

Figure 1 | CNS evolution and dorsoventral patterning. **a**, A nerve net is ancestral for Cnidaria and Bilateria. The neuroanatomical diversity hampers the reconstruction of the CNS evolution in Bilateria. **b**, A central argument for an ancestral medially condensed VNC for Bilateria is the

sensory structures and neural condensations in these species. However, the dorsoventral transcription factor *nkx6* does not co-localize with the motor neuron marker *ChAT* in the trunk of *M. stichopi* and *I. pulchra*, and the relation of *pax6*⁺ cells to this and another motor neuron marker (*Hb9*) is unclear in both species (Fig. 2b, e). Therefore, the diversity of neuroanatomies of Xenacoelomorpha contrasts with the

similar deployment of dorsoventral transcription factors in vertebrates, *Drosophila*, and the *P. dumerilii* larva. The staggered expression of these genes concurs with specific neuronal populations. D, dorsal; V, ventral; A, anterior; P, posterior; 5-HT, serotonin; ACh, acetylcholine.

more conserved deployment of ectodermal anteroposterior and BMP patterning systems. This, and the observation that disruption of BMP signalling does not affect CNS development (Extended Data Fig. 6), support the idea that the anti-neural role of the BMP pathway evolved after the Xenacoelomorpha–Nephrozoa split. Likewise, the expression of dorsoventral transcription factors unrelated to the distinct trunk

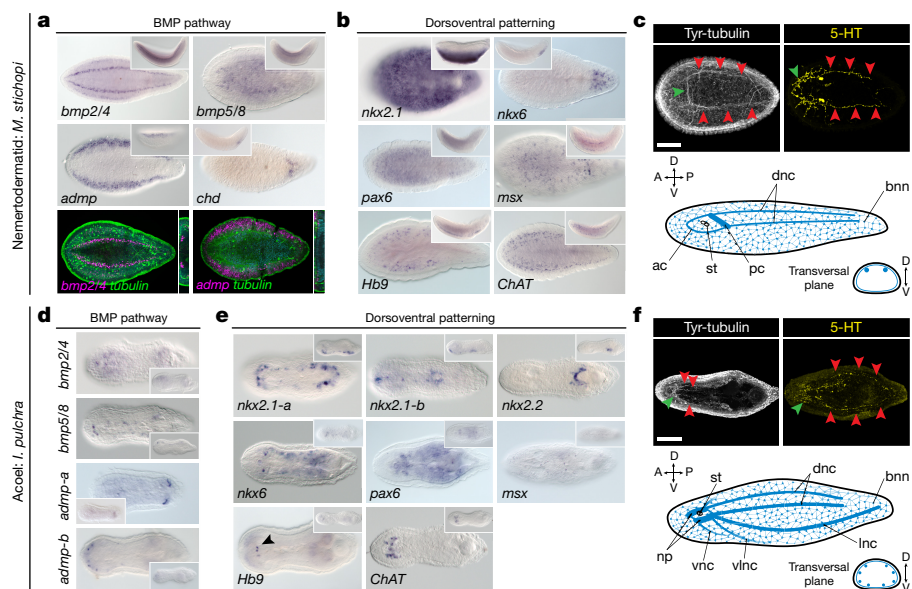


Figure 2 | Dorsoventral patterning in Xenacoelomorpha. **a**, The *bmp* ligands and *admp* are expressed dorsally; *chd* is expressed ventroposteriorly. **b**, Transcription factors *nkx2.1*, *nkx6*, and *msx* are expressed ventrally; *pax6* is expressed broadly; *Hb9* and *ChAT* are in the nerve cords. **c**, *M. stichopi* CNS (green arrowheads indicate the anterior commissures; red arrowheads indicate the nerve cords). Tyr-tubulin, tyrosinated tubulin. **d**, The *bmp* ligands are expressed dorsally; *admp-a* is expressed posteroventrally; *admp-b* is expressed anterolaterally. **e**, The

nkx2.1 paralogues and *nkx2.2* are expressed ventrally; *nkx6* is expressed laterally; *pax6* throughout the body; *msx* in isolated cells; *Hb9* and *ChAT* are in the brain. **f**, *I. pulchra* CNS (green arrowheads indicate the brain; red arrowheads indicate the nerve cords). Insets are lateral views. Abbreviations: ac, anterior commissure; bnn, basiepidermal nerve net; dnc, dorsal nerve cord; lnc, lateral nerve cord; np, neuropile; pc, posterior commissure; st, statocyst; vlnc, ventrolateral nerve cord; vnc, ventral nerve cord. Scale bars, 100 μ m.