Antioxidant quinonoid pigments from ceolomic fluid of Far Eastern sea urchins

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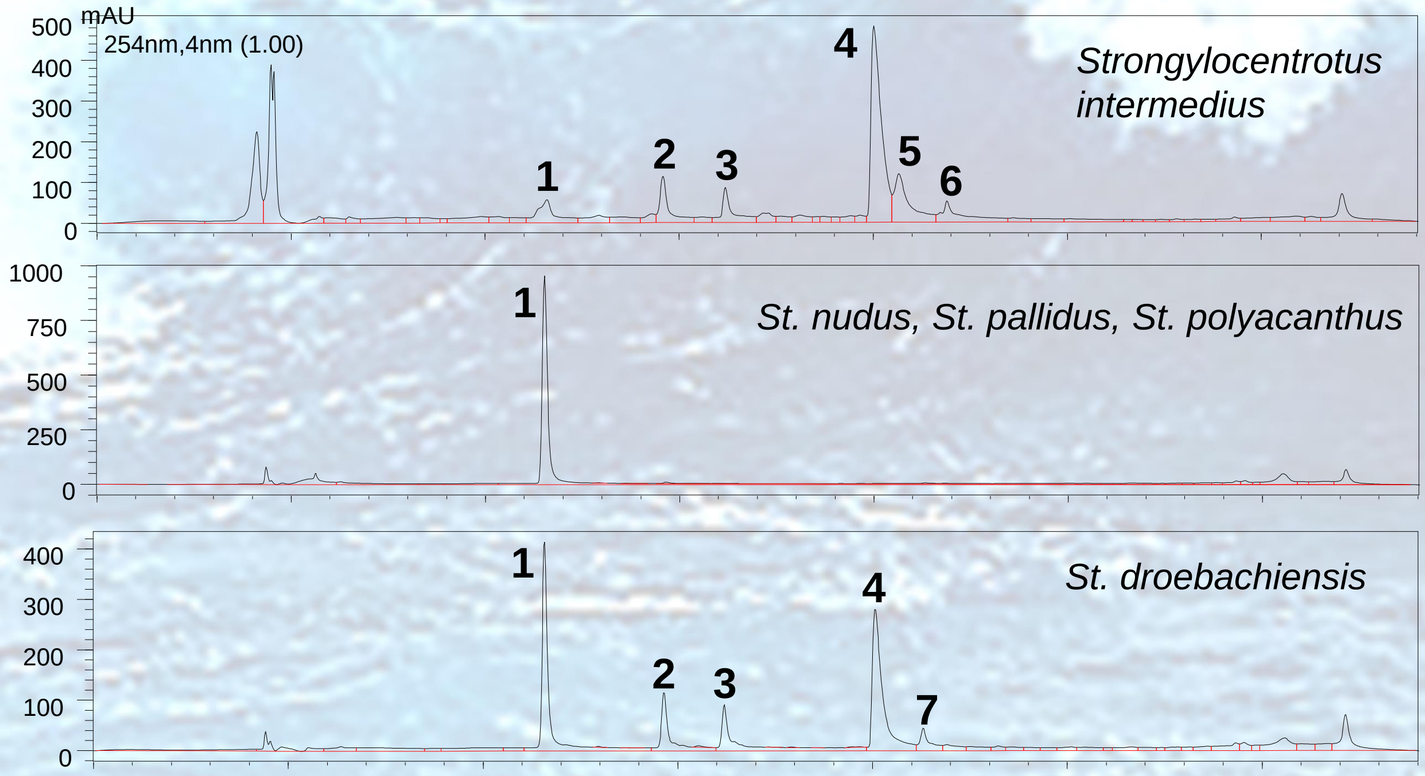
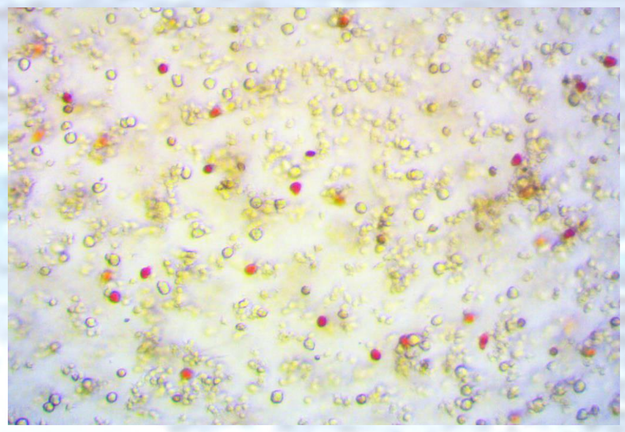
The coelomic fluid of the sea urchin contains cells, generically called coelomocytes, that have been studied for many decades. Due to their capability to respond to injuries, host invasion, and cytotoxic agents, coelomocytes are regarded as the immune effectors of the sea urchin. One of the subpopulations of coelomocytes are spherical cells with red cytoplasmic granules. In 1885 MacMunn isolated the red pigment from the ceolomic fluid of *Echinus esculentus* and called it echinochrome A. Since that there is an opinion, that ceolomic fluid of sea urchins contains echinochrome A regardless from their species.

We investigated <u>for the first time</u> the composition of quinonoid pigments from ceolomic fluid of eight sea urchin species of the Sea of Japan and the Sea of Okhotsk (*Strongylocentrotus intermedius*, *St. pallidus*, *St. droebachiensis*, *St. polyacanthus*, *Mesocentrotus nudus*, *Echinocardium cordatum*, *Scaphechinus mirabilis*, *Echinarachnius parma*) using HPLC-DAD-MS:





Ceolomic fluid of *Scaphechinus mirabilis*

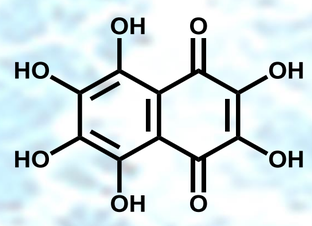
Sc. mirabilis

St. intermedius

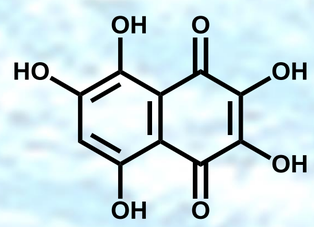
E. cordatum

St. droebachiensis

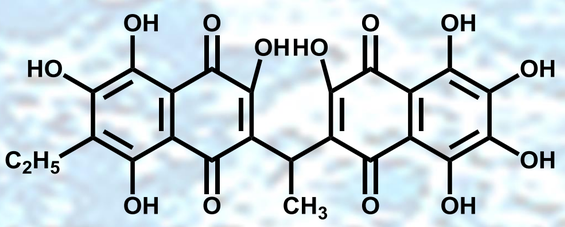
№	Compound
1	Spinochrome E
2	Spinochrome D
3	$[M-H]^-$ m/z 535
4	Ethylidene-6,6’-bis(2,3,7-trihydroxynaphthazarin)
5	$[M-H]^-$ m/z 765
6	$[M-H]^-$ m/z 483, 527
7	Echinochrome A
8	Dehydroechinochrome



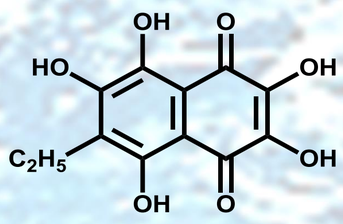
Spinochrome E (1)



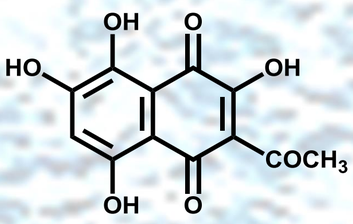
Spinochrome D (2)



Ethylidene-6,6’-bis(2,3,7-trihydroxynaphthazarin) (4)

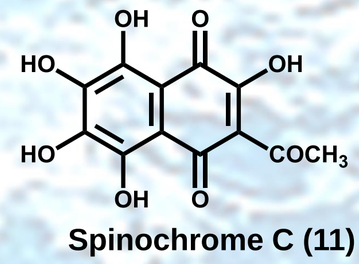


Echinochrome A (7)



Spinochrome A (9)

Spinochrome B (10)



Spinochrome C (11)

We discovered that <u>composition of ceolomic fluid pigments differs between the species</u> and includes all known naphthoquinones of sea urchins – spinochromes E (**1**) and D (**2**), echinochrome A (**7**), binaphthoquinone **4** and some new compounds (**3**, **5**-**6**). Main ceolomic fluid pigments along with spinochromes A, B, and C (**9**, **10**, **11**) were isolated previously from shells and spines of sea urchins *M. nudus* and *Sc. mirabilis* [1, 2] and were tested for their ability to scavenge the stable DPPH radical and to inhibit lipid peroxidation [methods description in 1]. Echinochrome A, spinochromes C and E showed the highest antioxidant activity on both models.

Table 1: DPPH scavenging activities of compounds **1-2, 7, 9-11** compared with α-tocopherol[a]

Compound	EC_{50}, μM
1	12.32
2	48.36
7	7.86
9	37.06
10	23.04
11	17.04
α-tocopherol	16.24

[a] Each value is presented as the mean ± standard deviation (n = 3).

Table 2. Antioxidant activity of compounds **1-2, 7, 9-11** compared with vitamin C in lipid peroxidation test[a]

Compound	0.4 мМ		0.8 мМ	
	τ, h	AOA	τ, h	AOA
1	6.2±0.2	1.3	8.6±0.2	1.4
2	29.6±0.9	2.4	35.0±1.0	2.6
7	161.0±9.6	8.6	318.4±15.9	16.2
9	10.5±0.4	1.5	15.1±0.6	1.7
10	20.1±1.0	1.9	33.8±1.7	2.6
11	96.5±5.8	5.6	168.0±10.1	9.0
Vitamin C	1.3±0.01	1.1	1.5±0.01	1.1

The induction times τ and τ_0 for which the substrate weight started to increase in the presence and absence of the tested compound because of peroxide production, respectively, were determined, and relative antioxidant activity (AOA) was calculated as the ratio of τ and $\tau + \tau_0$. The well-known antioxidant vitamin C was used as positive control.

References:

1. E. A. Vasileva, N. P. Mishchenko, P. A. Zadorozhny, S. A. Fedoreyev. Nat. Prod. Communications, 2016, Vol. 11, No. 6, P. 821-824.
2. N. P. Mishchenko, E. A. Vasileva, S. A. Fedoreyev. Tetrahedron Letters, 2014, Vol. 55, P. 5967-5969.

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