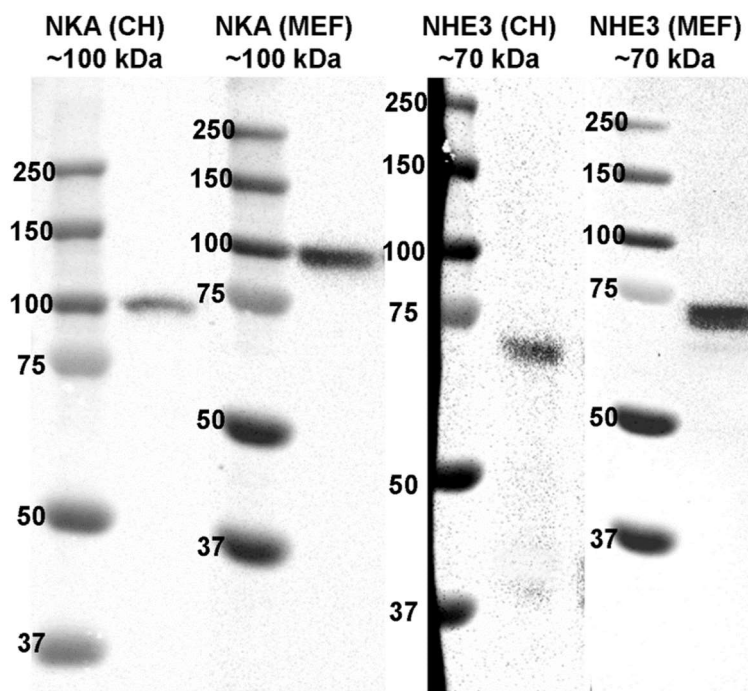


**Fig S1. A.** Na<sup>+</sup>/K<sup>+</sup>-ATPase (NKA, red) and Na<sup>+</sup>/H<sup>+</sup> Exchanger 3 (NHE3, green) immunostaining within European sea bass gill. Ionocytes containing NKA and NHE3 (NKA-I) were observed on the filament (F) and base of lamellae (L). **B.** Negative controls (no primary antibodies) had no discernible signal. Nuclei (blue) are stained with DAPI.



**Fig. S2.** Antibody verification on European sea bass gill tissue. The crude homogenate (CH) and membrane-enriched fraction (MEF) of sea bass gill tissue were immunolabeled with antibodies against Na<sup>+</sup>/K<sup>+</sup>-ATPase (NKA) and Na<sup>+</sup>/H<sup>+</sup> Exchanger 3 (NHE3). Molecular markers are shown on the left of each respective blot. Each antibody immunodetected a single band matching the predicted size of respective protein (~100 kDa for NKA, ~70 kDa for NHE3).

**Table S1.** Water chemistry of gill irrigation chambers used while blood sampling fish

	Exposure length			
	0 min	~10 min	~40 min	~135 min
Temperature (°C)	14.00 ± 0.00	14.05 ± 0.15	14.00 ± 0.30	13.95 ± 0.15
pH (NBS)	8.14 ± 0.02	6.95 ± 0.02	6.91 ± 0.01	6.91 ± 0.04
Salinity	34.6 ± 0.4	35.6 ± 0.5	34.8 ± 0.7	34.6 ± 0.7
pCO <sub>2</sub> (kPa)	0.060 ± 0.010	0.965 ± 0.045	1.227 ± 0.164	1.024 ± 0.088
TA (μM)	3120 ± 372	2834 ± 36	3301 ± 361	2750 ± 27

**Table S2.** Mean ± s.e.m. of water chemistry parameters within isolation tanks and gill irrigation chambers during low alkalinity hypercapnia exposure

Tank	Duration (min)	pH (NBS)	Temperature (°C)	Salinity	pCO <sub>2</sub> (kPa)	TA (μM)
Isolation	135.0 ± 4.6	5.70 ± 0.03	13.78 ± 0.07	34.0 ± 0.1	1.141 ± 0.020	188 ± 12
Gill irrigation	n/a	5.53 ± 0.31	14.05 ± 0.05	35.1 ± 0.3	1.288 ± 0.104	188 ± 129

**Table S3.** Mean ± s.e.m. of water chemistry parameters within isolation boxes prior to gill sampling. Gill samples from sea bass exposed to hypercapnia were taken from 5 sea bass immediately after flux measurements were completed.

Treatment	Duration (min)	pH (NBS)	Temperature (°C)	Salinity	pCO <sub>2</sub> (kPa)	TA (μM)
Normocapnia	n/a	8.05 ± 0.00	14.12 ± 0.06	33.6 ± 0.0	0.055 ± 0.001	2335 ± 7
Hypercapnia	132.9 ± 2.6	6.97 ± 0.02	14.14 ± 0.02	33.5 ± 0.0	0.770 ± 0.037	2365 ± 9

**Table S4.** Theoretical calculations of H<sup>+</sup> excretion by NHE in response to environmental hypercapnia. Calculations based on Parks *et al.* 2008. If Na<sup>+</sup><sub>i</sub>/Na<sup>+</sup><sub>e</sub> > H<sup>+</sup><sub>i</sub>/H<sup>+</sup><sub>e</sub> then H<sup>+</sup> excretion by NHE is thermodynamically unviable.

Treatment	Ionocyte Na <sup>+</sup> (Na <sup>+</sup> <sub>i</sub> , mM)	Seawater Na <sup>+</sup> (Na <sup>+</sup> <sub>e</sub> , mM)	Ionocyte H <sup>+</sup> (H <sup>+</sup> <sub>i</sub> , mM)	Seawater H <sup>+</sup> (H <sup>+</sup> <sub>e</sub> , mM)	Na <sup>+</sup> <sub>i</sub> /Na <sup>+</sup> <sub>e</sub>	H <sup>+</sup> <sub>i</sub> /H <sup>+</sup> <sub>e</sub>
Control	140	480	3.98e <sup>-08</sup>	8.91e <sup>-09</sup>	0.292	4.467
Hypercapnia	140	480	5.01e <sup>-08</sup>	1.12e <sup>-07</sup>	0.292	0.447
Hypercapnia + low TA	140	480	5.01e <sup>-08</sup>	2.00e <sup>-06</sup>	0.292	0.025