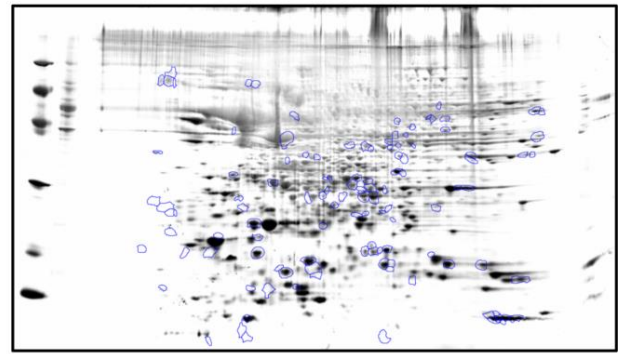
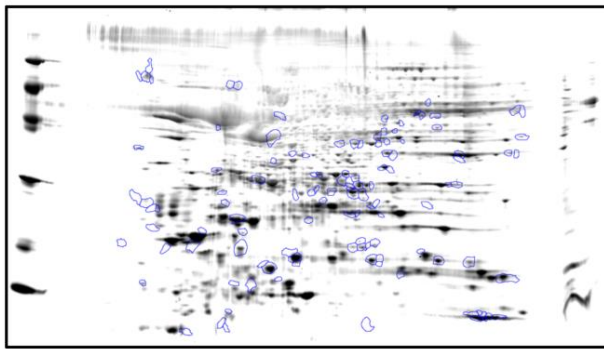


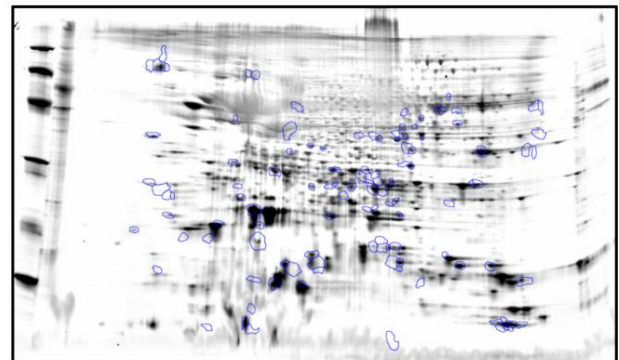
Control



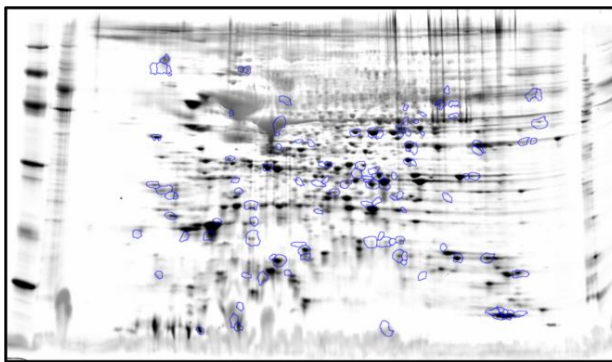
0 hour



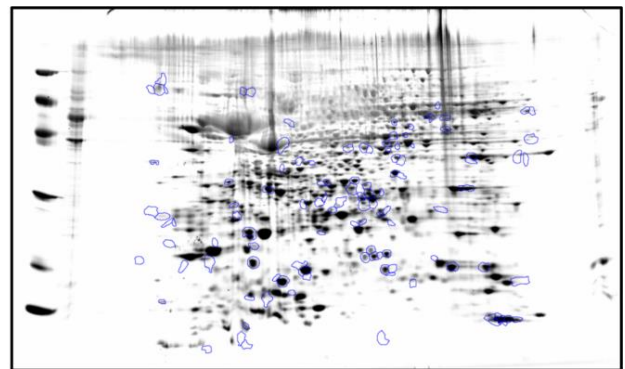
3 hour



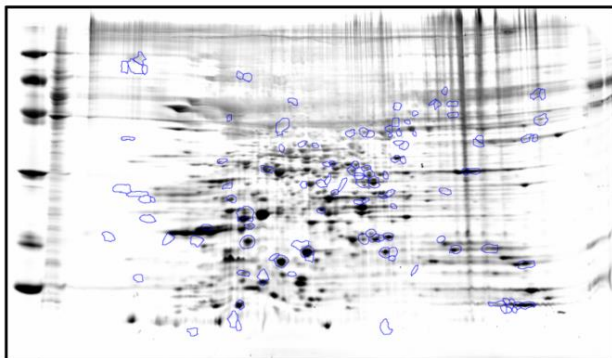
6 hour



12 hour



18 hour



24 hour

Fig. S1. Representative two dimensional SDSg PAGE gel images from each of the seven treatment groups: Control, 0 hour, 3 hour, 6 hour, 12 hour, 18 hour and 24 hour. Proteins are separated by isoelectric point (pI) (pH range 3 –) along the horizontal dimension and by relative molecular mass (M_r) along the vertical dimension. Biorad low range molecular weight standards were used for the ladder. The 115 spots selected for identification are outlined on each gel, and are labeled by spot number on the Control gel. See Table S1 for further information on protein identities.

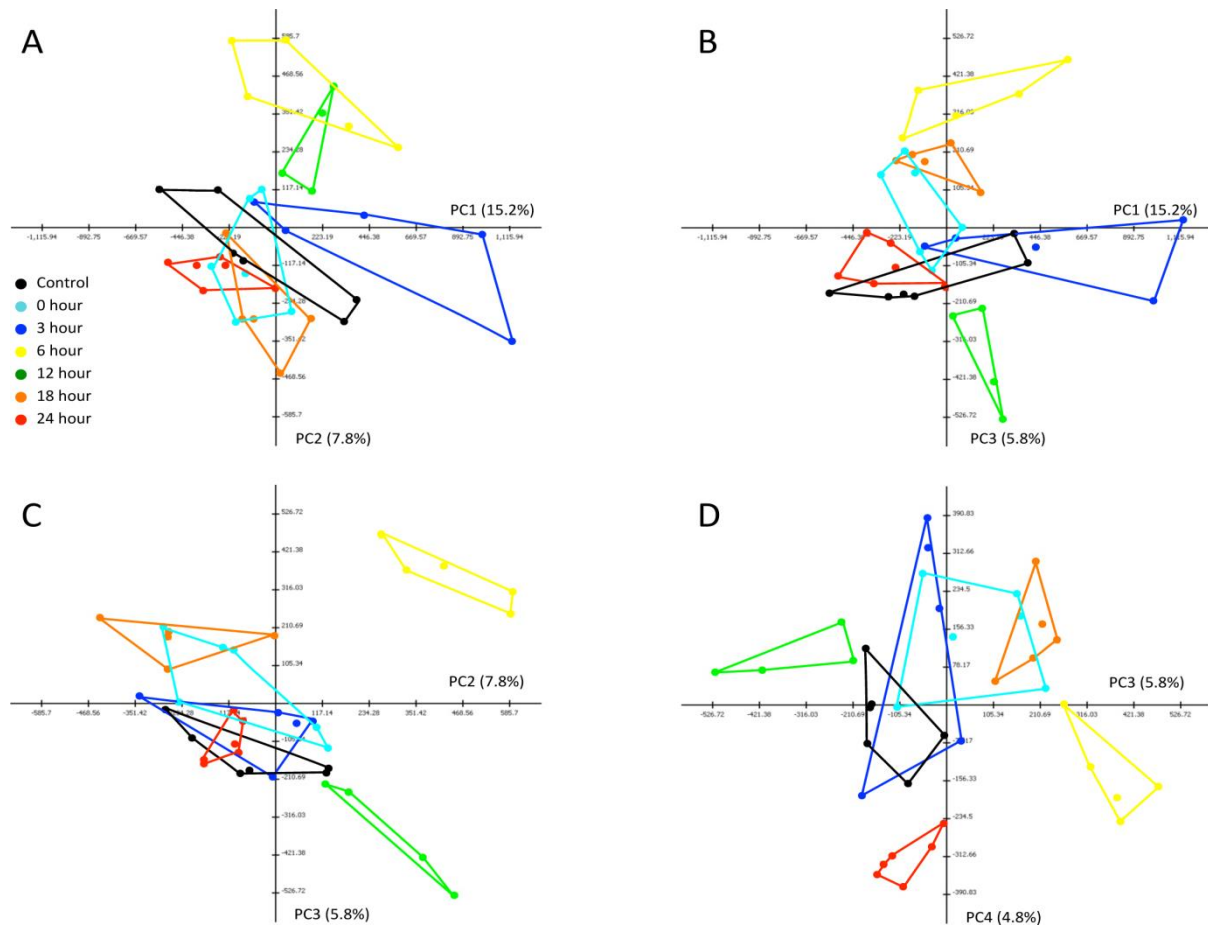


Fig. S2. Principal components analysis of 933 spots detected on twog dimensional gels using Delta2D software. Symbols represent individual mussels from the seven groups (black — ntrol; cyan — h; blue — h; yellow — h; green — h; orange — h; red — h).

- A) PC1 vs. PC2. The first PC (horizontal axis) explains the greatest amount of variance among samples 15.2 , but does not clearly separate treatment groups. We interpret this to mean that when analyzing all 933 spots on the 2D gels, most variation in PEPs among mussels is associated with inter---individual differences not associated with recovery time. The second PC (vertical axis) explains 7.8% of variance and separates the 6 h and 12 h treatments from the other groups.
- B) PC1 vs. PC3. The third PC explains 5.8% of total variance, and separates the 6 h group (positive oadings) from the 12 h group;
- C) PC2 vs. PC3. The 6 h and 12 h groups are strongly separated from the other treatments. Compare with Fig. 2A, where the 6 and 12 h treatment groups show a similar separation when the analysis is limited to the 115 spots detected as differing significantly among groups by ANOVA.
- D) PC3 vs. PC4. The fourth PC explains 4.8% of total variance, and separates the 24 h group from other treatments.

Table S1. Proteins from *Geukensia demissa* gill isolated by 2D gel electrophoresis and identified by tandem mass spectrometry. Gray shading indicates protein spots that were contiguous on the 2D gels and had identical IDs, including Genbank accession numbers, and so were combined (i.e., the Delta2D software differentiated multiple spots when there was only one). Proteins with identical names and Genbank accession numbers that are listed in separate rows exist as discrete and non-contiguous spots on the 2D gels (see Fig. S1). Red text indicates protein spots that decreased in abundance significantly at the indicated time point relative to control (all others increased in abundance). If a protein was significantly different from control at multiple time points, it is listed under the first time point at which significance was reached.

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
A. Protein spots significantly increased/decreased in abundance relative to control at 0 hours. See Fig. 3 for expression profiles.									
57	Prohibitin	182	5	XP_012945186.1	<i>Haliotis diversicolor</i>	1E-92	81	6.11	30.7
78/79	Voltage-dependent ion channel-2 (high pl)	77/255	3/8	ADI56517.1	<i>Haliotis diversicolor</i>	2E-156/ 1E-155	75/75	7.9/8.0	31.2/31.2
77	Voltage-dependent ion channel-2 (mid pl)	147	5	ADI56517.1	<i>Haliotis diversicolor</i>	1E-155	75	6.9	31.4
76	Voltage-dependent ion channel-2 (low pl)	213	8	ADI56517.1	<i>Haliotis diversicolor</i>	1E-155	75	6.7	31.5
25/26	Glyceraldehyde-3-phosphate dehydrogenase (high pl)	329/ 152	9/4	AEF33398.1	<i>Crassostrea ariakensis</i>	2E-86/ 6E-93	50/81	8.7/8.6	40.2/40.3
22/27	Glyceraldehyde-3-phosphate dehydrogenase (mid pl)	552/ 483	15/14	AEF33398.1	<i>Crassostrea ariakensis</i>	0E+00/ 0E+00	82/82	8.0/8.0	39.1/40.9
23/24	Glyceraldehyde-3-phosphate dehydrogenase (low pl)	64/96	2/3	AEF33398.1	<i>Crassostrea ariakensis</i>	4E-148/ 4E-148	72/72	7.0/7.1	40.5/40.6

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
B. Protein spots significantly increased/decreased in abundance relative to control at 3 hours. See Fig. 4 for expression profiles.									
3/4	78 kDa Glucose regulated protein	698/831	19/24	BAD15288.1	<i>Crassostrea gigas</i>	0/0	88/90	4.9/5.0	71.6/72.1
64	Small heat shock protein 24.1	345	8	AEP02968.1	<i>Mytilus galloprovincialis</i>	6E-114	68	5.5	33.0
49	Peptidylprolyl cis-trans isomerase (high pl)	418	13	EKC29243.1	<i>Crassostrea gigas</i>	3E-118	90	6.8	28.9
47/50/51	Peptidylprolyl cis-trans isomerase (mid pl)	80/259/413	3/6/16	EKC29243.1	<i>Crassostrea gigas</i>	3E-118/ 3E-118/ 3E-118	90/90/90	6.6/6.6/6.6	28.7/30.3/30.7
48	Peptidylprolyl cis-trans isomerase (low pl)	551	18	EKC29243.1	<i>Crassostrea gigas</i>	3E-118	90	6.3	29.7
58/59	Protein deglycase DJ-1	307	10	EKC37254.1	<i>Crassostrea gigas</i>	5E-70/ 5E-70	67/67	6.8/6.9	17.1/18.4
73	Universal stress protein A-like	260	9	XP_011448812.1	<i>Crassostrea gigas</i>	6E-20	82	7.9	14.4
43	Nacre protein (high pl)	171	6	BAK57311.1	<i>Pinctada fucata</i>	2E-14	31	7.0	23.9
44	Nacre protein (low pl)	101	3	BAK57311.1	<i>Pinctada fucata</i>	1E-22	31	6.6	24.6
82	mitochondrial H ⁺ ATPase subunit α (high pl)	612	18	EKC39329.1	<i>Crassostrea gigas</i>	0E+00	88	7.3	55.4
5	mitochondrial H ⁺ ATPase subunit α (low pl)	93	2	EKC39329.1	<i>Crassostrea gigas</i>	2E-164	94	6.9	53.4
6	Actin 2 (high pl)	725	22	EKC38058.1	<i>Crassostrea gigas</i>	0E+00	97	7.6	49.4
7	Actin 2 (low pl)	180	5	EKC38058.1	<i>Crassostrea gigas</i>	0E+00	99	5.5	45.9
18	Actin 2 (low pl, high Mr)	759	27	EKC38058.1	<i>Crassostrea gigas</i>	0E+00	99	5.5	55.4

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
C. Protein spots significantly increased/decreased in abundance relative to control at 6 hours. See Fig. 5 for expression profiles.									
55	Peroxisredoxin-6	152	4	ABO26614.1	<i>Haliotis discus</i>	2E-96	72	7.0	26.8
45	Nucleoredoxin	80	1	EKC27452.1	<i>Crassostrea gigas</i>	2E-40	42	5.9	11.6
72	Dyp-type peroxidase (high pI)	559	17	EKC43063.1	<i>Crassostrea gigas</i>	1E-109	71	6.5	34.4
70/71	Dyp-type peroxidase (low pI)	223/797	5/19	EKC43063.1	<i>Crassostrea gigas</i>	1E-109/5E-97	71/71	6.0/6.1	34.3/34.4
39	NADP+-dependent isocitrate dehydrogenase (high pI)	244	9	AFI56373.1	<i>Mytilus trossulus</i>	0E+00	92	7.1	48.3
40	NADP+-dependent isocitrate dehydrogenase (low pI)	364	10	AFI56373.1	<i>Mytilus trossulus</i>	0E+00	92	6.9	48.3
21	Fructose biphosphate aldolase	281	6	EKC30386.1	<i>Crassostrea gigas</i>	0E+00	85	7.0	44.2
65	Triosephosphate isomerase	143	5	AEF33397.1	<i>Crassostrea ariakensis</i>	3E-86	78	5.8	29.7
41	Malate dehydrogenase, mitochondrial	621	15	XP_005096166.1	<i>Aplysia californica</i>	1E-140	70	5.7	40.7
14	Citrate synthase	338	10	EKC35491.1	<i>Crassostrea gigas</i>	0E+00	77	7.1	52.0
12	Arginine kinase (high pI)	437	11	AGN95434.1	<i>Semisulcospira libertina</i>	3E-176	73	7.2	44.6
11	Arginine kinase (low pI)	488	10	AGN95434.1	<i>Semisulcospira libertina</i>	2E-179	73	6.8	45.7
28	Hemicentin-1	250	9	EKC35524.1	<i>Crassostrea gigas</i>	2E-20	36	6.1	23.5
56	Profilin-like	455	10	XP_005111300.1	<i>Aplysia californica</i>	3E-04	28	5.6	14.1

74/75	Vitelline membrane outer layer protein 1	147/341	6/11	EKC25506.1	<i>Crassostrea gigas</i>	2E-51/4E-50	48/48	4.7/4.7	17.6/20.4
114/116	Putative C1q domain containing protein MgC1q96 (low Mw, high pl)	445/592	9/18	CBX41745.1	<i>Mytilus galloprovincialis</i>	1E-52/1E-52	52/52	5.2/5.3	20.5/23.5
115	Putative C1q domain containing protein MgC1q96 (high Mw, low pl)	310	7	CBX41745.1	<i>Mytilus galloprovincialis</i>	1E-52	52	5.0	26.8
29/30/32/34/36/37	Heterogeneous nuclear ribonucleoprotein 27C (high pl)	171/158/174/141/117/205	5/5/8/7/4/6	EKC41770.1	<i>Crassostrea gigas</i>	6E-28/6E-28/3E-30/2E-26/1E-28/1E-27	67/67/67/59/60/67	8.8/8.7/9.0/8.8/8.7/8.7	4.4/4.7/4.9/5.0/4.6/5.2
31/33/35	Heterogeneous nuclear ribonucleoprotein 27C (low pl)	231/221/112	6/5/3	EKC41770.1	<i>Crassostrea gigas</i>	1E-28/8E-28/1E-27	60/67/61	5.2/5.3/5.3	6.6/4.8/3.6
42	N(G), N(G) dimethylarginine dimethylamino-hydrolase 1	128	2	EKC40016.1	<i>Crassostrea gigas</i>	1E-70	71	5.9	40.6

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
D. Protein spots significantly increased/decreased in abundance relative to control at <u>12 hours</u>. See Fig. 6 for expression profiles.									
8/10	Alcohol dehydrogenase type 3 (high pI)	413/437	10/13	EKC37227.1	<i>Crassostrea gigas</i>	1E-60/ 1E-60	58/58	6.6/6.7	44.0/44.0
9	Alcohol dehydrogenase type 3 (low pI)	263	8	EKC37227.1	<i>Crassostrea gigas</i>	1E-60	58	6.3	44.3
67/68/ 69	Universal stress protein-like isoform 2	178/82/ 81	7/2/2	AEF33379.1	<i>Crassostrea ariakensis</i>	3E-26/ 3E-26/ 3E-26	37/37/37	7.0/7.1/7.1	14.7/14.9/ 13.5
52	Perlucin-like	123	4	P86854.1	<i>Mytilus galloprovincialis</i>	1E-24	34	4.0	27.0
61	Putative perlucin-4	146	4	ABO26593.1	<i>Haliotis discus</i>	1E-20	38	4.4	17.0

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
E. Protein spots significantly increased/decreased in abundance relative to control at <u>18 hours</u>. See Fig. 7 for expression profiles.									
13	Cathepsin B	67	2	AEJ08755.1	<i>Crassostrea ariakensis</i>	3E-162	70	6.6	32.8
20	Flotillin-2	74	3	EKC35912.1	<i>Crassostrea gigas</i>	3E-158	70	7.6	55.2

Spot # ^a	Protein identification	Mowse score ^b	# of peptide matches ^c	Genbank accession. version	Species ^d	E value ^e	Identity (%) ^f	pI ^g (observed)	Mr (kDa) ^g (observed)
F. Protein spots significantly increased/decreased in abundance relative to control at 24 hours. See Fig. 8 for expression profiles.									
19	Eukaryotic translation initiation factor 5A-1-like	248	6	XP_005094246.1	<i>Aplysia californica</i>	4E-67	68	5.3	17.1
2	5'-AMP-activated protein kinase subunit β -2	67	2	EKC38929.1	<i>Crassostrea gigas</i>	2E-21	56	3.7	79.8
46	Nucleoside diphosphate kinase	254	9	AFK73702.1	<i>Ostrea edulis</i>	1E-64	84	8.3	14.1
1	15-hydroxyprostaglandin dehydrogenase	59	2	EKC19471.1	<i>Crassostrea gigas</i>	1E-55	55	4.9	29.2
54	Peroxisome oxidin-5, mitochondrial	167	5	EKC39509.1	<i>Crassostrea gigas</i>	1E-77	77	7.7	16.5

^aSpot numbers correspond to spot labels on the gel images in Figure S1.

^bMolecular Weight Search score returned by Mascot. Scores above 45 indicate homology between the MS/MS peptides and the *G. demissa* EST entry ($P < 0.05$).

^cNumber of non-overlapping peptides detected by MS/MS found in the *G. demissa* EST entry. If one spot matched more than one EST entry, and all EST entries matched the same Genbank sequence, this is the sum of peptides.

^dSpecies from which the most significant BLAST protein match was derived. BLAST searches were limited to molluscan nucleotide sequences.

^eE-value indicates the likelihood that the *G. demissa* EST entry matches the specified Genbank sequence by chance.

^fThe percent amino acid identity between the *G. demissa* EST entry and the specified Genbank sequence.

^gpI and Mr values were estimated by Mascot software based on location on the 2D gels.