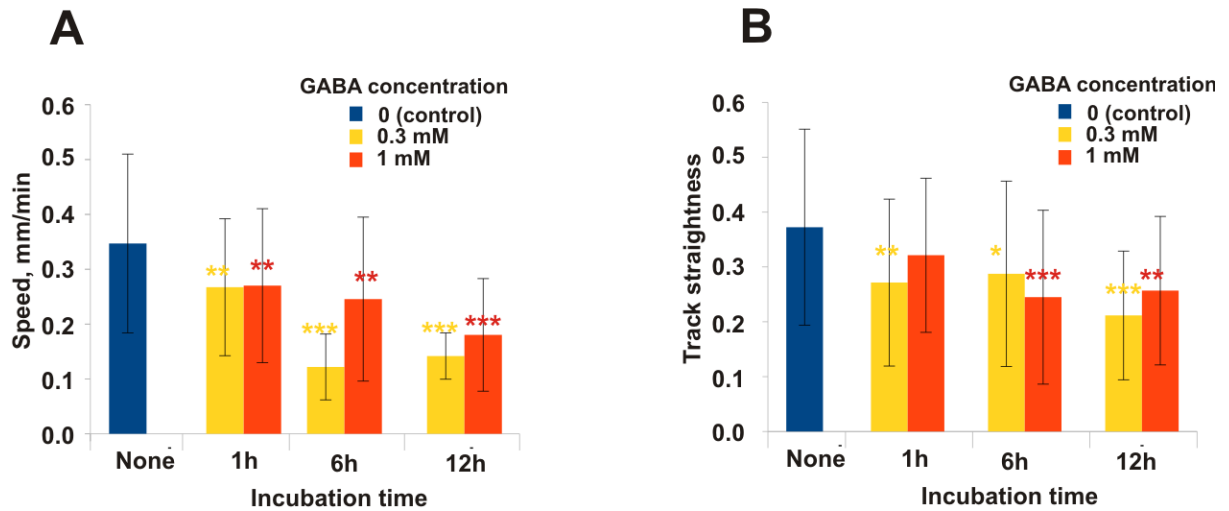


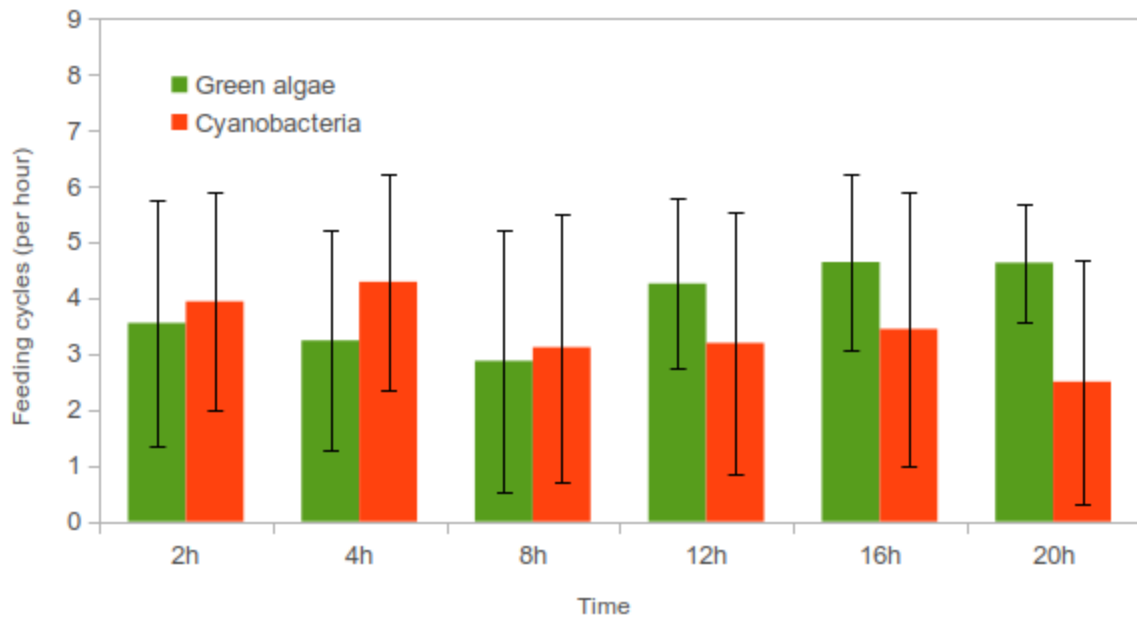
Supplemental Figures:

Glutamate, aspartate, GABA, glycine, and ATP integrate locomotion and feeding behaviors in placozoans

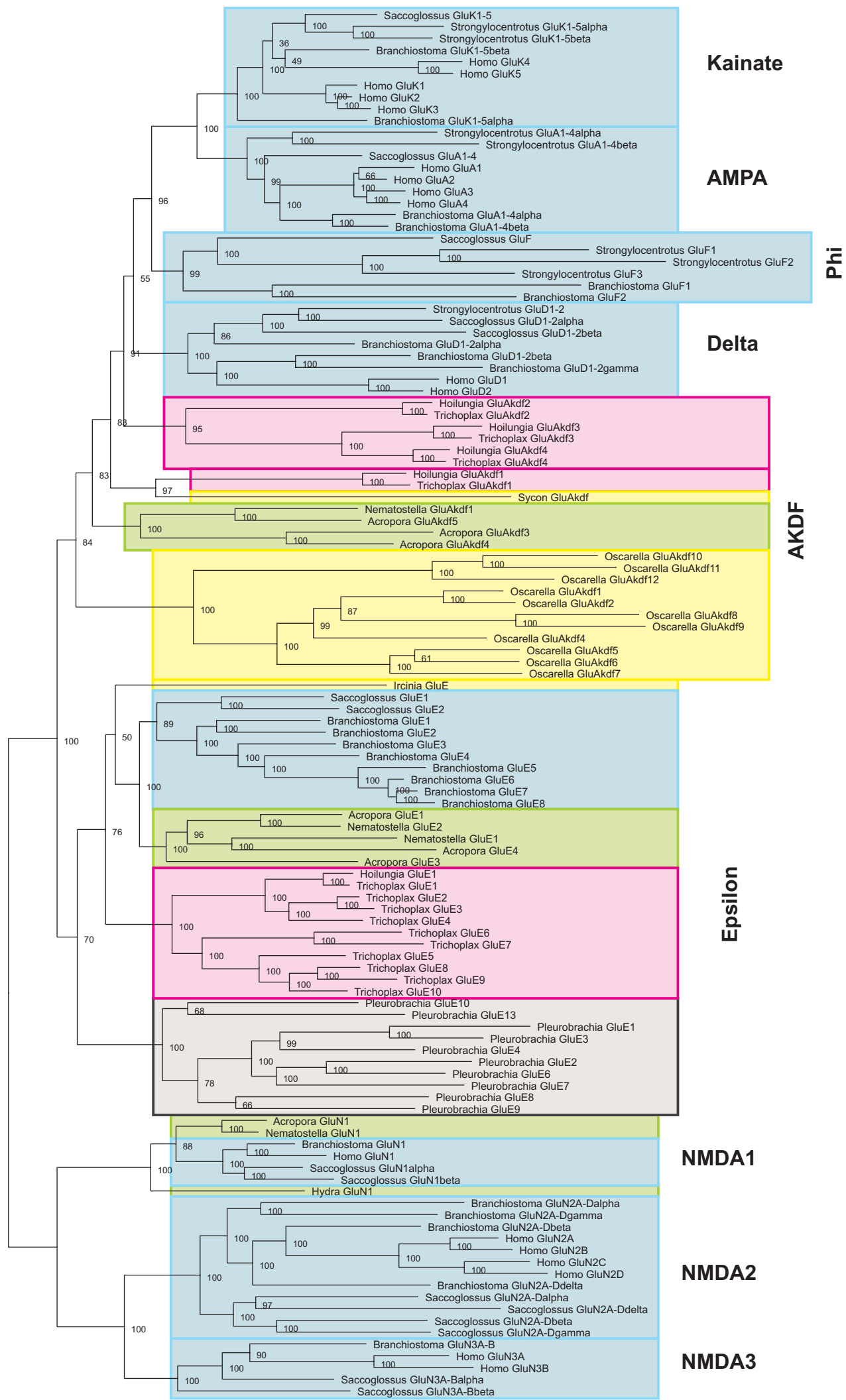
Mikhail A. Nikitin, Darja Y. Romanova, Simkha I. Borman, Leonid L. Moroz



Supplemental Figure 1. Effects of GABA on the locomotion of *Trichoplax* on a conditioned dish. **(A)** Animal locomotion on a clean dish is increased over time in control but slightly decreases during incubation with 0.3 or 1 mM GABA. Asterisks denote a significant difference in speed from the control (Welch t-test, *** - $p < 0.001$). N animals = 98, 50, 74, 29, 46, 69, 35, 44, and 72, respectively. **(B)**. Track straightness on a clean dish does not change over time in control but decreases during incubation with GABA (tracks become more convoluted). N animals are the same as in A.



Supplemental figure S2. Comparison of feeding dynamics on green algae *Tetraselmis* and cyanobacteria *Oscillatoria*. After 12 hours, feeding on cyanobacteria is less active than on green algae, and the standard deviation of feeding activity is higher on cyanobacteria. N animals = 40 for green algae and 48 for cyanobacteria.

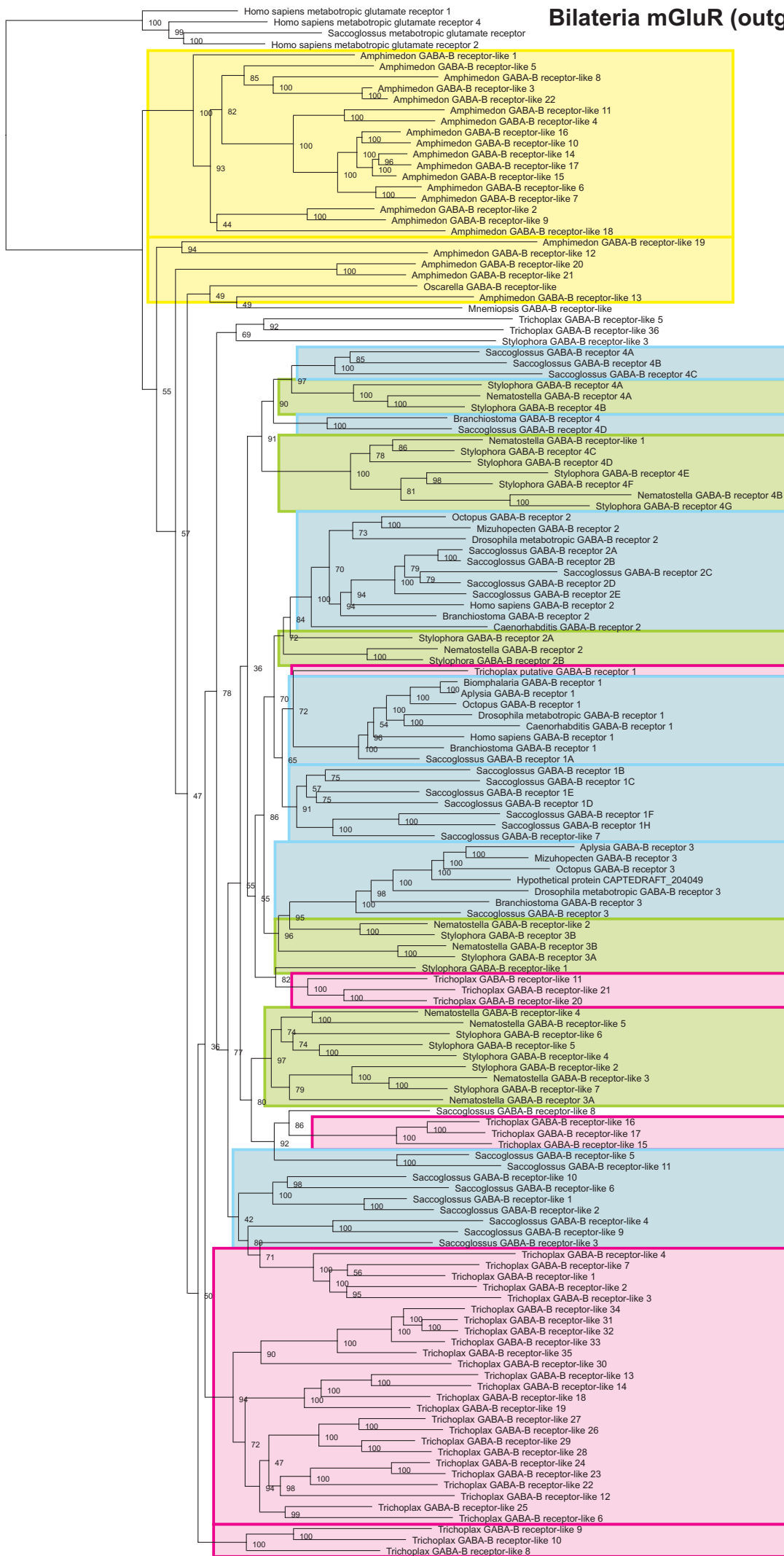


0.6

Placozoa
 Porifera
 Cnidaria
 Bilateria

Supplemental figure S3. Bayesian phylogenetic tree of ionotropic glutamate receptors. Numbers denote bayesian posterior probability near nodes. Analysis was run for 1.42 million generations, and convergence was checked with Tracer. Sequences were aligned using E-INS-i algorithm implemented in MAFFT online. Alignment consisted of 2472 positions, 1348 of them parsimony-informative.

Bilateria mGluR (outgroup)



**Deuterostomia
and Cnidaria
GABA-B R4**

**Bilateria
GABA-B R2**

**Bilateria
GABA-B R1**

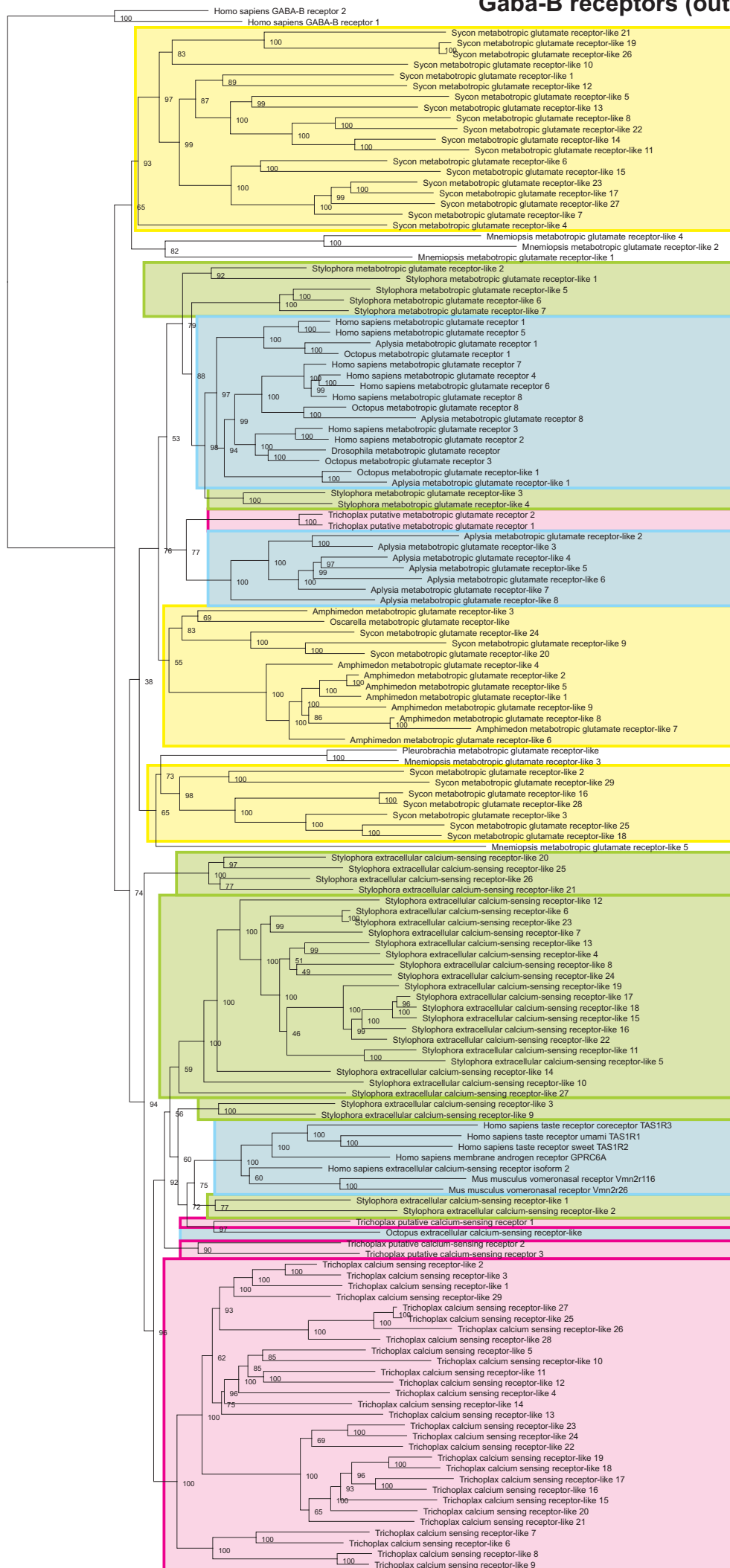
**Bilateria
GABA-B R3**

0.7

Placozoa
 Porifera
 Cnidaria
 Bilateria

Supplemental figure S4. Maximum Likelihood phylogenetic tree of metabotropic glutamate and related receptors. Numbers denote ultrafast bootstrap support near nodes. Sequences were aligned using E-INS-i algorithm implemented in MAFFT online. Alignment consisted of 4071 positions, 1464 of them parsimony-informative.

Gaba-B receptors (outgroup)



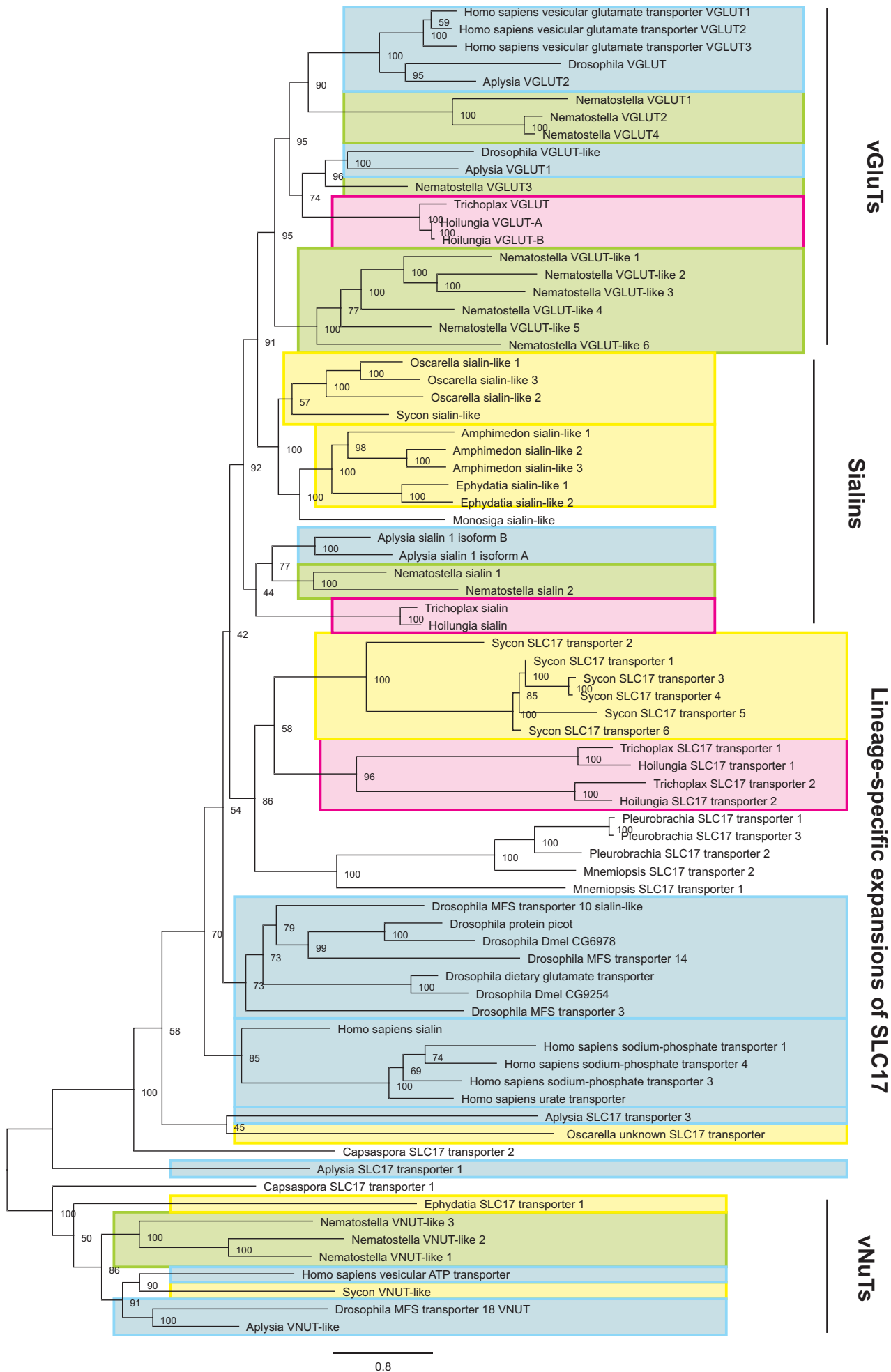
**Bilateria
mGluR**

**Bilateria
extracellular
calcium-sensing,
taste and
vomeronasal**

0.6

Placozoa
 Porifera
 Cnidaria
 Bilateria

Supplemental figure S5. Maximum Likelihood phylogenetic tree of metabotropic GABA receptors. Numbers denote ultrafast bootstrap support near nodes. Sequences were aligned using E-INS-i algorithm implemented in MAFFT online. Alignment consisted of 5312 positions, 1376 of them parsimony-informative.



Placozoa
 Porifera
 Cnidaria
 Bilateria

Supplemental figure S6. Bayesian phylogenetic tree of vesicular glutamate and related transporters (SLC17 family). Numbers denote bayesian posterior probability near nodes. Analysis was run for 10 million generations, and convergence was checked with Tracer. Sequences were aligned using E-INS-i algorithm implemented in MAFFT online. Alignment consisted of 1036 positions, 603 of them parsimony-informative.