.	N.A.
560	histone H2A-like pr. 2	13	0.22	0.04	N.A.	0.95	N.A.	N.A.
527	Hp DDB_G0277507	28	0.00	0.03	N.A.	N.A.	N.A.	N.A.
266	Hp DicpuDraft_159622	13	1.09	0.58	1.36	1.35	0.19	0.02
417	Hp KGM_05474	38	0.50	0.01	N.A.	N.A.	N.A.	N.A.
245	Hp KGM_06733	77	0.58	0.04	1.10	0.98	0.92	0.96
80	Hp secreted	22	0.60	0.03	0.66	0.60	0.55	0.01
131	hydroxypyruvate isomerase	29	0.86	0.16	0.92	0.74	0.42	0.00
119	isocitrate DH	46	0.83	0.06	0.96	1.16	0.17	0.00
336	JH esterase	65	0.03	0.00	1.17	0.25	N.A.	N.A.
340	larval cuticle pr. 16/17	12	0.98	0.96	0.08	N.A.	0.04	0.00
531	mating type pr. MAT1-1-1	10	0.00	0.00	0.03	0.34	0.00	0.00
230	Msex010104-RA	26	0.55	0.04	3.06	5.83	0.52	0.01
142	OBP1	20	0.73	0.28	0.29	N.A.	0.48	0.03
48	Ommochrome BP	31	0.81	0.04	0.94	N.A.	0.43	0.02
99	peroxisomal N1-acetyl-spermine/spermidine oxidase-1	53	0.65	0.01	N.A.	N.A.	0.25	0.01
227	peroxisomal N1-acetyl-spermine/spermidine oxidase-2	53	0.64	0.04	N.A.	N.A.	0.00	0.00
660	pr. disulfide isomerase A6	46	0.00	0.00	1.41	0.75	N.A.	N.A.
566	proteasome subunit α3	28	0.00	0.02	0.38	1.44	N.A.	N.A.
446	rpL4	49	0.21	0.04	1.02	0.96	N.A.	N.A.
606	rpL5	34	0.07	0.01	0.91	0.80	N.A.	N.A.
521	rpL7	31	0.29	0.04	0.00	0.10	N.A.	N.A.
604	rpL7A	30	0.11	0.00	1.13	1.01	N.A.	N.A.
512	rpL13A	23	0.22	0.05	0.68	1.16	N.A.	N.A.
598	rpL15	24	0.00	0.01	1.09	0.91	N.A.	N.A.
631	rpL22	17	0.18	0.04	0.87	1.05	N.A.	N.A.
477	rpS3 40S	27	0.11	0.01	0.84	1.11	N.A.	N.A.
550	rpS3A	30	0.11	0.01	1.44	1.10	0.00	0.37
433	rpS4	30	0.35	0.03	1.07	0.93	N.A.	N.A.
353	rpS9	23	0.37	0.01	1.65	1.47	0.00	0.37
593	rpS13	17	0.00	0.00	1.67	1.17	N.A.	N.A.
575	rpS15A	15	0.10	0.03	1.74	1.25	N.A.	N.A.
536	rpS16	17	0.00	0.02	0.75	1.04	N.A.	N.A.
422	rpS18	18	0.25	0.04	1.53	1.08	N.A.	N.A.
537	rpS20	14	0.18	0.03	1.23	1.25	N.A.	N.A.
661	rpS27	9	0.00	0.00	1.44	1.09	N.A.	N.A.
83	secreted acidic Cys-rich pr.	38	0.32	0.01	0.66	0.72	0.20	0.00
75	tolloid-like pr. 2	158	1.12	0.46	2.07	0.59	0.45	0.01
406	transitional ER ATPase TER94	89	0.36	0.05	1.25	1.68	N.A.	N.A.
159	transl. EF 1α	50	0.57	0.02	0.94	1.05	N.A.	N.A.
565	transl. EF 1β'	24	0.00	0.03	1.00	0.89	N.A.	N.A.
449	uncharacterized inducible pr. A	7	1.70	0.59	N.A.	N.A.	0.22	0.01
149	venom acid phosphatase	45	0.48	0.00	0.79	2.92	0.13	0.00

Table S4. A list of 78 down-regulated proteins in gel-fractionated and ACN-treated samples\*

441	vitellogenic carboxypeptidase	55	0.00	0.01	0.61	0.97	N.A.	N.A.
53	α-amylase 3	58	0.66	0.14	0.77	5.83	0.41	0.00
240	β-1,3-glucanase	42	0.81	0.32	1.27	3.56	0.40	0.02
156	βA mannosidase, lysosomal	102	0.72	0.01	0.71	1.02	0.05	0.01
87	β-N-acetylglucosaminidase 1	70	0.73	0.03	1.36	0.89	0.43	0.00
113	γ-glutamyl cyclotransferase-like venom pr. 1	23	0.71	0.01	1.12	N.A.	0.54	0.02
179	γ-glutamyl hydrolase	38	0.57	0.00	N.A.	N.A.	0.23	0.07

\* See Table 1 footnotes

										3			
	Mr	slice #	1	2	3	4	5	6	7	8	9		Ι
name	(kDa)	Mr	500-200	200–120	120-80	80-70	70–50	50-45	45–25	25–20	20–10	ΣUSC	or U
nroDO1	70	Ctrl	0	10.3	49.5	171.5	121.8	27.5	43.2	0	0.7	424.5	
ριοροι	79	Ind	0	18	41.9	162.7	77.4	16.7	30.4	0	0.6	347.7	

## Table S5. Distribution of USCs of the 654 proteins identified in the gel slices \*

\* Sums of USCs for *highly abundant* ( $\Sigma$ USC: 6000–500), *abundant* (500–150), *moderate* (150–50), *low* (50–15), and *rare* (15–0) proteins are listed in cells corresponding to the gel slices; The protein names are highlighted yellow, grey, orange, green, and white accordingly. If USC = 0, the cell is shaded in light blue; If USC > 0, they are shaded in a color gradient from white to red. The USC spreading patterns are shown in the column of  $M_r$  in different colors: blue for  $M_r$  down shifted 2 or more gel slices; light blue for consistent  $M_r$  (-1 to +1 slice); green for  $M_r$  up shifted 2 slices; yellow for  $M_r$  up shifted 3 or more gel slices. In the column  $\Sigma$ USC, up- and down-regulated proteins are shaded orange and green, respectively. I for immunity-related; U for immunity-unrelated.

See the attached Excel file for Table S5. A small portion of it is shown above

щ	2222	# of distinct peptides							% coverage						identification probability %					
#	# Hame	CH1	CH2	СНЗ	IH1	IH2	IH3	CH1	CH2	CH3	IH1	IH2	IH3	CH1	CH2	CH3	IH1	IH2	IH3	
670	4Tox_SV3	0	0	0	0	0	2	0	0	0	0	0	11	0	0	0	0	0	100	
298	MGD2b	3	4	3	4	3	3	21	35	21	35	21	21	100	100	100	100	100	100	
14	apolipo3	28	25	25	24	24	24	68	66	66	66	65	66	100	100	100	100	100	100	
369	attacin1	0	0	0	3	0	3	0	0	0	78	0	83	0	0	0	100	0	100	
117	attacin2	0	0	0	15	16	14	0	0	0	79	80	80	0	0	0	100	100	100	

#### Table S6. Detailed information for the 702 identified proteins

See the attached Excel file for Table S6. A small portion of it is shown above

## **Figure legends**

## 1 Fig. S1. Correlation of the mRNA and protein abundances (A) and their *changes* (B) for the

2 **ACN-treated samples.** See Fig. 3 for the definitions of x- and y-axes.

# **Fig. S2. Relationships of ΣUSCs of proteins and numbers of the gel slices they were identified.**

- 4 Open circles (blue), control samples; open diamonds (red), induced samples. Proteins with similar
- 5  $\Sigma$ USCs were slightly shifted to reduce overlap.







