

Differences of semi-static systems on growth and sensitivity of *Myriophyllum spicatum* compared to static test in accordance with OECD 239

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The ring-test on the rooted aquatic plant *M. spicatum* in a water/sediment system was conducted under static conditions. However, guideline OECD 239 also mentions to use systems with several water changes in case of degradation of test item. The basic idea behind is to prove a constant exposure for risk assessment. But an influence on the test system was not clarified and the system is more artificial, since natural degradation was not taken into account. This might under or overestimate effects of degrading test items.

Further, water changes influence the pH and might increase the amount of nutrients in the water or wash out nutrients from the sediment. This can lead to a higher or lower sensitivity to chemicals. Stabilisation of test items can also be reached by buffering the test system for pH sensitive test items. We present data with buffered and non-buffered test systems and compare growth of control plants from semi-static and static designs.

Material & Methods

Test Substance: 3,5-DCP
Test Duration: 14 days
Test Concentrations: 1.50, 3.00, 6.00 and 12.0 mg/L (nominal)
Test Design: based on OECD Guideline 239; static or semi-static (water change on day 4, 7, 9, 12)

Treatment	pH		
	static	semi-static	static + MES
Control	8.68	8.38	6.63
1.50	8.64	8.26	6.61
3.00	8.64	8.03	6.62
6.00	7.64	7.59	6.60

Treatment	O ₂ [mg/L]		
	static	semi-static	static + MES
Control	10.52	9.61	10.27
1.50	10.15	9.39	9.48
3.00	11.20	9.04	8.53
6.00	7.46	7.31	7.42

Effects of 3,5-DCP on *Myriophyllum spicatum* plants



Increasing effects due to increasing concentration of 3,5-DCP and increasing time

Results

Growth of control plants from static buffered (+ MES buffer) and semi-static test design compared to static designs expressed in % reduction

Design	Total Shoot Length			Fresh Weight			Dry Weight		
	[cm]	Yield	Growth rate	[mg]	Yield	Growth rate	[mg]	Yield	Growth rate
Static	35.0			612			32.6		
Semi-static	29.8	17.9	9.1	828	-41.7	-16.3	50.0	-80.1	-41.0
Static + MES	24.6	35.9	20.2	869	-49.5	-18.7	50.0	-80.0	-40.9

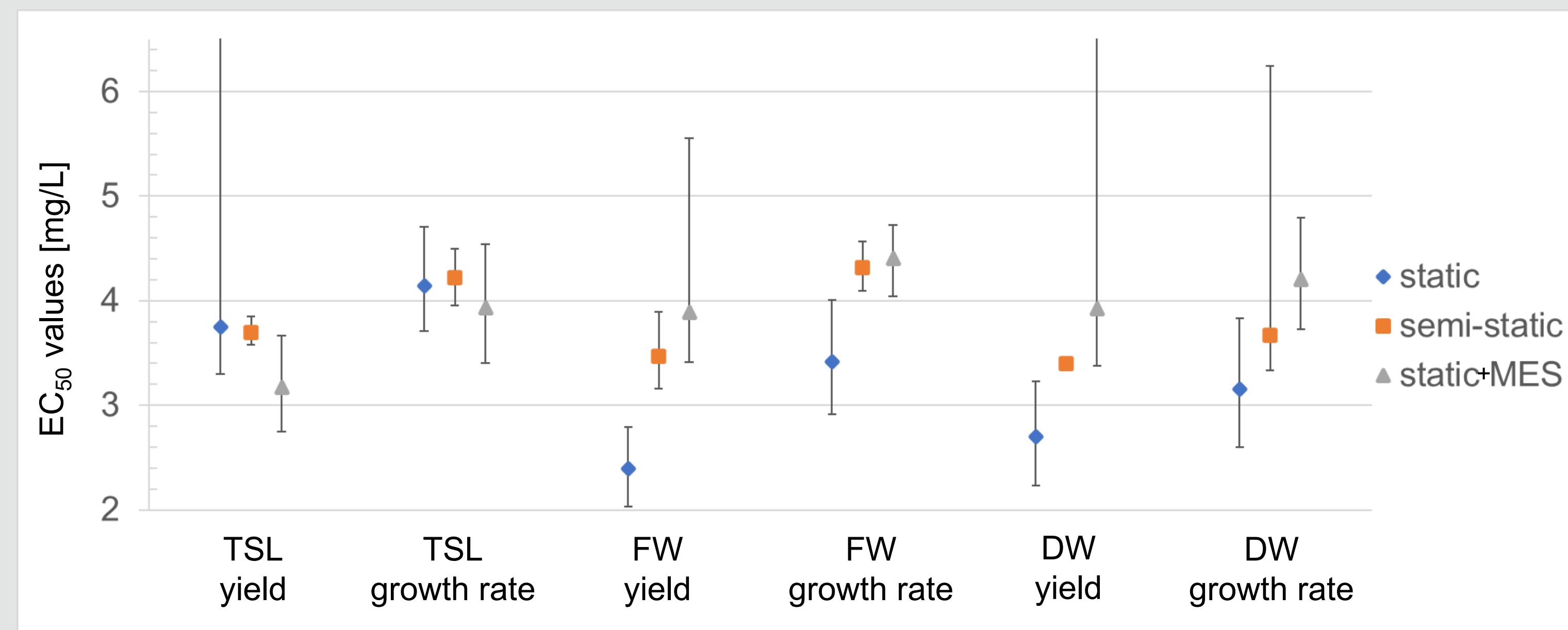
Compared to the static design, control plants of the semi-static und buffered design showed **reduced total shoot length**.

- 17.9% reduction in yield and 9.1% reduction in growth rate for the semi-static design.
- 35.9% reduction in yield and 20.2% reduction in growth rate for the buffered design.

For **biomass** an **increase** in the semi-static as well as in the buffered design was observed.

- **for fresh weight** -41.7% reduction in yield and -16.3% reduction in growth rate for the semi-static design.
- -49.5% reduction in yield and -18.7% reduction in growth rate for the buffered design (fresh weight).
- **for dry weight** -80.1% reduction in yield and -41.0% reduction in growth rate for the semi-static design.
- -80.0% reduction in yield and -40.9% reduction in growth rate for the buffered design (dry weight).

Endpoints from static (+ MES buffer) and semi-static tests of *Myriophyllum spicatum* with 3, 5-DCP



EC₅₀ values of total shoot length of the semi-static und buffered design are comparable to EC₅₀ values of the static design.

For biomass EC₅₀ **values are lower in** the semi-static and buffered design compared to the static design.

Taking the hydrolyzation of 3,5-DCP into account endpoints of the static test would be even lower (by ~20%).

Chlorosis, necrosis and reduced number of roots were observed for all test systems at day 14 for the concentration levels 6.00 and 12.0 mg/L.

Discussion

All test designs fulfilled validity criteria according to OECD 239. But it became obvious that water renewals have an influence on the growth of plants. The surprising results that water renewals and buffered systems indicates an increase in plant weight but decrease plant length compared to static designs needs further clarification. Other studies in the past indicates that control plants in static designs with buffer showed an increase in all growth parameters compared to non-buffered test. And in case tests are performed in a semi-static buffered systems an decreased growth compared to non-buffered systems could be observed. Further different growth between seasons or plant source or age of shoots was observed. It could also be observed that shoot length and fresh weight is more sensitive than dry weight. But in recovery phases the dry weight becomes more sensitive compared to fresh weight. All this indicates that depending on the test design effects on the ECx values are likely. Based on these findings further studies will be needed in order to clarify all upcoming questions.