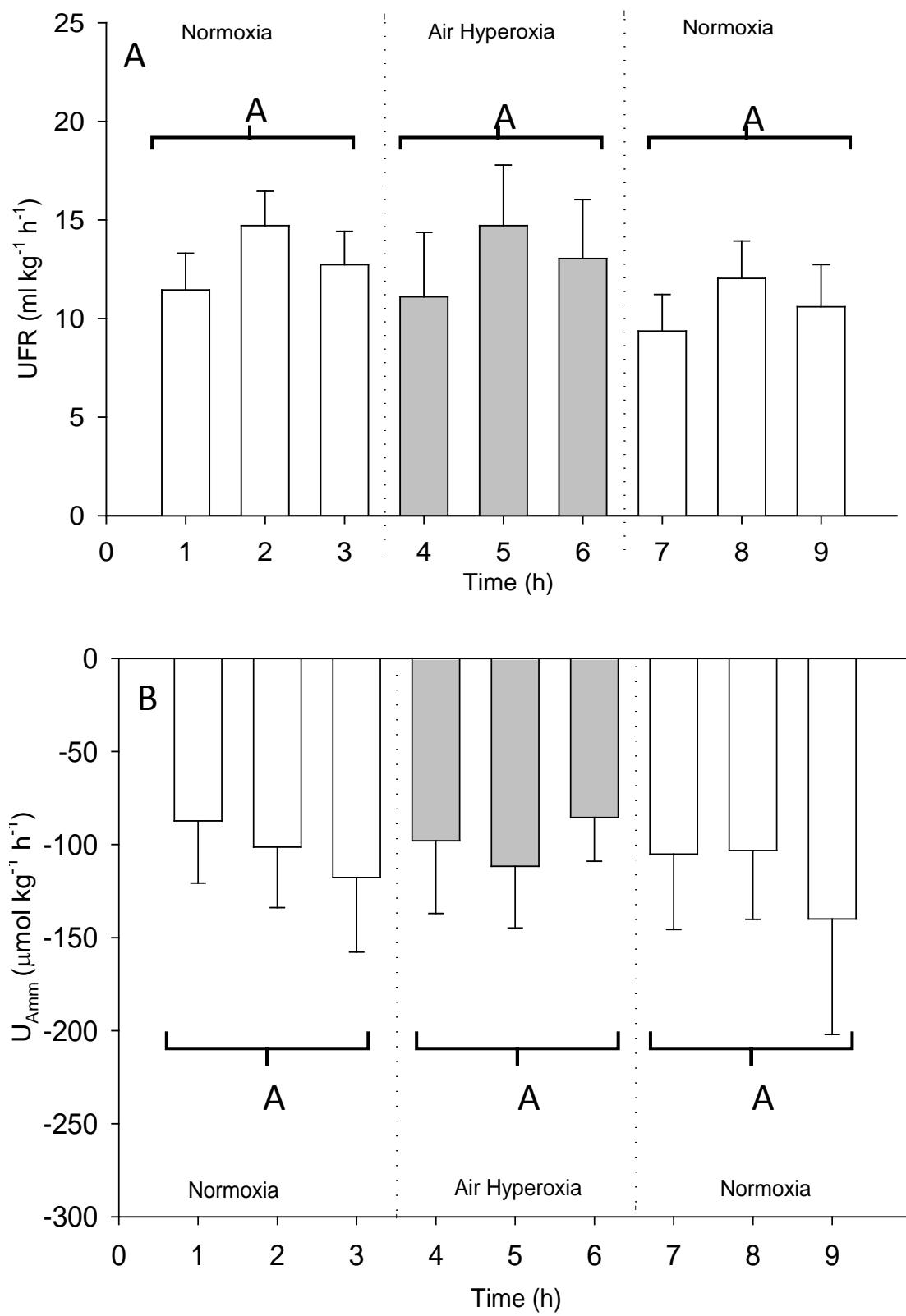


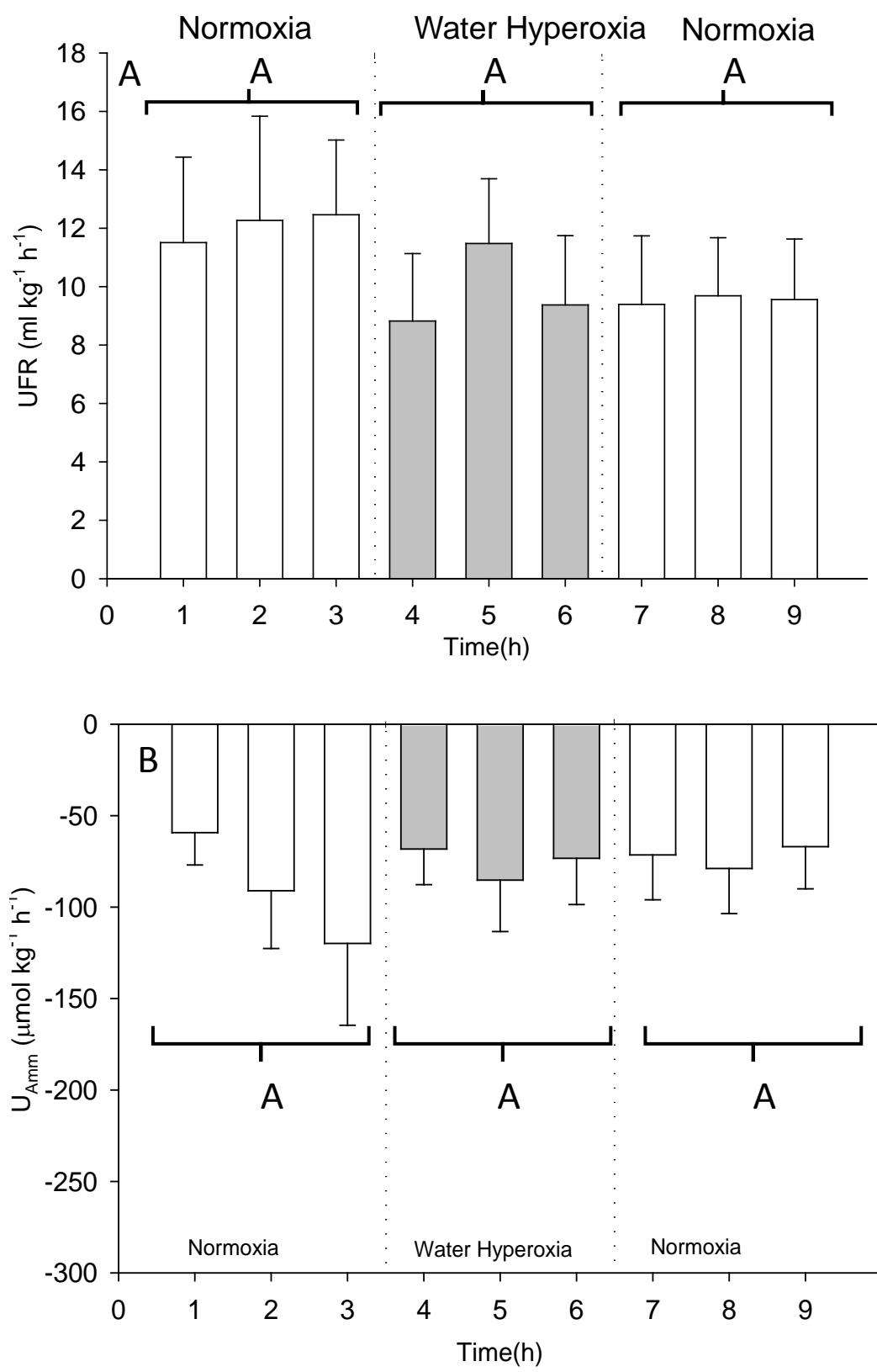
Supplementary Fig. S1

Fig. S1. The effect of **aerial hypoxia** (aquatic normoxia) on (A) urine flow rate (UFR) and (B) urinary excretion rate of total ammonia (U_{Amm}) in *A. gigas*. Aerial hypoxia was applied during a 3-h experimental treatment period, following a 3-h normoxic control period, and was followed by a 3-h normoxic recovery period. Measurements were made over 1-h intervals. Means \pm SEM ($N = 9$). There were no significant differences (i.e. $P > 0.05$) among the overall means of the 3-h periods, nor were any of the means in the 1-h intervals significantly different from the overall normoxic control mean (i.e. $P > 0.05$).



Supplementary Fig. S2

Fig. S2. The effect of **aerial hyperoxia** (aquatic normoxia) on (A) urine flow rate (UFR) and (B) urinary excretion rate of total ammonia (U_{Amm}) in *A. gigas*. Aerial hyperoxia was applied during a 3-h experimental treatment period, following a 3-h normoxic control period, and was followed by a 3-h normoxic recovery period. Measurements were made over 1-h intervals. Means \pm SEM ($N = 9$). There were no significant differences (i.e. $P > 0.05$) among the overall means of the 3-h periods, nor were any of the means in the 1-h intervals significantly different from the overall normoxic control mean (i.e. $P > 0.05$).



Supplementary Fig. S3

Fig. S3. The effect of **aquatic hyperoxia** (aerial normoxia) on (A) urine flow rate (UFR) and (B) urinary excretion rate of total ammonia (U_{Amm}) in *A. gigas*. Aquatic hyperoxia was applied during a 3-h experimental treatment period, following a 3-h normoxic control period, and was followed by a 3-h normoxic recovery period. Measurements were made over 1-h intervals. Means \pm SEM ($N = 9$). There were no significant differences (i.e. $P > 0.05$) among the overall means of the 3-h periods, nor were any of the means in the 1-h intervals significantly different from the overall normoxic control mean (i.e. $P > 0.05$).

Supplementary Table S1

Urine composition and flow rate (UFR) in *A. gigas* under control day-time conditions. Means \pm SEM (N = 29 measurements on 11 animals).

[Na ⁺] _u (mmol L ⁻¹)	0.22 \pm 0.06
[K ⁺] _u (mmol L ⁻¹)	0.10 \pm 0.01
[Cl ⁻] _u (mmol L ⁻¹)	0.17 \pm 0.03
[Total Ammonia] _u (mmol L ⁻¹)	10.81 \pm 1.37
[Urea-N] _u (mmol-N L ⁻¹)	6.79 \pm 1.58
Urine Flow Rate (ml kg ⁻¹ h ⁻¹)	12.23 \pm 1.05

Supplementary Table S2

A. Pearson's correlation coefficients for plasma components of *A. gigas* (N = 8). The one significant correlation is highlighted in bold.

	[Total CO ₂] _p	[Ammonia] _p	[Urea-N] _p	[Cl] _p	[Na ⁺] _p
[Ammonia] _p	-0.738*				
[Urea-N] _p	-0.581	0.276			
[Cl] _p	-0.354	0.178	0.101		
[Na ⁺] _p	-0.429	0.280	0.272	0.611	
[K ⁺] _p	0.175	-0.600	0.322	-0.350	-0.115

*P < 0.05

B. Pearson's correlation coefficients for components of overnight urine collections of *A. gigas* (N = 28 collections from 11 fish). Significant correlations are highlighted in bold.

	UFR	[TotalCO ₂]	[Amm]	[Urea-N]	[Cl]	[Na ⁺]
[TotalCO ₂]	0.167					
[Amm]	0.112	0.645***				
[Urea-N]	0.056	0.420*	0.281			
[Cl]	0.142	0.056	0.213	-0.156		
[Na ⁺]	-0.196	0.171	-0.210	0.089	0.146	
[K ⁺]	-0.107	0.748***	0.400*	0.160	-0.086	0.506**

*P < 0.05, **P < 0.01, ***P < 0.001

Supplementary Table S3

A. Urinary concentrations (mmol L⁻¹) of Na⁺ ([Na⁺]_u), K⁺ ([K⁺]_u), Cl⁻ ([Cl⁻]_u), total ammonia ([Amm]_u), and urea-N ([Urea-N]_u) of *A. gigas* subjected to **aerial hypoxia** (aquatic normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means ± SEM (N). Overall means in 3-h periods not sharing the same letter are significantly different (P < 0.05).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aerial Hypoxia	4-5h Aerial Hypoxia	5-6h Aerial Hypoxia	Mean Aerial Hypoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
[Na ⁺] _u	0.29	0.36	0.36	0.34^A	0.17	0.15	0.16	0.16^B	0.17	0.11*	0.29	0.19^B
SEM (8)	0.16	0.20	0.20	0.19	0.07	0.05	0.08	0.07	0.09	0.02	0.21	0.10
[K ⁺] _u	0.09	0.10	0.12	0.10	0.14	0.12	0.07	0.11	0.10	0.10	0.10	0.10
SEM (8)	0.01	0.02	0.03	0.02	0.05	0.04	0.01	0.03	0.03	0.03	0.03	0.03
[Cl ⁻] _u	0.22	0.21	0.23	0.22	0.13	0.16	0.17	0.15	0.12	0.12	0.12	0.12
SEM (8)	0.09	0.08	0.08	0.08	0.03	0.06	0.05	0.04	0.02	0.02	0.02	0.02
[Amm] _u	12.10	9.20	11.12	10.81	11.20	12.70	14.07	12.66	12.01	11.83	12.60	12.15
SEM (8)	3.32	1.66	2.14	2.09	2.71	2.66	2.89	2.68	2.72	2.87	2.77	2.64
[Urea-N] _u	0.78	0.80	0.92	0.83	0.77	0.85	0.78	0.80	0.84	0.89	0.78	0.84
SEM (8)	0.23	0.22	0.24	0.21	0.23	0.23	0.23	0.23	0.24	0.26	0.24	0.25

B.Urinary concentrations (mmol L⁻¹) of Na⁺ ([Na⁺]_u), K⁺ ([K⁺]_u), Cl⁻ ([Cl⁻]_u, total ammonia ([Amm]_u), and urea-N ([Urea-N]_u) of *A. gigas* subjected to **aerial hyperoxia** (aquatic normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means ± SEM (N).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aerial Hyperoxia	4-5h Aerial Hyperoxia	5-6h Aerial Hyperoxia	Mean Aerial Hyperoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
[Na ⁺] _u	0.19	0.15	0.17	0.17	0.18	0.15	0.13	0.15	0.12	0.14	0.12	0.13
SEM (5)	0.03	0.05	0.06	0.03	0.06	0.05	0.04	0.05	0.04	0.06	0.04	0.05
[K ⁺] _u	0.08	0.06	0.07	0.07	0.07	0.06	0.06	0.06	0.06	0.07	0.07	0.07
SEM (5)	0.01	0.02	0.02	0.02	0.02	0.02	0.01	0.02	0.01	0.02	0.01	0.01
[Cl ⁻] _u	0.09	0.08	0.08	0.08	0.08	0.07	0.08	0.08	0.08	0.08	0.09	0.08
SEM (5)	0.01	0.01	0.01	0.00	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
[Amm] _u	9.13	7.15	10.30	8.86	11.35	12.40	9.25	10.35	10.27	10.12	12.89	11.10
SEM (5)	3.80	2.74	4.20	3.24	4.50	4.40	3.86	4.17	4.04	4.10	5.23	4.42
[Urea-N] _u	0.50	0.63	0.77	0.63	0.84	0.83	0.82	0.83	0.86	0.92	0.71	0.83
SEM (5)	0.31	0.29	0.32	0.31	0.34	0.34	0.33	0.34	0.34	0.28	0.33	0.31

C. Urinary concentrations (mmol L⁻¹) of Na⁺ ([Na⁺]_u), K⁺ ([K⁺]_u), Cl⁻ ([Cl⁻]_u), total ammonia ([Amm]_u), and urea-N ([Urea-N]_u) of *A. gigas* subjected to **aquatic hypoxia** (aerial normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means ± SEM (N). * indicates significant difference from mean Control value (P < 0.05).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aquatic Hypoxia	4-5h Aquatic Hypoxia	5-6h Aquatic Hypoxia	Mean Aquatic Hypoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
[Na ⁺] _u	0.24	0.14	0.16	0.15	0.19	0.16	0.15	0.17	0.17	0.25	0.26	0.23
SEM (9)	0.04	0.04	0.04	0.04	0.06	0.04	0.04	0.04	0.04	0.11	0.09	0.07
[K ⁺] _u	0.09	0.11	0.19	0.13	0.11	0.08*	0.07*	0.09	0.09	0.08*	0.10	0.09
SEM (9)	0.01	0.02	0.09	0.03	0.02	0.01	0.01	0.01	0.02	0.01	0.01	0.01
[Cl ⁻] _u	0.18	0.17	0.18	0.18	0.25	0.21	0.19	0.22	0.16	0.21	0.19	0.18
SEM (9)	0.02	0.02	0.05	0.02	0.07	0.04	0.04	0.04	0.04	0.03	0.03	0.03
[Amm] _u	11.93	12.94	11.93	13.64	11.02	14.91	13.08	12.72	14.33	11.31	12.29	12.64
SEM (9)	3.41	3.58	3.05	3.04	3.12	4.67	4.12	3.86	4.08	2.86	3.29	3.32
[Urea-N] _u	0.34	0.54	0.47	0.45	0.56	0.54	0.41	0.50	0.56	0.55	0.82	0.64
SEM (9)	0.20	0.25	0.22	0.22	0.28	0.26	0.21	0.25	0.24	0.27	0.32	0.27

D. Urinary concentrations (mmol L⁻¹) of Na⁺ ([Na⁺]_u), K⁺ ([K⁺]_u), Cl⁻ ([Cl⁻]_u), total ammonia ([Amm]_u), and urea-N ([Urea-N]_u) of *A. gigas* subjected to **aquatic hyperoxia** (aerial normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means ± SEM (N).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aquatic Hyperoxia	4-5h Aquatic Hyperoxia	5-6h Aquatic Hyperoxia	Mean Aquatic Hyperoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
[Na ⁺] _u	0.15	0.14	0.13	0.14	0.12	0.12	0.12	0.12	0.13	0.13	0.13	0.13
SEM (7)	0.13	0.15	0.03	0.10	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03
[K ⁺] _u	0.07	0.08	0.09	0.08	0.07	0.06	0.07	0.07	0.08	0.08	0.08	0.08
SEM (7)	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02
[Cl ⁻] _u	0.21	0.15	0.15	0.17	0.15	0.15	0.16	0.15	0.16	0.181	0.15	0.17
SEM (7)	0.05	0.03	0.02	0.03	0.03	0.04	0.04	0.04	0.05	0.03	0.04	0.04
[Amm] _u	7.34	10.45	9.19	8.99	8.57	9.61	8.30	8.29	9.19	9.41	7.01	8.53
SEM (7)	2.48	3.27	2.94	2.56	2.87	3.37	2.97	3.02	3.44	3.03	2.60	2.83
[Urea-N] _u	0.65	0.59	0.71	0.65	0.59	0.76	0.60	0.65	0.68	0.63	0.59	0.63
SEM (7)	0.24	0.22	0.27	0.25	0.23	0.29	0.23	0.25	0.25	0.24	0.22	0.24

E.Urinary excretion rates ($\mu\text{mol kg}^{-1} \text{h}^{-1}$) of Na^+ (U_{Na}), K^+ (U_{K}), Cl^- (U_{Cl}), total ammonia (U_{Amm}), and urea-N ($U_{\text{Urea-N}}$), and urine flow rates (UFR) of *A. gigas* subjected to **aerial hypoxia** (aquatic normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means \pm SEM (N). Overall means in 3-h periods not sharing the same letter are significantly different ($P < 0.05$).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aerial Hypoxia	4-5h Aerial Hypoxia	5-6h Aerial Hypoxia	Mean Aerial Hypoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
U_{Na}	3.65	4.43	4.06	4.05^A	1.61	1.40	2.03	1.68^B	2.12	1.58	3.05	2.25^{AB}
SEM (8)	2.16	2.54	2.50	2.40	0.73	0.65	0.89	0.73	1.07	0.47	1.93	1.03
U_{K}	1.00	0.93	0.88	0.94	1.43	1.33	0.79	1.18	1.28	1.39	1.37	1.35
SEM (8)	0.15	0.22	0.09	0.12	0.74	0.55	0.10	0.41	0.48	0.61	0.62	0.57
U_{Cl}	2.72	2.25	1.83	2.27	1.06	1.64	2.16	1.62	1.43	1.70	1.43	1.52
SEM (8)	1.34	0.90	0.52	0.85	0.20	0.78	0.76	0.49	0.37	0.51	0.36	0.41
U_{Amm}	120.40	81.03	91.67	97.70	96.75	127.77	155.14	126.55	126.10	141.16	139.29	135.52
SEM (8)	41.68	16.11	19.61	18.45	21.32	27.26	35.11	26.24	31.23	41.23	44.16	36.81
$U_{\text{Urea-N}}$	7.28	6.78	8.03	7.36	6.99	9.01	8.48	8.16	7.90	9.03	7.06	7.99
SEM (8)	2.19	2.06	2.51	2.18	2.40	2.47	2.69	2.46	2.12	2.63	2.16	2.25
UFR	11.27	10.57	10.37	10.73	8.94	8.86	12.78	10.19	11.57	13.71	11.84	12.37
SEM (8)	1.77	2.11	2.42	2.00	1.03	1.49	2.17	0.87	1.60	2.55	2.04	2.03

F.Urinary excretion rates ($\mu\text{mol kg}^{-1} \text{h}^{-1}$) of Na^+ (U_{Na}), K^+ (U_{K}), Cl^- (U_{Cl}), total ammonia (U_{Amm}), and urea-N ($U_{\text{Urea-N}}$), and urine flow rates (UFR) of *A. gigas* subjected to **aerial hyperoxia** (aquatic normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means \pm SEM (N). Overall means in 3-h periods not sharing the same letter are significantly different, and asterisks indicate means in 1-h intervals that are significantly different from the overall normoxic control mean ($P < 0.05$).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aerial Hyperoxia	4-5h Aerial Hyperoxia	5-6h Aerial Hyperoxia	Mean Aerial Hyperoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
U_{Na}	2.09	2.35	2.49	2.31^A	2.09	2.25	1.84	2.06^A	0.76*	1.55	1.01	1.11^B
SEM (5)	0.30	0.88	1.02	0.46	0.98	0.99	0.97	0.98	0.28	0.68	0.2	0.30
U_{K}	0.92	0.86	0.91	0.90	0.69	0.80	0.70	0.73	0.61	0.78	0.76	0.72
SEM (5)	0.26	0.21	0.28	0.23	0.22	0.13	0.15	0.17	0.18	0.12	0.18	0.14
U_{Cl}	1.03	1.18	1.05	1.09	0.89	1.01	1.05	0.98	0.70	0.97	0.90	0.86
SEM (5)	0.18	0.10	0.15	0.12	0.31	0.26	0.25	0.27	0.14	0.16	0.15	0.11
U_{Amm}	87.26	101.40	117.7	102.1	98.00	111.7	85.5	98.4	105.12	103.22	140.00	116.11
SEM (5)	29.94	29.05	35.83	30.76	34.92	29.61	21.02	27.71	36.22	33.05	55.49	40.53
$U_{\text{Urea-N}}$	5.29	10.57	10.19	8.64	7.82	9.68	8.53	8.67	9.59	10.31	8.14	9.34
SEM (5)	3.98	5.34	5.23	4.67	3.61	4.53	4.05	3.99	4.09	4.15	4.47	4.08
UFR	11.45	14.71	12.73	12.97	11.10	14.71	13.05	12.95	9.36	12.04	10.60	10.67
SEM (5)	1.86	1.74	1.69	1.02	3.27	3.07	2.98	3.04	1.85	1.89	2.14	1.43

G.Urinary excretion rates ($\mu\text{mol kg}^{-1} \text{h}^{-1}$) of Na^+ (U_{Na}), K^+ (U_{K}), Cl^- (U_{Cl}), total ammonia (U_{Amm}), and urea-N ($U_{\text{Urea-N}}$), and urine flow rates (UFR) of *A. gigas* subjected to **aquatic hypoxia** (aerial normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means \pm SEM (N). * indicates significant difference from mean Control value ($P < 0.05$).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aquatic Hypoxia	4-5h Aquatic Hypoxia	5-6h Aquatic Hypoxia	Mean Aquatic Hypoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
U_{Na}	2.30	2.33	2.57	2.40	1.93	2.21	2.93	2.36	2.89	4.82	3.74	3.82
SEM (9)	1.06	1.21	1.20	1.14	1.16	1.18	1.21	1.16	1.24	2.19	1.31	1.48
U_{K}	1.14	1.36	2.35	1.62	0.70*	0.84*	1.11	0.88	1.16*	1.21*	1.36	1.24
SEM (9)	0.34	0.25	1.51	0.58	0.15	0.20	0.14	0.14	0.30	0.27	0.30	0.25
U_{Cl}	1.46	1.54	2.55	1.76	1.28	1.39	1.54	1.37	1.66	2.58	1.88	1.95
SEM (9)	0.35	0.40	0.41	0.33	0.41	0.42	0.48	0.43	0.44	0.85	0.52	0.59
U_{Amm}	140.88	172.77	136.53	155.67	74.19*	120.21	183.58	126.00	157.27	149.17	158.42	154.95
SEM (9)	46.87	57.84	34.93	42.47	26.22	38.29	67.55	41.06	44.00	45.07	53.25	46.60
$U_{\text{Urea-N}}$	3.64	0.54	5.15	3.11	5.11	6.95	4.63	5.56	6.42	5.55	10.18	7.38
SEM (9)	1.54	0.25	2.77	1.47	2.85	4.11	1.96	2.93	2.99	3.15	4.07	3.31
UFR	13.73	12.83	13.66	13.39	7.95*	10.04	13.87	10.62	12.41	14.19	12.60	13.07
SEM (9)	2.19	2.58	2.95	2.31	2.51	2.67	2.84	2.37	1.97	3.08	2.12	2.34

H.Urinary excretion rates ($\mu\text{mol kg}^{-1} \text{h}^{-1}$) of Na^+ (U_{Na}), K^+ (U_{K}), Cl^- (U_{Cl}), total ammonia (U_{Amm}), and urea-N ($U_{\text{Urea-N}}$), and urine flow rates (UFR) of *A. gigas* subjected to **aquatic hyperoxia** (aerial normoxia) during the experimental treatment period. Hourly values, as well as mean values (in bold) for the 3-h pre-treatment control period, 3-h treatment period, and 3-h post-treatment recovery period are shown. Means \pm SEM (N).

	0-1h Control	1-2h Control	2-3h Control	Mean Control	3-4h Aquatic Hyperoxia	4-5h Aquatic Hyperoxia	5-6h Aquatic Hyperoxia	Mean Aquatic Hyperoxia	6-7h Recovery	7-8h Recovery	8-9h Recovery	Mean Recovery
U_{Na}	1.90	2.16	1.77	1.94	0.96	1.43	1.02	1.14	1.17	1.26	1.02	1.15
SEM (7)	0.55	0.85	0.48	0.53	0.36	0.42	0.37	0.35	0.44	0.40	0.30	0.38
U_{K}	0.62	0.69	1.07	0.79	0.55	0.59	0.60	0.58	0.65	0.66	0.75	0.69
SEM (7)	0.17	0.23	0.27	0.19	0.16	0.14	0.16	0.15	0.18	0.12	0.19	0.15
U_{Cl}	1.80	1.63	1.85	1.76	1.06	1.52	1.31	1.30	1.21	1.41	1.16	1.26
SEM (7)	0.35	0.50	0.37	0.28	0.25	0.34	0.34	0.23	0.31	0.22	0.17	0.22
U_{Amm}	59.30	91.08	119.89	90.09	68.28	85.23	73.34	75.61	71.50	78.91	64.90	71.77
SEM (7)	17.65	31.57	44.74	27.89	19.48	28.13	25.27	23.71	24.49	24.62	23.07	23.18
$U_{\text{Urea-N}}$	8.15	6.17	12.67	9.00	6.73	8.32	7.86	7.64	7.68	7.18	6.62	7.16
SEM (7)	4.79	4.02	6.14	4.56	3.47	3.92	4.04	3.78	3.97	3.86	3.66	3.83
UFR	11.50	12.27	12.46	12.08	8.82	11.48	9.37	9.89	9.39	9.69	9.56	9.54
SEM (7)	2.93	3.57	2.56	2.25	2.31	2.21	2.37	2.06	2.35	1.99	2.07	2.09