



Fig. S1. Steps involved in the analysis of protein synthesis rate in larvae of *Crassostrea gigas*.

(A) High-performance liquid chromatography (HPLC) analysis showing the amounts (moles in relative fluorescence units) of free amino acids extracted from 7-d-old larvae of *C. gigas* (Family 6). Labels on peaks are glycine, G, and serine, S. The corresponding analysis of all ^{14}C -labeled amino acids in this sample is shown in panel D.

(B) HPLC analysis for purity of the ^{14}C -labeled glycine stock solution (purchased commercially from Perkin Elmer, Inc.). HPLC eluent was collected continuously throughout the 30 minute analysis using a fraction collector every 30 seconds; each fraction was assayed for radioactivity. ^{14}C -labeled glycine contained 95.4% of the radioactivity, with 4.6% in ^{14}C -labeled serine.

(C) Distribution of ^{14}C -label in the free amino acid pool following a 6-min exposure of larvae to seawater containing the stock solution of ^{14}C -glycine. Note the ratio of radiolabeled glycine and serine present in larvae, is similar to that in the original stock of commercially-supplied ^{14}C -labeled glycine (Panel B).

(D) Distribution of ^{14}C -label in the free amino acid pool after a 36-min exposure of larvae to seawater containing ^{14}C -glycine. The percentage of ^{14}C -label in serine increased slightly to 7.9% at this last, 36-min time point of the protein synthesis assay (see panel E). This represents only a 3.3% (i.e., the difference between 7.9% and 4.6%) interconversion of the original stock of ^{14}C -labeled glycine to ^{14}C -labeled serine during the 36-min assay. Since the mole-percent of serine in whole-body protein of larvae of *C. gigas* is $6.1\% \pm 0.1$ (Lee et al., 2016) – approximately half of that of glycine, at $12\% \pm 0.2$ mole-percent – the small percent of ^{14}C -serine in the free amino acid pool has a minimal effect on the calculation of protein synthesis rate.

(E) Specific activity of ^{14}C -glycine in the precursor pool of free amino acids in larvae sampled at different time points during a 36-min time-course assay. Specific activity of glycine [ratio of ^{14}C -labeled (Bq) glycine to total glycine (moles)] was calculated for each of the six different time points using, for example, the moles of total glycine (36 min, panel A) and the corresponding amount of ^{14}C -labeled glycine present in larvae at a given time point (36 min, panel D). Change in the specific activity of ^{14}C -glycine was used to correct the total amount of glycine that was incorporated into protein. Regression: Specific activity = $2.20 \times \text{time} + 16.77$, $N=6$, $r^2=0.98$, $P<0.001$.

(F) Duplicate time-course assays to measure protein synthesis rates in 7-day-old larvae. For each synthesis rate, two time-course assays were conducted using different aliquots of larvae from the same larval culture (open and closed circles represents duplicate time-series assays). By comparison of slopes of the two different regressions shown, the rates were not statistically different ($P=0.06$). Data points were pooled to calculate a single rate of protein synthesis for larvae of this size and age (i.e., corresponding to one data point, as shown in Fig. 3C). Regression: Protein synthesis = $0.04 \times \text{time} + 0.48$, $N=12$, $r^2=0.92$, $P<0.001$.