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Dopamine-like immunoreactivity in sponge larvae

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ABSTRACT: Sponges differ from the majority of multicellular animals by lack of specialized neural cells. Although sponge genomes show the toolkits for metabolism of some neuroactive substances, only few genes of monoamines metabolism were found. We studied larvae of the freshwater sponge *Eunapius fragilis* to analyze the immunoreaction to one of the monoamines – dopamine (DA). We found dopamine-like immunoreactivity in structures located under every flagellum in the larval epithelial cells. Double labeling with anti-DA and anti-58K Golgi protein antibodies, confocal microscopy with improved signal-to-noise ratio and super-resolution (Airyscan), and ultrastructural electron microscopy analysis revealed that the DA-like-immunopositive structures are closely associated with the Golgi apparatus. A similar pattern of immunolabeling was reported in the blastulae of sea urchins, whose ciliary activity is known to be affected by monoamines. Our finding of DA-like immunoreactive structures in sponge ciliated cells provide morphological basis for considering monoamines as potential intracellular regulators of flagellar/ciliary activity.
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KEY WORDS: monoamines, dopamine, Porifera, freshwater sponge, flagella/cilium.

Дофамин-подобная иммунореактивность у личинок губок

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РЕЗЮМЕ: Губки отличаются от большинства многоклеточных животных отсутствием специализированных нервных клеток. Несмотря на то, что в изученных геномах губок обнаружен инструментарий для метаболизма ряда нейроактивных веществ, для метаболизма моноаминов имеются не все необходимые гены. Мы провели иммунохимическое маркирование личинок пресноводной губки *Eunapius fragilis* для визуализации одного из моноаминов — дофамина (ДА). Дофамин-подобная иммунопозитивная реакция была выявлена в структурах, расположенных под жгутиком в каждой эпителиальной клетке личинки. Двойное маркирование с антителами к ДА и 58К протеину аппарата Гольджи, конфокальная микроскопия с улучшенным отношением сигнал/шум и суперразрешением (Airyscan), а также ультраструктурный анализ с использованием электронной микроскопии, показали, что обнаруженные

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ДА-подобные иммунопозитивные структуры ассоциированы с аппаратом Гольджи. Сходный паттерн иммунореактивности был показан ранее на бластулах морского ежа, для которых известно влияние моноаминов на активность локомоторных ресничек. Обнаруженные нами ДА-подобные иммунопозитивные структуры, расположенные под жгутиками клеток личинок *E. fragilis*, позволяют предполагать участие внутриклеточных моноаминов в регуляции жгутиковой/ресничной активности у губок.

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КЛЮЧЕВЫЕ СЛОВА: моноамины, дофамин, Porifera, пресноводные губки, жгутик/ресничка.

Introduction

Monoamines, such as serotonin (5-HT) and dopamine (DA), have a wide range of signal activity in diverse organisms. Besides multicellular animals, monoamines were found in prokaryotes (Hsu *et al.,* 1986; Shahkolahi, Donahue, 1993; Strakhovskaya *et al*., 1993; Lyte, Ernst, 1992; 1993; Oleskin *et al*., 1998; Tsavkelova *et al*., 2000; Clarke *et al*., 2006; Shishov *et al*., 2009), protists (McGowan *et al*., 1985; Csaba 1993), plants (Roshchina, 1991, 2001; 2010; Murch, 2006; Wan *et al*., 2018), and fungi (Malikina *et al*., 2010; Roshchina, 2010). Taken together, these data suggest that monoamines are present in most living organisms and apparently underlay regulation of conservative vital functions. This assumption is also supported by the fact that these substances are functionally active from the earliest stages of animal development starting from the oocyte, cleavage, and cell differentiation (Buznikov, 2007). Thus, the function of monoamines is far beyond the participation in signal transmission between specialized cells like differentiated neurons within the nervous system.

In Bilateria, the crucial biological role of monoamines is well-known, while data on other groups of multicellular organisms are scarce. The most evidences for presence of serotonin and catecholamines concerns cnidarian (Mathias *et al.*, 1960; Welsh, 1960, 1968; Wood, Lentz, 1964; Kline, Weissbach, 1965; Carlyle, 1969; Carlberg *et al*., 1984; Kolber, Martin, 1988; Anctil, 1989; Chung *et al*., 1989; Chung, Spencer, 1991; Carlberg, 1992; MacCauley, 1997; Dergham, Anctil, 1998; Westfall *et al*., 2000; Zega *et al*., 2003; Mayorova, Kosevich, 2013, Moeller *et al*., 2019). Despite the fact that their genomes lack the genes coding monoamine synthetic enzymes tyrosine hydroxylase and tryptophan hydroxylase (Westfall *et al*., 2000; Anctil *et al*., 2002; Anctil, 2009; Moroz, Kohn, 2015; Moroz *et al*., 2021), monoamines exert various physiological effects in cnidarians. In particular, their role in metamorphosis (Edwards *et al*., 1987; MacCauley, 1997; Zega *et al*., 2003; Moeller *et al*., 2019), rhythmic contraction (Anctil, 1989, Anctil *et al*., 1991; Tsang *et al*., 1997), feeding (Hanai and Kitajima, 1984), regeneration (Lenicque and Feral, 1977), and bioluminescence (Anctil *et al*., 1982; Awad, Anctil, 1993) were shown. Presence in neurons (Kolberg, Martin, 1988; Umbriaco *et al*., 1990; Pani *et al*., 1995; Anctil *et al*., 2002; Westfall *et al*., 2005), biosynthesis and catabolism (Pani, Anctil, 1994), release by exocytosis (Gillis, Anctil, 2001), and inactivation (Kolberg, Martin, 1988) of various monoamines were noted in variety of cnidarians. There are also some data on receptor activity (Hanai, Kitajima, 1984; Awad, Anctil, 1993; Anctil, Bouchard, 2004; Kass-Simon, Pierobon, 2007; Zega *et al*., 2007; Chen *et al*., 2017).

Sponges (phylum Porifera) represent a more basal group than cnidarian and completely lack homologues of monoamine receptors (Riesgo *et al*., 2014; Moroz, 2021). The other components

of the classical monoaminergic system (such as synthesis and degradation enzymes, transporters) do not form a coherent group but are rather scattered across the transcriptomes of different sponge species (Riesgo *et al*., 2014). Nevertheless, it is known that sponges react to the application of serotonin, dopamine, and epinephrine (Ellwanger *et al*., 2004; Ellwanger, Nickel, 2006). In addition, serotonin was visualized in the larvae of the sponge *Tedania ignis* (Duchassaing et Michelotti, 1864) by immunochemical labeling (Weyrer *et al*., 1999). Histochemical method revealed monoamine oxidase, serotonin, epinephrine in the calcareous sponge *Sycon*, and the staining was even blocked by inhibitors of their metabolism (Lentz, 1966), while presented histochemical work needs repetition.

In the current work, we present an evidence for the presence of specific DA-like-immunopositive structures in the sponge larvae cells and suggest their correspondence to the intracellular organelles (cilium/flagellum and Golgi apparatus).

Material and methods

Larvae of the freshwater sponge *Eunapius fragilis* (Leidy, 1851) were collected in the Moscow Canal in 15 June 2018 – 15 July 2021. The presence of larvae in sponges was determined visually, then parts of the sponges were separated from the substrate and placed in a container with fresh water. After a few hours, larvae left the tissues of the mother's body and were collected using a Pasteur glass pipette.

To fix larvae, we used a 4% paraformaldehyde solution (PFA) prepared in the phosphate buffer (PBS, pH = 7.4). The larvae were fixed for 4 h at room temperature, then washed three times in PBS and immersed for a day in a PBS-based solution with 1% Triton X-100, antibodies to DA (1: 150, rabbit polyclonal, Enzo Life Sciences BML-DA1140), acetylated alpha-tubulin (1: 3000, mouse monoclonal, *Sigma* Aldrich *T6793*), and Golgi marker (formiminotransferase cyclodeaminase, 58K Golgi protein, Sigma, G2707) (1: 100, mouse monoclonal). After that, the samples were washed three times in PBS and immersed for a day in a PBS-based solution with 0.1% Triton X-100 and secondary antibodies (Goat anti-Rabbit Alexa 488 and Goat anti-Mouse Alexa 555, ThermoFisher), and Hoechst nuclear dye (Sigma). If necessary, preparations were stained with phalloidin-Alexa 488 (1: 600, ThermoFisher) to visualize cell borders. Finally, the samples were washed three times in PBS and transferred to 80% glycerol in PBS. The specific character of antibody binding was checked by the reaction lacking primary antibodies and other rabbit antibodies.

Confocal scanning microscope Zeiss 880 (Carl Zeiss, Germany) was used for the detailed analysis of the preparations. Stacks of optical sections taken with x40 objectives and 0.3 ìm intervals were processed by Image J (NIH, USA) to obtain two-dimensional images. The optimal number of stacks was selected to demonstrate the structures of interest. We applied the Airyscan mode of Zeiss 880 scanning microscope to improve signal-to-noise ratio and to get super-resolution images.

Ultrathin sections were gained and processed as described in Sokolova *et al*., 2019.

Results

The larva of *E. fragilis* is a free-swimming parenchymella with large cavity in the anterior part (Fig. 1A). It is covered by dense layer of epithelial monociliated cells underlined with amoeboid cells (Fig. 1B). Epithelial cells have pear-shaped nuclei with prominent apical beak (Fig. 1C).

The immunofluorescence revealed a single DA-like immunopositive granule located asymmetrically within the cell body underneath the cilia and adjacent to nuclei (Fig. 1C–E). Confocal images with improved signal-to-noise ratio and super-resolution (Airyscan mode) allowed to visualize DA-like-positive specific structure and position adjacent to each nucleus of epithelial cells (Fig. 2A, B). Note, that each structures contain three-four parts combined in circle (~1 μm in diameter) (Fig. 2C, E), and DA-like positive circles look like thick stick (~1 μm

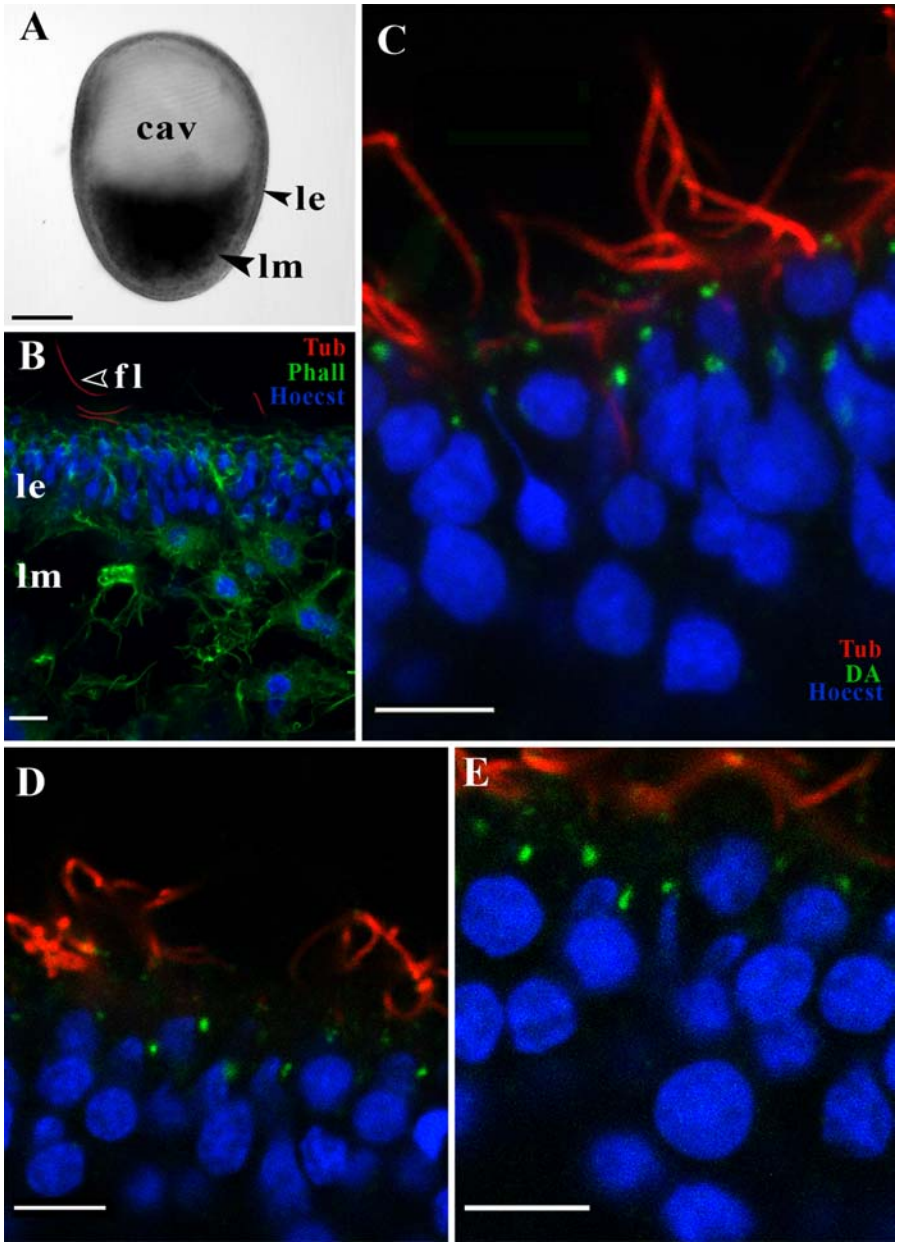

Fig. 1. Larva of *Eunapius fragilis* and its peripheral cells. A — larva, general view (light microscopy); B — peripheral fragment of larva: larval epithelium underlined with amebocytes (confocal microscopy); C–E — high magnification of the epithelial cells labeled with anti-DA antibodies.
Abbreviations: cav — cavity; le — larval epithelium; lm — larval mesohyl; Phall — phalloidin, Tub — tubulin-positive label, DA — dopamine-positive label. Scale bars: A — 100 μm, B — 10 μm, C–E — 5 μm.
Рис. 1. Личинка *Eunapius fragilis* и ее периферические клетки. A — общий вид личинки (световая микроскопия); B — фрагмент периферического участки личинки: личиночный эпителий и лежащие под ним амебоциты (конфокальная микроскопия); C–E — большое увеличение эпителиального слоя личинки, маркирование антителами против ДА.
Обозначения: cav — полость; le —личиночный эпителий; lm —личиночный мезохил; Phall — фаллоидин, Tub — тубулин-позитивная метка, DA — дофамин-позитивная метка. Масштаб: A — 100 μм, B — 10 μм, C–E — 5 μм.

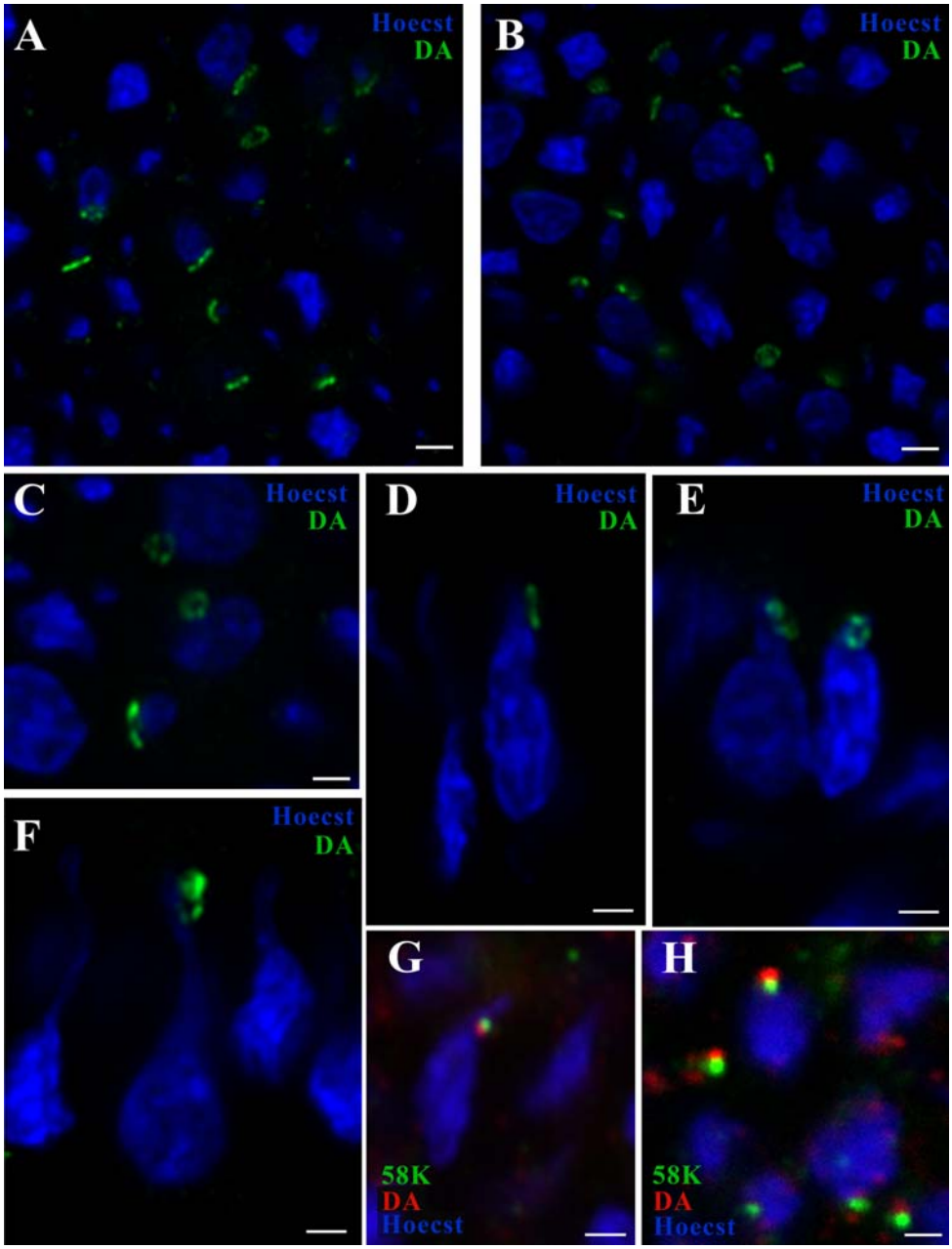

Fig. 2. Immunolabeling of epithelial cells. A–C — Airyscan images of transversal sections, anti-DA labeling (green); D–F — Airyscan images of longitudinal sections, anti-DA labeling (green); G–H — anti-DA (red) and anti-Golgi 58K protein (green) double labeling.
Scale bars: A–C — 1 μm; D–F — 1 μm; G — 2 μm; H — 1 μm.
Рис. 2. Иммунохимическое маркирование эпителиальных клеток. A–C— поперечный срез, Airyscan изображение, маркирование антителами к ДА (зеленый); D–F — продольный срез, Airyscan изображение, маркирование антителами к ДА (зеленый); G–H — двойное мечение: против ДА (красный) и 58K протеина комплекса Гольджи (зеленый).
Масштаб: A–C — 1 μм; D–F — 1 μм; G — 2 μм; H — 1 μм.

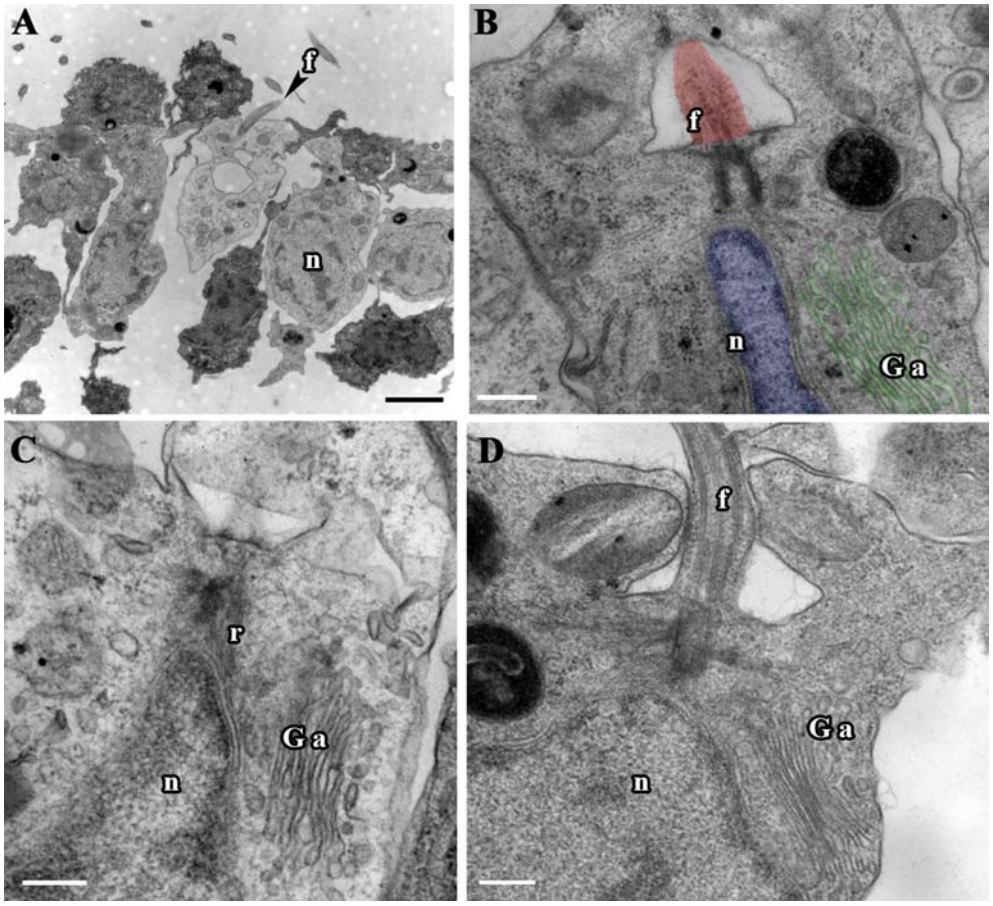

Fig. 3. Ultrastructure of epithelial cells. A — section of the epithelium; B–D — apical parts of the flagellated cells. Structures on B are colored in accordance to the colors of immune labels on Fig. 1: red — flagellum, blue — nucleus, green — Golgi apparatus.

Abbreviations: f — flagellum; Ga — Golgi apparatus; n — nucleus; r — rootlets. Scale bars: A — 1 μm, C–D — 0.2 μm.

Рис. 3. Ультраструктура эпителиальных клеток. A — срез через эпителий; B–D — апикальная часть жгутиковой клетки. Структуры на B окрашены в соответствии с цветами иммунохимического маркирования на Рис. 1.

Обозначения: f — жгутик; Ga — аппарат Гольджи; n — ядро; r — корешки. Масштаб: A — 1 μм, C–D — 0,2 μм.

long) from the side (Fig. 2A, C, D, E). Only single DA-like-positive structure locates in one cell, and this structure is adjacent to the nuclei apical beak (Fig. 2D, E, F).

Double staining with DA antibodies and the 58K Golgi protein antibodies (convenient and widely used Golgi apparatus marker) demonstrated close location of both markers in the specific position adjacent to the nuclei apical beak (Fig. 2G, H). Note the absence of full colocalization between markers (Fig. 2H), and basal location of DA-like positive structure relative to the 58K Golgi protein-positive mark (Fig. 2G).

The ultrastructural analysis demonstrated characteristic structures of *E. fragilis* larval epithelial cells. Single flagellum emerged from the deep pit at the apical pole of the cell (Fig. 3A, B, D). The kinetid (flagellar apparatus) was connected with the nucleus via rootlets (Fig.

3C). Golgi apparatus demonstrated asymmetrical positioning on one side from the nuclear beak (Fig. 3B–D). Note the size of Golgi apparatus ~1 μm (Fig. 3B, C, D). The described arrangement of the flagellum, nucleus, and Golgi apparatus were observed in every ciliated cell of larval epithelium.

Discussion

The Golgi apparatus located aside the ciliary/flagellar rootlets is an ordinary feature of monociliated cells in diverse taxa (e.g. Nerrevang, Wingstrand, 1970). This organelle is shown to be a site where the flagellar scaffold proteins are produced, maturated and modified (Deane *et al.*, 2001). It also participates in the intraflagellar transport functioning and correct cilia assembly (Follit *et al*., 2006; Carvalho-Santos *et al*., 2011). In sponges, the Golgi apparatus often adjoins the flagellar rootlets, even when the nucleus is located away from the kinetid (Pozdnyakov *et al*., 2018, 2020; Sokolova *et al*., 2019, 2020).

In larval epithelial cells of *Eunapius fragilis,* the Golgi apparatus forms a complex with the nucleus and kinetid rootlets. Juxtaposition of immunochemical labeling and ultrastructural analysis demonstrates the spatial correspondence of the DA-like immunoreactive structures and Golgi apparatus, which is confirmed by specific anti-Golgi labeling. Interestingly, some components of monoaminergic system, such as monoamine synthesis enzymes (tryptophane hydroxylase, tyrosine hydroxylase and dopamine-â-hydroxylase) are shown to be associated with the endoplasmic reticulum, microtubules and Golgi apparatus in the 5-HT- and DA-containing central neurons of mammals (Iijima, Awazi, 1973; Pickel *et al.*, 1975, 1976).

A similar pattern of DA- and 5-HT-positive immunolabeling associated with locomotory cilia was observed in the larvae of sea urchins (Katow *et al*., 2010; Obukhova *et al*., 2014, 2015). The monoamine-positive structures occurred in association with gamma-tubulin at the base of the cilium at sea urchin blastula stage (Katow *et al*., 2010). Their correspondence to the Golgi apparatuses was suggested by ultrastructural analysis (Obukhova *et al*., 2014, 2015). The experimental survey demonstrated that in the sea urchin larvae the Golgi-located monoamines are involved in regulation of cilia growth and regeneration (Obukhova *et al*., 2014, 2015). It is known that serotonin and dopamine are active molecules affecting ciliary beating frequency in invertebrate larvae (Marinković *et al*., 2019), including sea urchins (Wada *et al*., 1997; Katow *et al*., 2010) and freshwater gastropods (Goldberg *et al*., 2011). Probably, the monoamine-dependent ciliary regulation is a universal mechanism within invertebrates. We suggested that despite the absence of complete genetic tools for the monoaminergic system, sponge may utilize monoamines for the same purpose. The observed dopamine-like-positive immunoreactivity in sponge larvae supports this suggestion which, of cause, needs to be proved experimentally.

Our work presents the first evidence for intracellular DA-like-immunopositive structures in sponges, the multicellular organisms lacking nerve cells and monoamine receptors. Further studies of monoamines in sponges are very promising to uncover the functions of monoamines as ancient intracellular regulators of cell activity.

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References

Anctil M., Boulay D., Lariviére L. 1982. Monoaminergic mechanisms associated with control of luminescence and contractile activities in the coelenterate *Renilla köllikeri* // Journal of Experimental Zoology. Vol.223. P.11–24.

Anctil M. 1989. Modulation of a rhythmic activity by serotonin via cyclic AMP in the coelenterate *Renilla köllikeri* // Journal of Comparative Physiology. Part B. Vol.159. P.491–500.

Anctil M., Pani A.K., Ali M.A. 1991. Modulation of rhythmic contractions by melatonin via cyclic GMP in the coelenterate *Renilla koellikeri* // Journal of Comparative Physiology B. Vol.161. P.569–575.

Anctil M. 2009. Chemical transmission in the sea anemone *Nematostella vectensis*: a genomic perspective // Comparative Biochemistry and Physiology. Part D: Genomics and Proteomics. Vol. 4. P.268–289. doi:10.1016/j.cbd.2009.07.001.

Anctil M., Hurtubise P., Gillis M.-A. 2002. Tyrosine hydroxylase and dopamine-â-hydroxylase immunoreactivities in the cnidarian *Renilla koellikeri* // Cell Tissue Research. Vol.310. P.109–117.

Anctil M., Bouchard C. 2004. Biogenic amine receptors in the sea pansy: activity, molecular structure, and physiological significance // Hydrobiologia. Vol.530/531. P.35–40.

Awad E., Anctil M. 1993. Identification of â-like adrenoceptors associated with bioluminescence in the sea pansy, *Renilla koellikeri* // Journal of Experimental Biology. Vol.177. P.181–200.

Buznikov, G. A. 2007. Preneural transmitters as regulators of embryogenesis. Current state of problem // Russian Journal of Developmental Biology. Vol.38. P.213–220.

Carlyle R. 1969. The occurrence of catecholamines in the sea anemone *Actinia equina* // British Journal of Pharmacology. Vol.36. P.182.

Carlberg M. 1992. Localization of dopamine in the freshwater hydrozoan *Hydra attenuata* // Cell Tissue Research. Vol.270. P.601–607.

Carlberg M., Jergil B., Lindbladh C., Rosengren E. 1984. Enzymatic 5-hydroxylation of l-dopa by a tyrosinase isolated from the sea anemone *Metridium senile* // General Pharmacology: The Vascular System. Vol.15. P.301–307.

Carvalho-Santos Z., Azimzadeh J., Pereira-Leal J. B., Bettencourt-Dias M. 2011. Tracing the origins of centrioles, cilia, and flagella // Journal of Cell Biology. Vol.194. P.165–175.

Chen P., Chen P., Li T., Shen Q., Yan D.-F., Zhang L., Chen X., Li Y, Zhao W. 2017. Two dopamine D2-like receptor genes from the silkworm (*Bombyx mori*) and their evolutionary history in metazoan // Scientific Reports. Vol.7. P.1–12. doi:10.1038/s41598-017-07055-5.

Chung J.-M., Spencer A.N., Gahm K.H. 1989. Dopamine in tissues of the hydrozoan jellyfish *Polyorchis penicillatus* as revealed by HPLC and GC/MS // Journal of Comparative Physiology. Part B. Vol.159. P.173–181.

Chung J.-M., Spencer A.N. 1991. Dopamine as a neuroactive substance in the jellyfish *Polyorchis penicillatus* // Journal of Experimental Biology. Vol.156. P.433–451.

Clarke M.B., Hughes D.T., Zhu C., Boedeker E.C., Sperandio V. 2006. The QseC sensor kinase: a bacterial adrenergic receptor // Proceedings of the National Academy of Sciences of the USA. Vol.103. P.10420–10425. doi: 10.1073/pnas.0604343103

Csaba G. 1993. Presence in and effect of pineal indoleamines at very low level of phylogeny // Experientia. Vol.49. P.627–634.

Deane J.A., Cole D.G., Seeley E.S., Diener D.R., Rosenbaum J.L. 2001. Localization of intraflagellar transport protein IFT52 identifies basal body transitional fibers as the docking site for IFT particles // Current Biology. Vol.11. P.1586–1590. doi: 10.1016/s0960-9822(01)00484-5.

Dergham P., Anctil, M. 1998. Distribution of serotonin uptake and binding sites in the cnidarian *Renilla koellikeri*: an autoradiographic study // Tissue and Cell. Vol.30. P.205–215.

Edwards N.C., Thomas M.B., Long B.A., Amyotte S.J. 1987. Catecholamines induce metamorphosis in the hydrozoan *Halocordyle disticha* but not in *Hydractinia echinate* // Roux's Archives of Developmental Biology. Vol.196. P.381–384.

Ellwanger K., Nickel M. 2006. Neuroactive substances specifically modulate rhythmic body contractions in the nerveless metazoon *Tethya wilhelma* (Demospongiae, Porifera) // Frontiers in Zoology. Vol.3. P.1–14.

Ellwanger K., Brümmer F., Nickel M. 2004. Glutamate, GABA and serotonin induce contractions in the sponge *Tethya wilhelma* (Porifera: Demospongiae) // Kinzelbach Â.R. (ed.). Jahrestagung der Deutschen Zoologischen Gesellschaft. Rostock: Abstractband. P.157.

Elliott G.R., Leys S. 2007. Coordinated contractions effectively expel water from the aquiferous system of a freshwater sponge // Journal of Experimental Biology. Vol.210. P.3736–3748.

Follit J.A., Tuft R.A., Fogarty K.E., Pazour G.J. 2006. The intraflagellar transport protein IFT20 is associated with the Golgi complex and is required for cilia assembly // Molecular biology of the cell. Vol.17. P.3781–3792.

Gillis M.-A., Anctil M. 2001. Monoamine release by neurons of a primitive nervous system: an amperometric study // Journal of Neurochemistry. Vol.76. P.1774–1784.

Goldberg J.I., Rich D.R., Muruganathan S.P., Liu M.B., Pon J.R., Tam R., Diefenbach T.J., Kuang S. 2011. Identification and evolutionary implications of neurotransmitter-ciliary interactions underlying the behavioral response to hypoxia in *Lymnaea stagnalis* embryos // The Journal of Experimental Biology. Vol.214. P.2660–2670.

Hanai K., Kitajima M. 1984. Two types of surface amine receptors modulating the feeding response in *Hydra japonica*: the depressing action of dopamine and related amines // Chemical Senses. Vol.9. P.355–367.

Hsu S.C., Johansson K., Donahue M. 1986. The bacterial flora of the intestine of *Ascaris suum* and 5-hydroxytryptamine production // Journal of Parasitology. Vol.72. P.545–549.

Iijima K., Awazi N. 1973. Histochemical studies on the morphology of the Golgi apparatus and its relation-

ship to catecholamine biosynthesis in the locus coeruleus of the rat // Zeitschrift für Zellforschung und Mikroskopische Anatomie. Vol.136. P.329–348.

Kass-Simon G., Pierobon P. 2007. Cnidarian chemical neurotransmission, an updated overview // Comparative Biochemistry and Physiology. Part A: Molecular, Integrative Physiology. Vol.146. P.9–25. doi:10.1016/j.cbpa.2006.09.008.

Katow H., Suyemitsu T., Ooka S., Yaguchi J., Jin-Nai T., *et al.* 2010. Development of a dopaminergic system in sea urchin embryos and larvae // Journal of Experimental Biology. Vol.213. P.2808–2819.

Kline E.S., Weissbach H. 1965. Hydroxyindoleamines in the nematocysts of *Hydra littoralis* // Life Sciences. Vol.4. P.63–67. doi:10.1016/0024-3205(65)90034-2

Kolberg K.J., Martin V. 1988. Morphological, cytochemical and neuropharmacological evidence for the presence of catecholamines in hydrozoan planulae // Development. Vol.103. P.249–258.

Lenicque P.M., Féral J.P. 1977. Effects of biogenic amines on the regeneration of small pieces of the pedal disc of the sea anemone *Metridium senile* (Linnaeus) // Comparative Biochemistry and Physiology Part C: Comparative Pharmacology. Vol.57. P.91–93.

Lentz T.L. 1966. Histochemical localization of neurohumors in a sponge // Journal of Experimental Zoology. Vol.162. P.171–179.

Lyte M., Ernst S. 1993. Alpha- and beta-adrenergic receptor involvement in catecholamine-induced growth of gram-negative bacteria // Biochemical and Biophysical Research Communications. Vol.190. P.447–452. doi: 10.1006/bbrc.1993.1068

Lyte M., Ernst S. 1992. Catecholamine induced growth of gram-negative bacteria // Life Sciences. Vol.50. P.203–212. doi: 10.1016/0024-3205(92)90273-R

McCauley D.W. 1997. Serotonin plays an early role in the metamorphosis of the hydrozoan *Phialidium gregarium* // Developmental biology. Vol.190. P.229–240.

Malikina K.D., Shishov V.A., Chuvelev D.I., Kudrin V.S., Oleskin A.V. 2010. Regulatory role of monoamine neurotransmitters in *Saccharomyces cerevisiae* cells // Applied Biochemistry and Microbiology. Vol.46. P.620–625.

Marinković M., Berger J., Jékely G. 2019. Neuronal coordination of motile cilia in locomotion and feeding // Philosophical Transactions of the Royal Society B. Vol.375. 20190165.

Mathias A.P., Ross D., Schachter M. 1960. The distribution 5-hydroxytryptamine, tetramethyammonium, homarine, and other substances in sea anemones // Journal of Physiology. Vol.151. P.296–311. doi:10.1113/jphysiol.1960.sp006439.

Mayorova T.D., Kosevich I. 2013. Serotonin-immunoreactive neural system and contractile system in the hydroid Cladonema (Cnidaria, Hydrozoa) // Invertebrate Neuroscience. Vol.13. P.99–106. doi:10.1007/s10158-013-0152-2

McGowan K., Guerina V., Wicks J., Donowitz M. 1985. Secretory hormones of *Entamoeba histolytica* // Ciba Foundation symposium. Vol.112. P.139–154.

Moeller M., Nietzer S., Schupp P.J. 2019. Neuroactive compounds induce larval settlement in the scleractinian coral *Leptastrea purpurea* // Scientific reports. Vol.9. P.1–9.

Moroz L.L., Kohn A.B. 2015. Unbiased view of synaptic and neuronal gene complement in Ctenophores: are there pan-neuronal and pan-synaptic genes across Metazoa? // Integrative and Comparative Biology. Vol.55. P.1028–1049. doi:10.1093/icb/icv104

Moroz L.L., Romanova D.Y., Kohn A.B. 2021. Neural versus alternative integrative systems: molecular insights into origins of neurotransmitters // Philosophical Transactions of the Royal Society B. Vol.376. P.20190762. doi:10.1098/rstb.2019.0762

Murch S.J. 2006. Neurotransmitters, neuroregulators and neurotoxins in plants // Baluska Â.F., Mancuso S., Volkmann D. (eds.). Communication in Plants – Neuronal Aspects of Plant Life. Berlin: Springer. P.137–151.

Nerrevang A., Wingstrand K.G. 1970. On the Occurrence and Structure of Choanocyte-like Cells in Some Echinoderms // Acta Zoologica. Vol.51. 249–270. doi:10.1111/j.1463-6395.1970.tb00436.x.

Obukhova A., Ivashkin E., Khabarova M., Voronezshkaya E. 2014. Novel non-neuronal cilia-associated monoamine-containing structure in sea urchin larvae // International congress on invertebrate morphology (ICIM 3), Berlin, Germany. P.243.

Obukhova A., Ivashkin E., Khabarova M., Yushin V.V., Voronezshkaya E.E. 2015. Catecholaminergic control of swimming behavior in sea urchin larvae // 13th International Symposium of Invertebrate Neurobiology (ISIN), Tihany, Hungary. P.54.

Oleskin A., Kirovskaya T., Botvinko I., Lysak L. 1998. Effects of serotonin (5-hydroxytryptamine) on the growth and differentiation of microorganisms // Microbiologia. Vol.67. P.305–312.

Pani A., Anctil M., Umbriaco D. 1995. Neuronal localization and evoked release of norepinephrine in the cnidarian *Renilla koellikeri* // Journal of Experimental Zoology. Vol.272. P.1–12.

Pani A.K., Anctil M. 1994. Evidence for biosynthesis and catabolism of monoamines in the sea pansy *Renilla koellikeri* (Cnidaria) // Neurochemistry International. Vol.25. P.465–474.

Pickel V.M., Joh T.H., Field P.M., Becker C.G., Reis D.J. 1975. Cellular localization of tyrosine hydroxylase by immunohistochemistry // Journal of Histochemistry & Cytochemistry. Vol.23. P.1–12.

Pickel V.M., Joh T.H., Reis D.J. 1976. Monoamine-synthesizing enzymes in central dopaminergic, noradrenergic and serotonergic neurons. Immunocytochemical localization by light and electron microscopy // Journal of Histochemistry & Cytochemistry. Vol.24. P.792–792.

Pozdnyakov I.R., Sokolova A.M., Ereskovsky A.V., Karpov S.A. 2018. Kinetid structure in sponge choanocytes of Spongillida in the light of evolutionary relationships within Demospongiae // Zoological Journal of the Linnean Society. Vol.184. P.255–272.

Pozdnyakov I.R., Sokolova A.M., Ereskovsky A.V., Karpov S.A. 2020. The kinetid structure of two oscarellid sponges (Class Homoscleromorpha) unveils plesiomorphies in kinetids of Homoscleromorpha–Calcarea lineage // Invertebrate Biology. Vol.139. P.e12299.

Riesgo A., Farrar N., Windsor P., Giribet G., Leys S. 2014. The analysis of eight transcriptomes from all poriferan classes reveals surprising genetic complexity in sponges // Molecular biology and evolution. Vol.31. P.1102–1120.

Roshchina V. 1991. [Biomediators in plants. Acetylcholine and biogenic amines]. Pushchino: Biologicheskiy Tsentr AN SSSR. [In Russian]

Roshchina V. 2001. Neurotransmitters in plant life. Plymouth: Science Publ.

Roshchina V. 2010. Evolutionary considerations of neurotransmitters in microbial, plant, and animal cells // M. Lyte, P.P.E. Freestone (eds.). Microbial endocrinology. Springer Science+Business Media, LLC P.17–52.

Shahkolahi A.M., Donahue M.J. 1993 Bacterial flora, a possible source of serotonin in the intestine of adult female *Ascaris suum* // Journal of Parasitology. Vol.79. P.17–22.

Shishov V.A., Kirovskaya T., Kudrin V., Oleskin A. 2009. Amine neuromediators, their precursors, and oxidation products in the culture of *Escherichia coli* K-12 // Applied biochemistry and microbiology. Vol.45. P.494–497.

Sokolova A.M., Pozdnyakov I.R., Ereskovsky A.V., Karpov S.A. 2019. Kinetid structure in larval and adult stages of the demosponges *Haliclona aquaeductus* (Haplosclerida) and *Halichondria panicea* (Suberitida) // Zoomorphology. Vol.138. P.171–184.

Sokolova A.M., Pozdnyakov I.R., Schepetov D.M., Ereskovsky A.V., Karpov S.A. 2020. Kinetid in larval cells of Spongillida (Porifera: Demospongiae): tracing the ancestral traits // Organisms Diversity and Evolution. Vol.20. P.669–680.

Strakhovskaya M., Ivanova E., Fraikin G. 1993. Stimulatory effect of serotonin on the growth of the yeast *Candida guillermondii* and the bacterium *Streptococcus faecalis* // Microbiologia. Vol.62. P.46–49.

Tsang W.H., McGaughey Y.H., Wong Y.H., Wong J.T.Y. 1997. Melatonin and 5-methoxytryptamine induced muscular contraction in sea anemones // Journal of Experimental Zoology. Vol.279. P.201–207.

Tsavkelova E., Botvinko I., Kudrin V., Oleskin A. 2000. Detection of neurotransmitter amines in microorganisms using of high performance liquid chromatography // Doklady Akademii Nauk. Vol.372. P.115–117.

Umbriaco D., Anctil M., Descarries L. 1990. Serotonin-immunoreactive neurons in the cnidarian *Renilla koellikeri* // Journal of Comparative Neurology. Vol.291. P.167–178.

Wada Y., Mogami Y., Baba S. 1997. Modification of ciliary beating in sea urchin larvae induced by neurotransmitters: beat-plane rotation and control of frequency fluctuation // The Journal of experimental biology. Vol.200. P.9–18.

Wan J., Zhang P., Wang R., Sun L., Ju Q., Hu J. 2018. Comparative physiological responses and transcriptome analysis reveal the roles of melatonin and serotonin in regulating growth and metabolism in *Arabidopsis* // BMC Plant Biology. Vol.18. P.362. doi: 10.1186/s12870-018-1548-2

Westfall J.A., Elliott S.R., MohanKumar P.S., Carlin R.W. 2005. Immunocytochemical evidence for biogenic amines and immunogold labeling of serotonergic synapses in tentacles of *Aiptasia pallida* (Cnidaria, Anthozoa) // Invertebrate Biology. Vol.119. P.370–378.

Welsh J.H. 1960. 5-Hydroxytryptamine in coelenterates // Nature. Vol.186. P.811–812. doi:10.1038/186811a0

Welsh J.H. 1968. Distribution of serotonin in the nervous system of various animal species // Advances in Pharmacology. Vol.6. P.171–188. doi:10.1016/s1054-3589(08)61171-0.

Weyrer S., Rutzler K., Rieger R. 1999. Serotonin in Porifera? Evidence from developing *Tedania ignis*, the Caribbean fire sponge (Demospongiae) // Memoirs of the Queensland Museum. Vol.44. P.659–666.

Wood J.G., Lentz T. 1964. Histochemical localization of amines in hydra and the sea anemone // Nature. Vol.201. P.88–90. doi:10.1038/201088b0.

Zega G., Pennati R., Fanzago A., De Bernardi F. 2004. Serotonin involvement in the metamorphosis of the hydroid *Eudendrium racemosum* // International Journal of Developmental Biology. Vol.51. P.307–313.

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