Does symbiotic relationship between *Hydra viridissima* and photoautotrophic alga provide evolutionary advantage in securing the DNA against damage by cytotoxic or genotoxic mode of action of environmental stressors?

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Abstract

Symbiosis is an evolutionary strategy that may provide biological advantages. The most complex and tight cooperation between symbiotic organisms is achieved in endosymbiosis, as it is the symbiosis between green hydra (*Hydra viridissima* Pallas, 1766) and unicellular photoautotrophic alga. We aimed to evaluate whether this symbiotic cooperation bestows higher resistance to environmental stressors of different nature, in terms of higher preservation of DNA integrity, compared to free-living brown hydra (*Hydra oligactis* Pallas, 1766). Hydras were exposed to herbicide norflurazon at of 2×10^{-7} or 2×10^{-6} mol/L and UV-B light of 254 nm, 0.023 mW/cm separately or simultaneously to detect additive or synergistic effect. We used alkaline comet assay to determine the level of primary DNA damage and fluorescent staining to detect apoptosis and necrosis. Norflurazon at 2×10-6 mol/L significantly increased both comet assay descriptors in brown hydra compared to negative control (6.17±0.6μm and 5.2±1.7% vs. 2.9 ± 0.2μm and 1.2 ± 0.2 %). The frequency of apoptotic and necrotic cells was significantly elevated either, being higher in brown hydra (25.7±3.5% and 8.2±0.2%) than green hydra (20.3±2.5% and 2.8±0.2%). UV-B irradiation induced significant DNA damage in brown hydra only (13.5±1.0μm, 4.1±1.0%). Simultaneous exposure to UV-B light and norflurazon led to synergistic increase in intensity of adverse effects. A concentration of 2×10^{-7} mol norflurazon/L combined with UV-B light induced significantly increased DNA lesion-level in brown hydra (15.8 ± 3.1%) and at concentration of 2×10^{-6} mol/L in both species (brown: 15.0±2.6%, green: 22.0±3.0%). Significant cytotoxicity and increased hedgehog nucleoids frequency were recorded, significantly more pronounced in brown (78.3±9.4% and 56.4±6.0%) compared to green hydra (34.7±2.5% and 24.2±0.6%). Besides genotoxicity, comet assay descriptors were significantly affected by and reflected cytotoxicity of norflurazon and/or UV-B light. We proved that evolutionary established symbiotic cooperation may contribute to higher resistance to cyto/genotoxic stressors.

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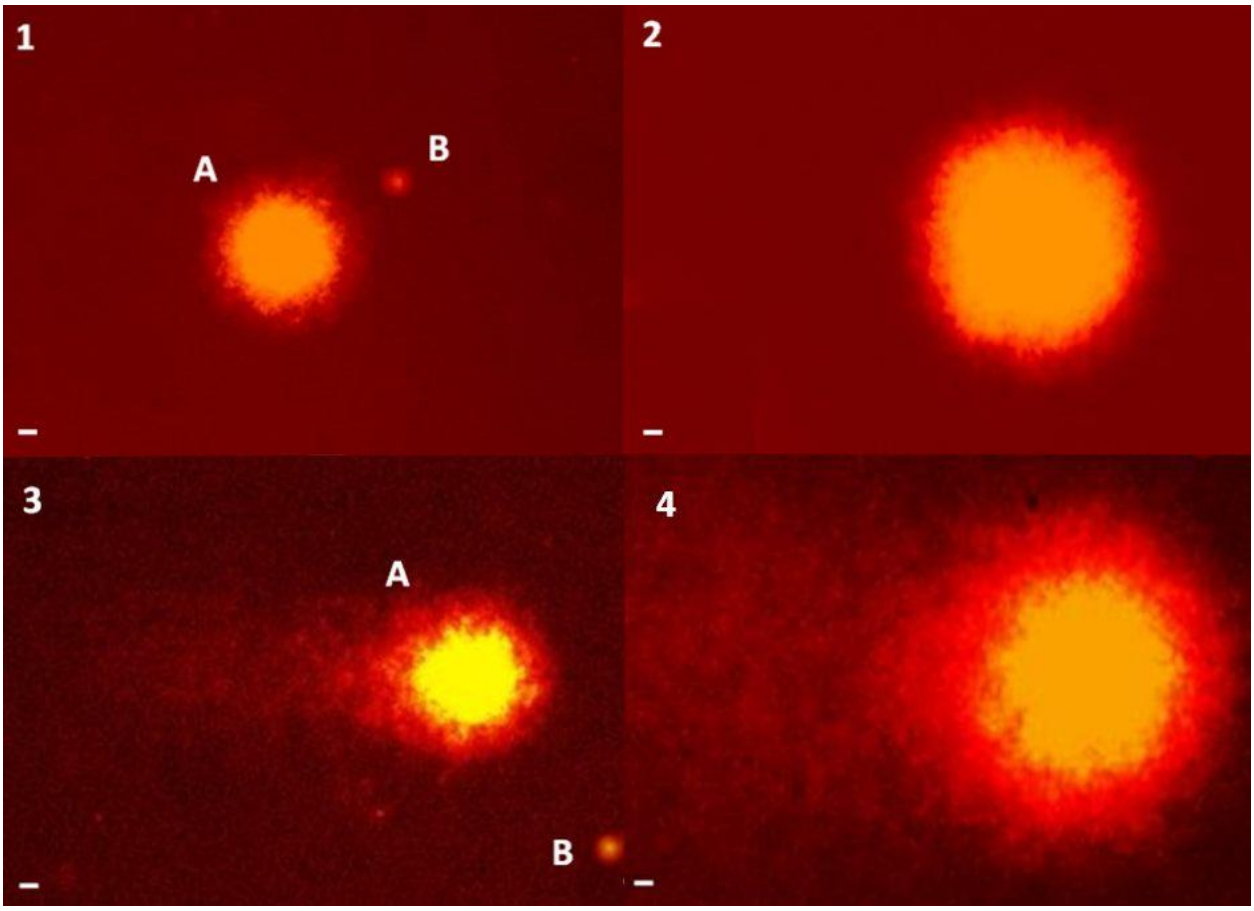
`Hydras Comet Final.docx` available at https://authorea.com/users/569499/articles/615263-does-symbiotic-relationship-between-hydra-viridissima-and-photoautotrophic-alga-provide-evolutionary-advantage-in-securing-the-dna-against-damage-by-cytotoxic-or-genotoxic-mode-of-action-of-environmental-stressors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B
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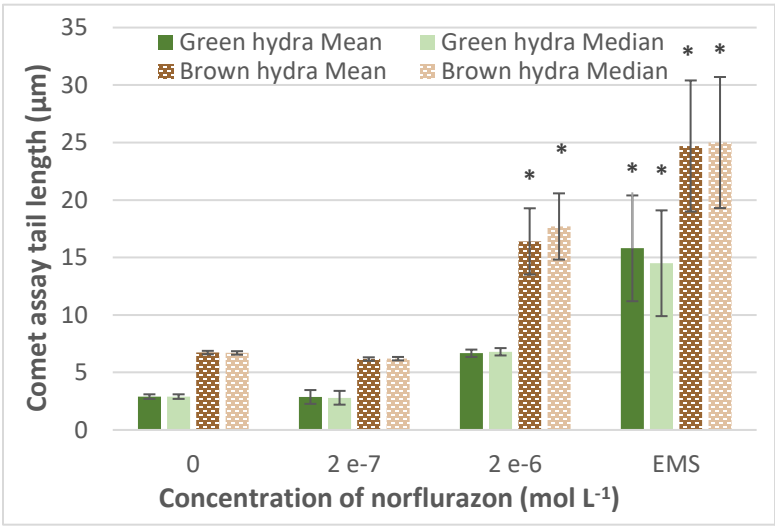
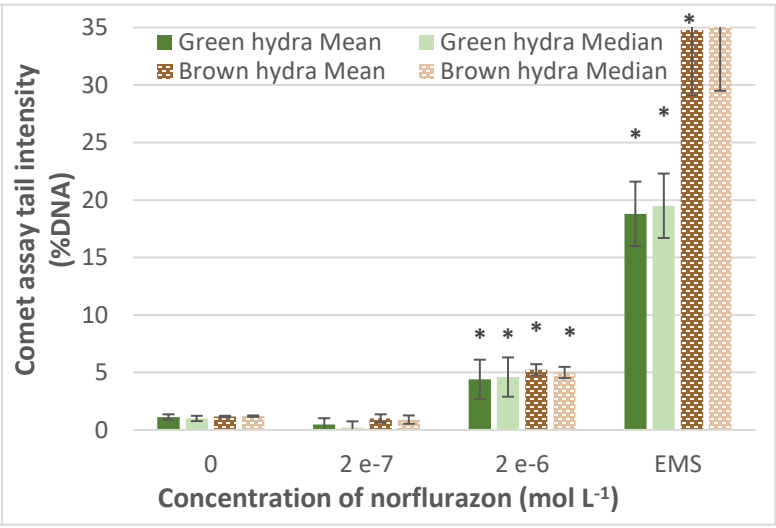
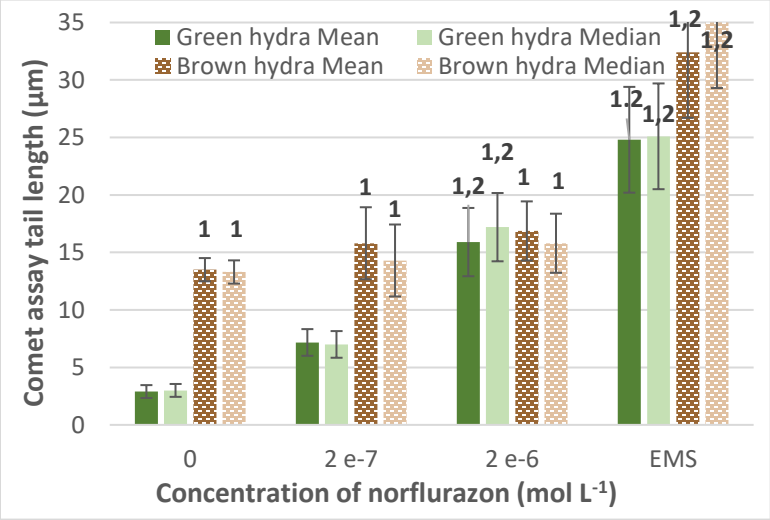
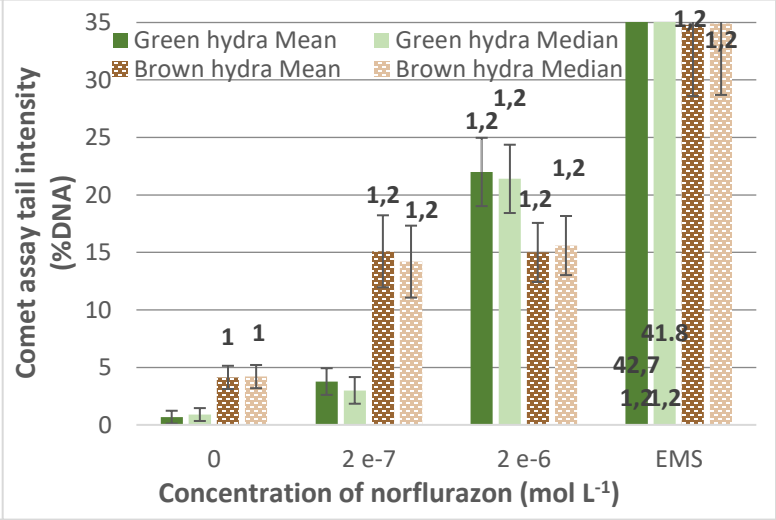
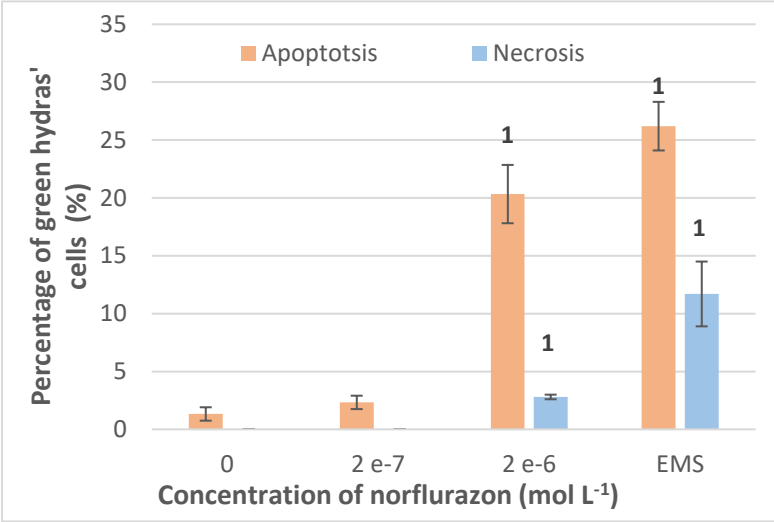
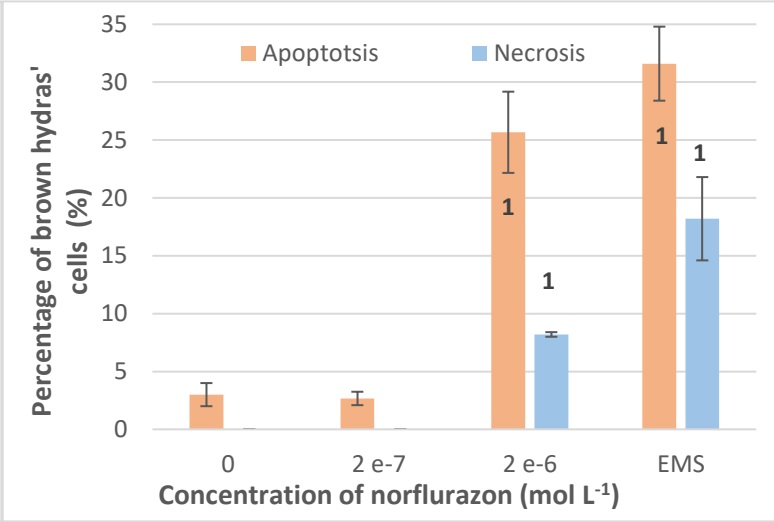

Green hydra Mean
Green hydra Median
Brown hydra Mean
Brown hydra Median
Comet assay tail length (µm)
35
30
25
20
15
10
5
0
*
*
*
*
*
*
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)

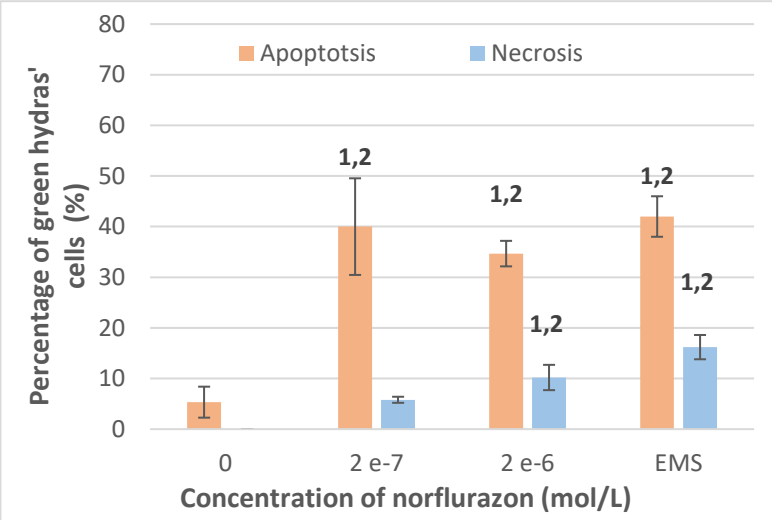

Green hydra Mean
Green hydra Median
Brown hydra Mean
Brown hydra Median
Comet assay tail intensity (%DNA)
35
30
25
20
15
10
5
0
*
*
*
*
*
*
*
*
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)

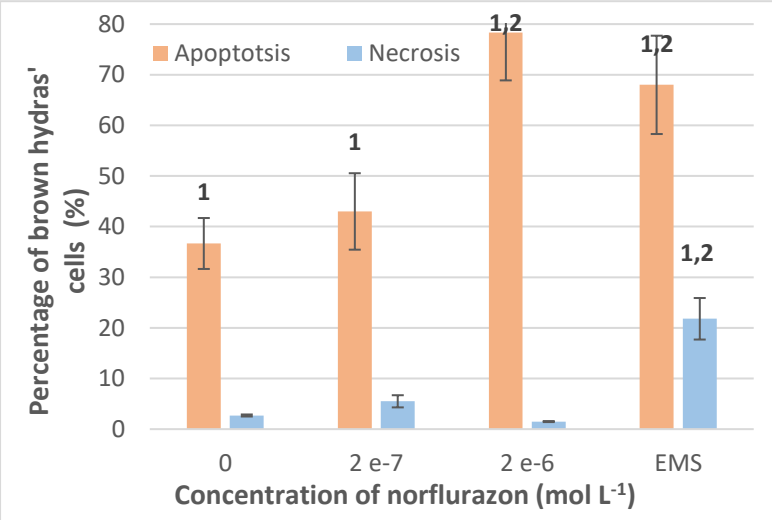

Green hydra Mean
Green hydra Median
Brown hydra Mean
Brown hydra Median
Comet assay tail length (µm)
0
5
10
15
20
25
30
35
1 1
1 1
1,2 1,2 1 1
1,2 1,2 1,2 1,2
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)


Green hydra Mean
Green hydra Median
Brown hydra Mean
Brown hydra Median
Comet assay tail intensity (%DNA)
0
5
10
15
20
25
30
35
1 1
1,2 1,2
1,2 1,2 1,2 1,2
42,7 41.8 1,2 1,2
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)


Percentage of green hydras' cells (%)
35
30
25
20
15
10
5
0
Apoptotsis
Necrosis
1
1
1
1
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)


Percentage of brown hydras' cells (%)
35
30
25
20
15
10
5
0
Apoptotsis
Necrosis
1
1
1
1
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)


Apoptotsis
Necrosis
Percentage of green hydras' cells (%)
80
70
60
50
40
30
20
10
0
1,2
1,2
1,2
1,2
1,2
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol/L)


Apoptotsis
Necrosis
Percentage of brown hydras' cells (%)
80
70
60
50
40
30
20
10
0
1
1
1,2
1,2
1,2
0
2 e-7
2 e-6
EMS
Concentration of norflurazon (mol L-1)