

PROLIFERATIVE STATUS DETERMINATION OF BENTHIC INVERTEBRATES COELOMOCYTES/HEMOCYTES

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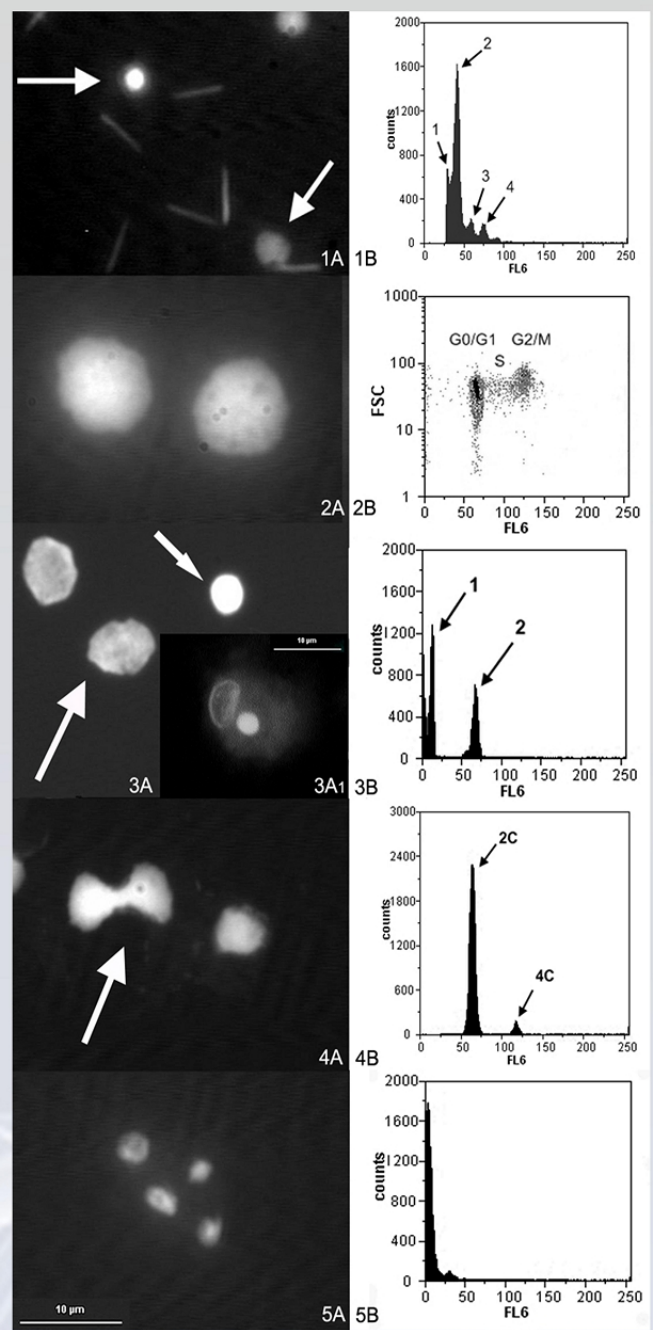
Marine invertebrates play a very important role in the maintenance of the health of marine ecosystems and serve as bio-indicators. Coelomocytes/hemocytes of benthic invertebrates, as primary immune effectors that respond to stress conditions, injuries, host invasion and cytotoxic/genotoxic agents, have been used for the monitoring of environmental conditions [1, 2]. As a free circulating they are easily collected and analyzed by flow cytometry. The DNA content is a marker of cellular maturity since the frequency of cells in G0, S and G2 phases could determine the proliferative status of the investigated cells. When the measurement of DNA content is performed with internal standard, flow cytometry provides information about genome size. [3]

Table 1. Descriptive statistics of genome size variation within 5 species of marine invertebrates calculated from distribution of DNA values of 10 specimens.

Species	DNA content (2C)/pg	Range	CV
<i>Aphrodita aculeata</i>	1.24±0.06	1.15-1.30	4.8
<i>Maja crispata</i>	7.76±0.10	7.63-7.91	1.3
<i>Echinaster sepositus</i>	1.52±0.03	1.46-1.54	2.0
<i>Paracentrotus lividus</i>	1.08±0.02	1.05-1.12	4.6
<i>Phallusia mammillata</i>	0.11±0.05	0.07-0.14	45

Conclusions

- Flow cytometry analyses of DAPI-stained nuclei identified the sea mouse *A. aculeata* coelomocytes as proliferating cells.
- Fluorescence microscopy of DAPI-stained cells identified the presence of phagocytic activity in unchallenged red starfish *E. sepositus*.
- Evidence of karyokinesis of both spiny crab hemocytes and sea urchin coelomocytes revealed that their division is completed in the hemolymph and coelom, respectively.
- The diploid DNA content (2C) in sea mouse *A. aculeata*, spiny crab *M. crispata*, red starfish *E. sepositus* and sea urchin *P. lividus* is 1.24, 7.76, 1.52 and 1.08 pg, respectively.
- DNA content of sea mouse *A. aculeata* and spiny crab *M. crispata* are the first records of their genome size. Furthermore, the DNA content of the sea mouse *A. aculeata* is the first genome size record among the species in the Aphroditidae family.



(1) *Aphrodita aculeata*. (2) *Maja crispata*. (3) *Echinaster sepositus*. (4) *Paracentrotus lividus*. (5) *Phallusia mammillata*. - (A) DAPI - stained nuclei and cells (3A1). (B) DAPI-fluorescence (FL6) distribution.

References

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