

A digital tool to monitor population dynamics

development of counting methods for Daphnia populations and possible use for other organisms

Harm van der Geest (UvA)
Simone Lucker (UvA)
Miriam Leon Paumen (UvA,VU)
Ruud Kampf (HHNK,VU)
Barry Pieters (UvA,UFZ)



Eversteoog
on Texel

Sewage treatment
plant
with constructed
wetland for effluent
polishing





Eversteekoog
Texel



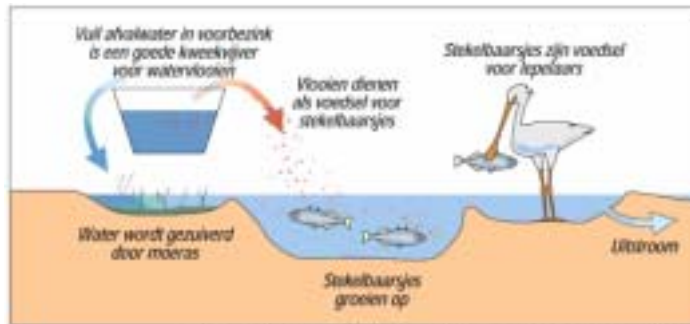
3



24 juni 2004

4

'Kwekelbaarsjes' 'Gricklebacks' project HHNK



UvA/LE/IER/HHNK - De Volkskrant - Bron: Hengeloort waterloop van Utrechtse Stroom, Holland Nieuwskrant



Daphnia as food for Sticklebacks



7

Experimental system



Bakken
Mesocosms



Vijvers
Ponds

8

Counting of Daphnia

- Ecotoxicological testing, 'Grickleback' project

- By hand
 - Labor intensive > expensive
 - Destructive methods > population dynamics / scale of experiments

- Need for a better method !

Aim of this project

- Development of a digital tool to count Daphnia
 - UFZ (living material)
 - UvA (fixated material)

- Monitor Daphnia populations in experimental setup 'grickelback' project

Eco-toxicological Research UFZ

- Method developed by the Effect Propagation group department of Chemical Ecotoxicology of UFZ Centre for Environmental Research Leipzig-Halle
- M. Liess and B. Pieters, in cooperation with UvA

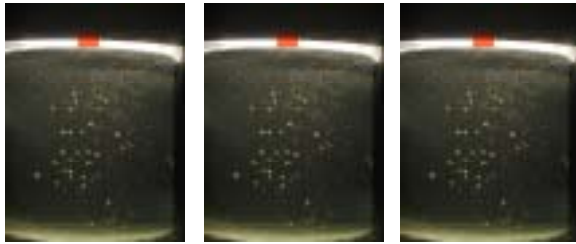


11

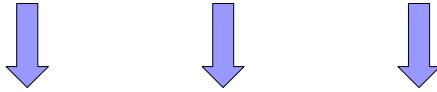


Toxicity test in 4.7 l round vessels with a black backside
Digital camera mounted in a device aimed on a vessel

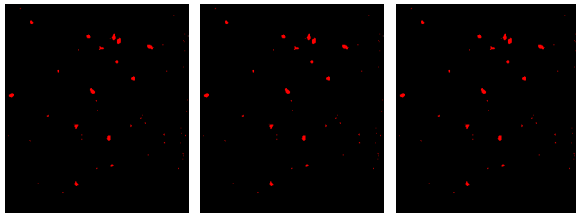
12



3 pictures
Short time interval



LIVING DAPHNIA !



Digital imaging
substraction of
particles not moved

13

UFZ method

- Very useable method !
- Makes it possible to follow population dynamics on life Daphnia in groups
- Making the pictures is an easy job
- But, processing data less easy
- Field samples ?
- Thesis Barry Pieters

14

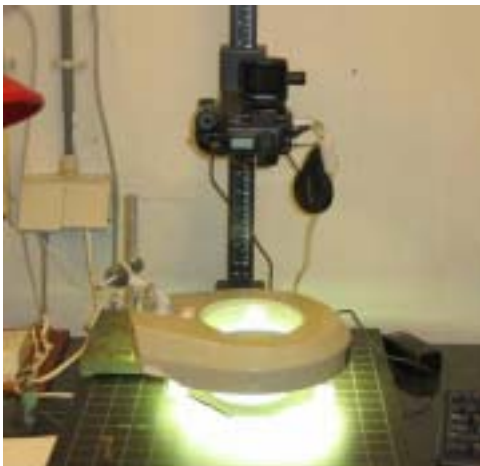
Counting fixated material (UvA)



Sampling in the field
Fixation in ethanol
Analysis in laboratory

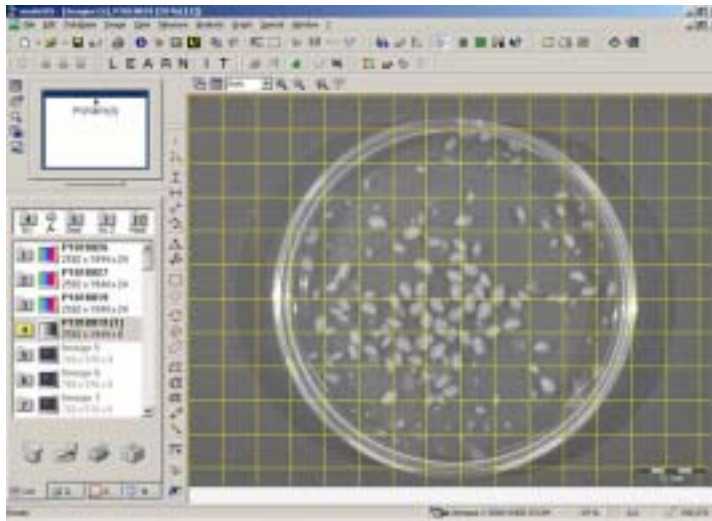
15

setup

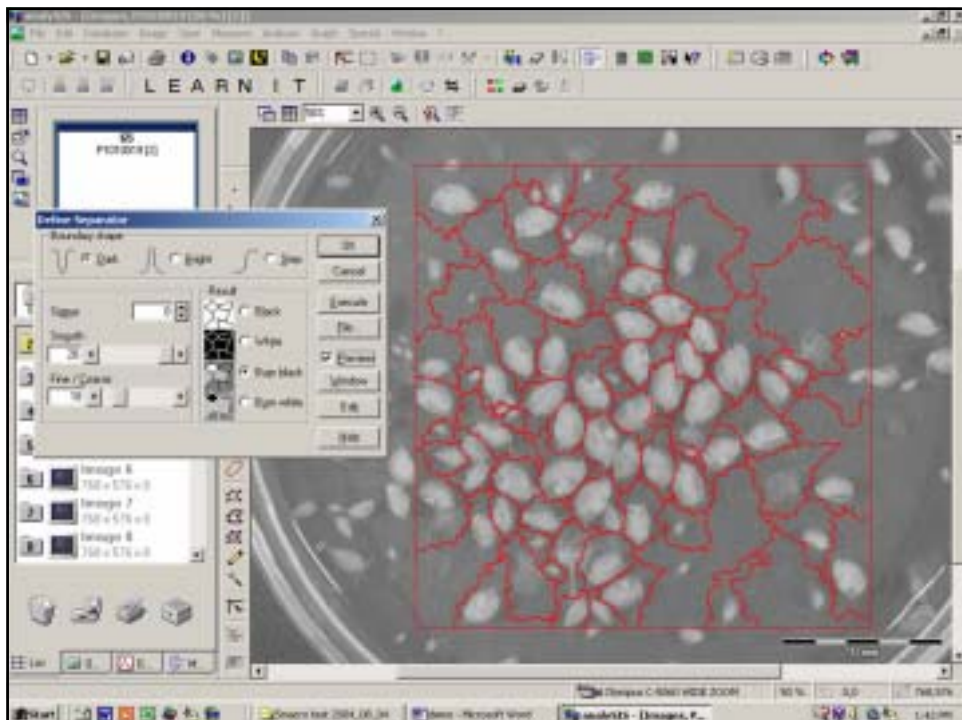


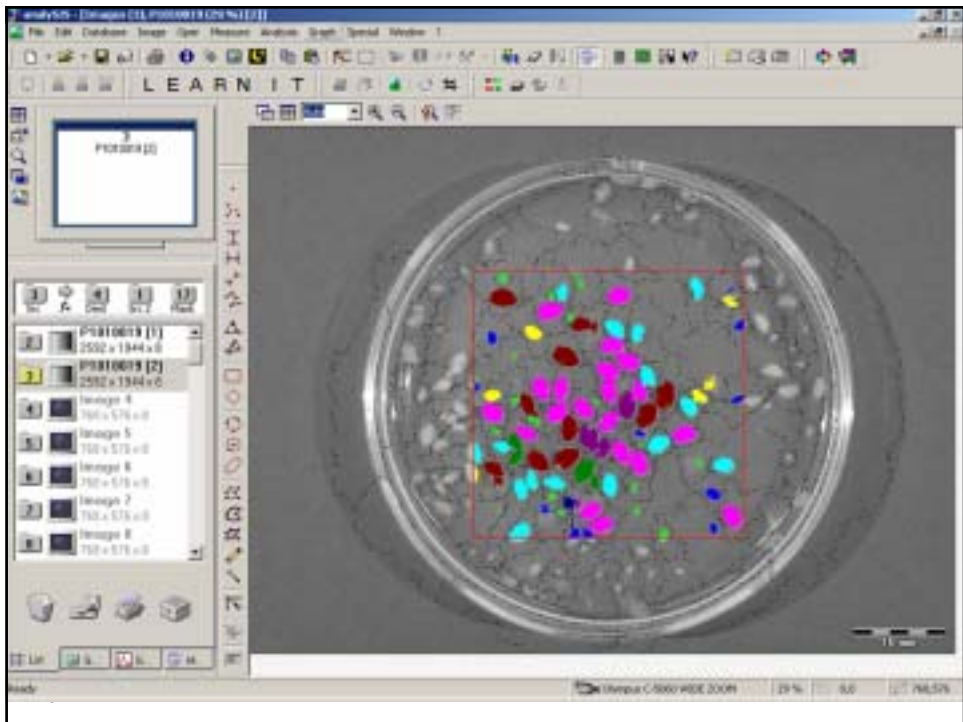
16





Standard software (Analysis FIVE)



17





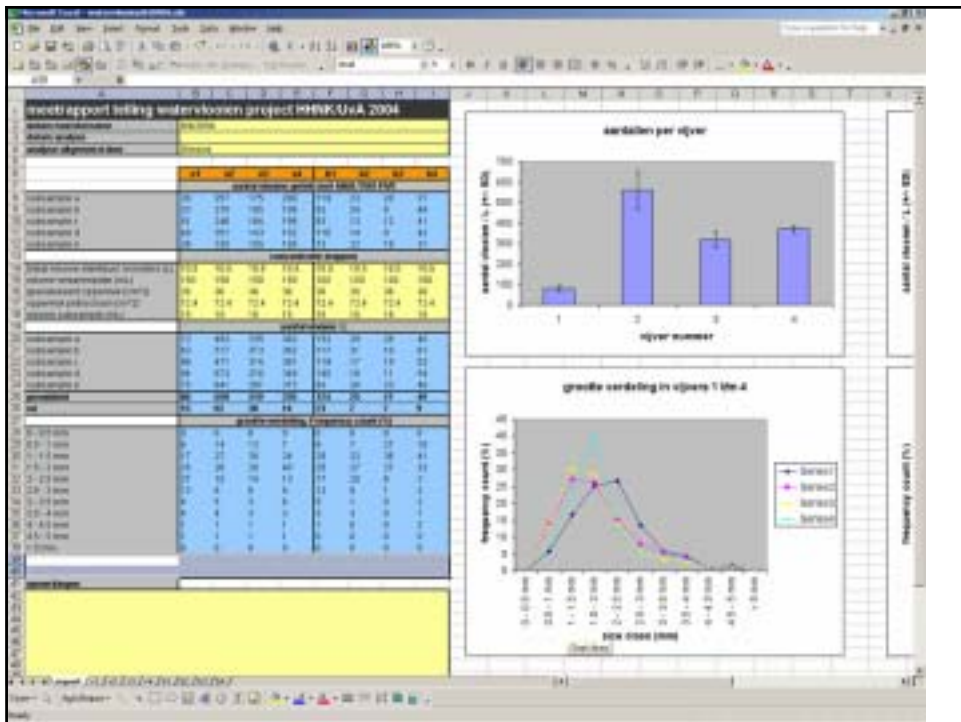
DE UNIVERSITEIT VAN AMSTERDAM  UvA   

Output: numbers/size distribution

The screenshot shows the LEARN IT software interface with a data table and a histogram. The data table displays the results of the analysis, including particle numbers and sizes. The histogram shows the distribution of these values, with the x-axis representing size and the y-axis representing the number of particles.

Particle ID	Size (µm)	Color
1	8	Green
2	8	Green
3	8	Green
4	8	Green
5	8	Green
6	8	Green
7	8	Green
8	8	Green
9	8	Green
10	8	Green
11	8	Green
12	8	Green
13	8	Green
14	8	Green
15	8	Green
16	8	Green
17	8	Green
18	8	Green
19	8	Green
20	8	Green
21	8	Green
22	8	Green
23	8	Green
24	8	Green
25	8	Green
26	8	Green
27	8	Green
28	8	Green
29	8	Green
30	8	Green
31	8	Green
32	8	Green
33	8	Green
34	8	Green
35	8	Green
36	8	Green
37	8	Green
38	8	Green
39	8	Green
40	8	Green
41	8	Green
42	8	Green
43	8	Green
44	8	Green
45	8	Green
46	8	Green
47	8	Green
48	8	Green
49	8	Green
50	8	Green
51	8	Green
52	8	Green
53	8	Green
54	8	Green
55	8	Green
56	8	Green
57	8	Green
58	8	Green
59	8	Green
60	8	Green
61	8	Green
62	8	Green
63	8	Green
64	8	Green
65	8	Green
66	8	Green
67	8	Green
68	8	Green
69	8	Green
70	8	Green
71	8	Green
72	8	Green
73	8	Green
74	8	Green
75	8	Green
76	8	Green
77	8	Green
78	8	Green
79	8	Green
80	8	Green
81	8	Green
82	8	Green
83	8	Green
84	8	Green
85	8	Green
86	8	Green
87	8	Green
88	8	Green
89	8	Green
90	8	Green
91	8	Green
92	8	Green
93	8	Green
94	8	Green
95	8	Green
96	8	Green
97	8	Green
98	8	Green
99	8	Green
100	8	Green

The histogram shows the distribution of particle sizes. The x-axis represents size (µm) and the y-axis represents the number of particles. The distribution is bimodal, with a peak at 8 µm and a secondary peak at 18 µm.



UvA method

- Works quick and easy
- Software easy to use (macro)
- Database
- Separate Daphnia from 'noise' (shape, size)

Application in the field



23

VU (adapted from UFZ)



24

VU (adapted from UFZ)

- Counting living Daphnia
- Field method based on work of UFZ works,
- BUT, problems with
 - 'front - side' effect
 - suspended solids, noise

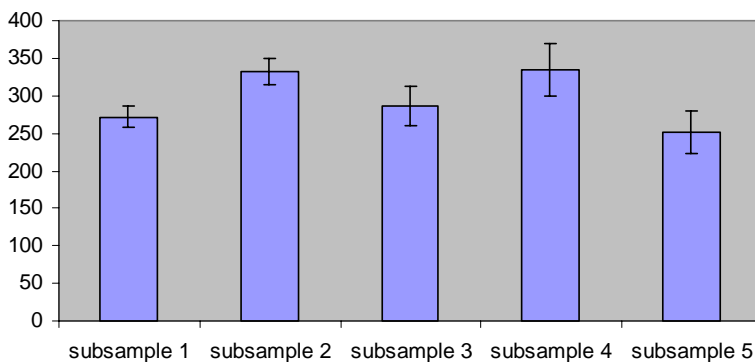
UvA method in the field
Shape/size recognition
is able to separate
Daphnia from 'noise'



25

Method evaluation

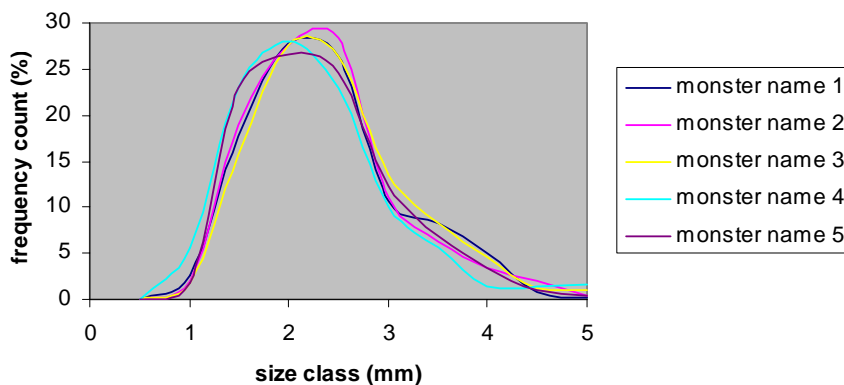
aantal individuen average



Manual counts, digital counts $R^2=0.94$

26

Method evaluation



27

Daphnia in 'Grickleback' system



2004

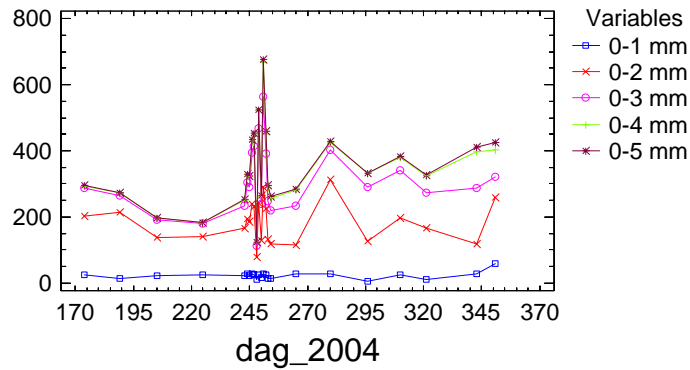
Every two weeks

Week 36/37
each day

28

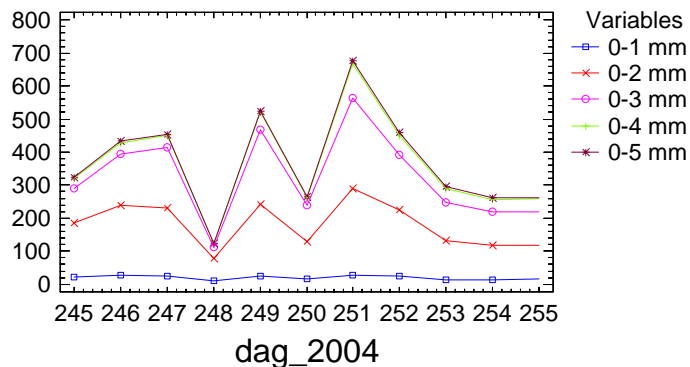
Daphnia in 'Grickleback' system

Bak 1 - Daphnia cumulative (per l)



Daphnia in 'Grickleback' system

Bak 1 - Daphnia cumulative (per l) - intensive



Potential other applications ?



31

Potential other applications ?

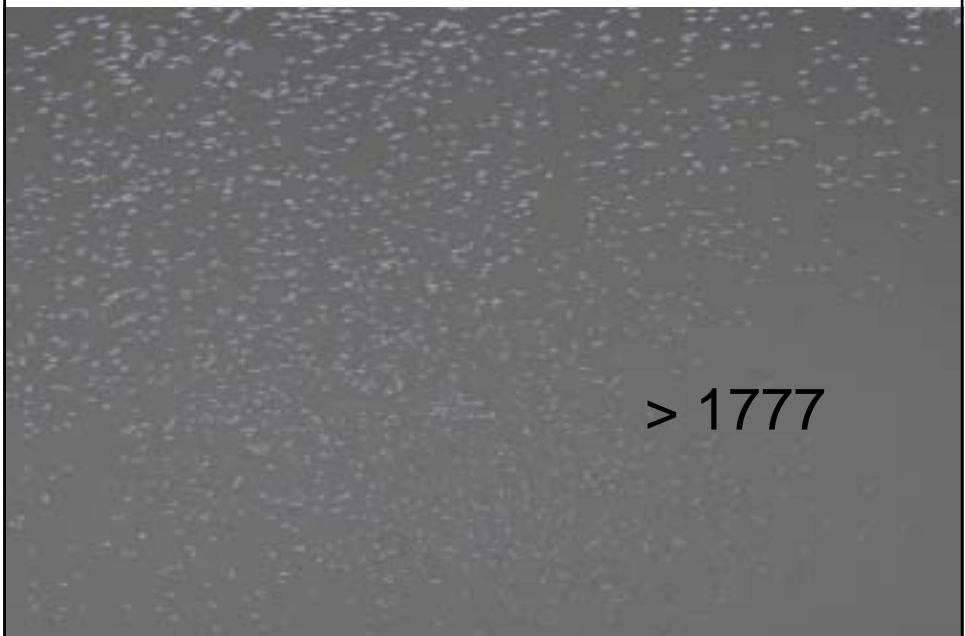


32



41 Golden Plovers





> 1777

Folsomia tox test

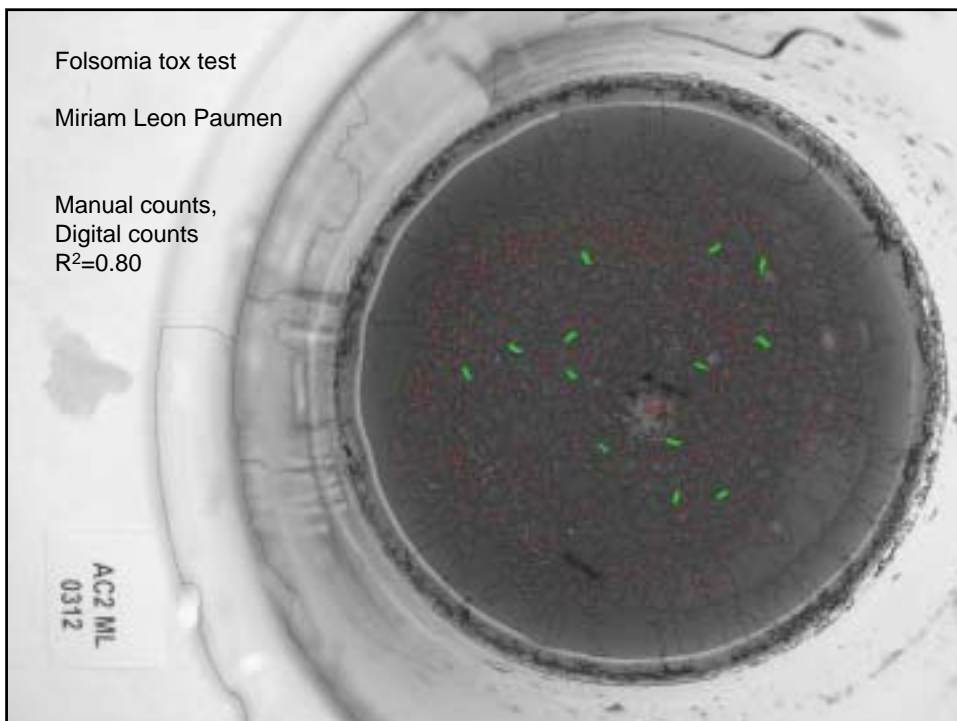
Miriam Leon Paumen



Folsomia tox test

Miriam Leon Paumen

Manual counts,
Digital counts
 $R^2=0.80$

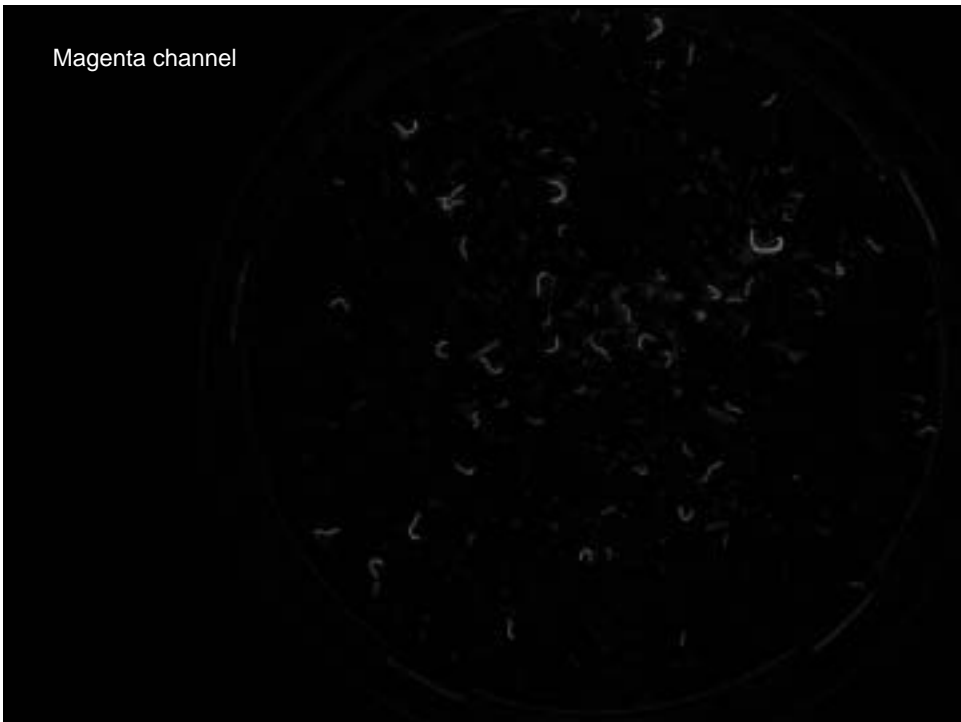


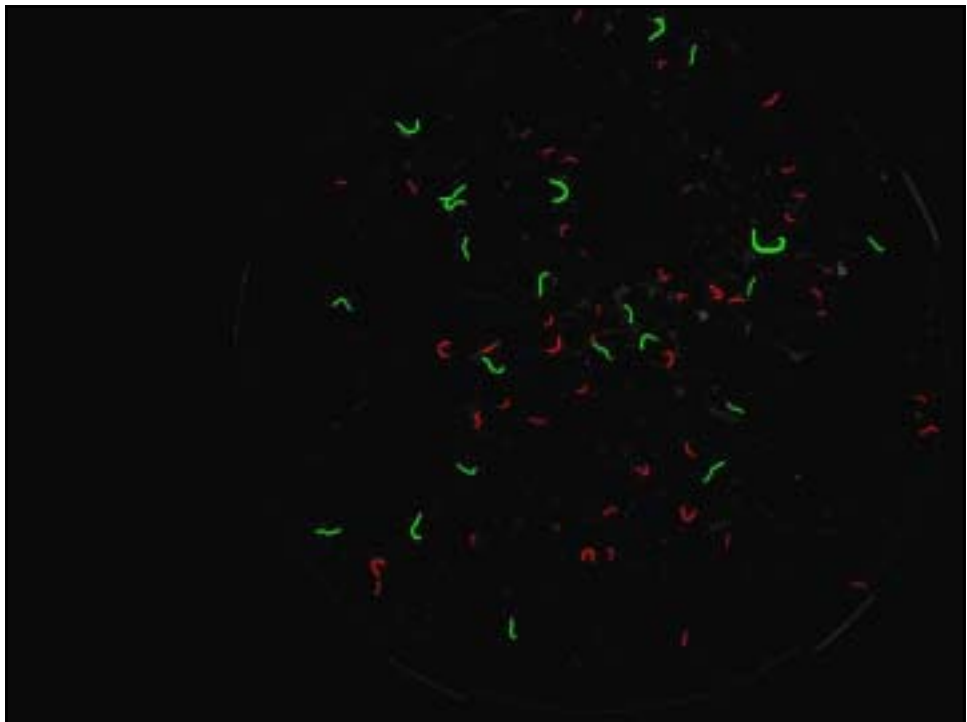
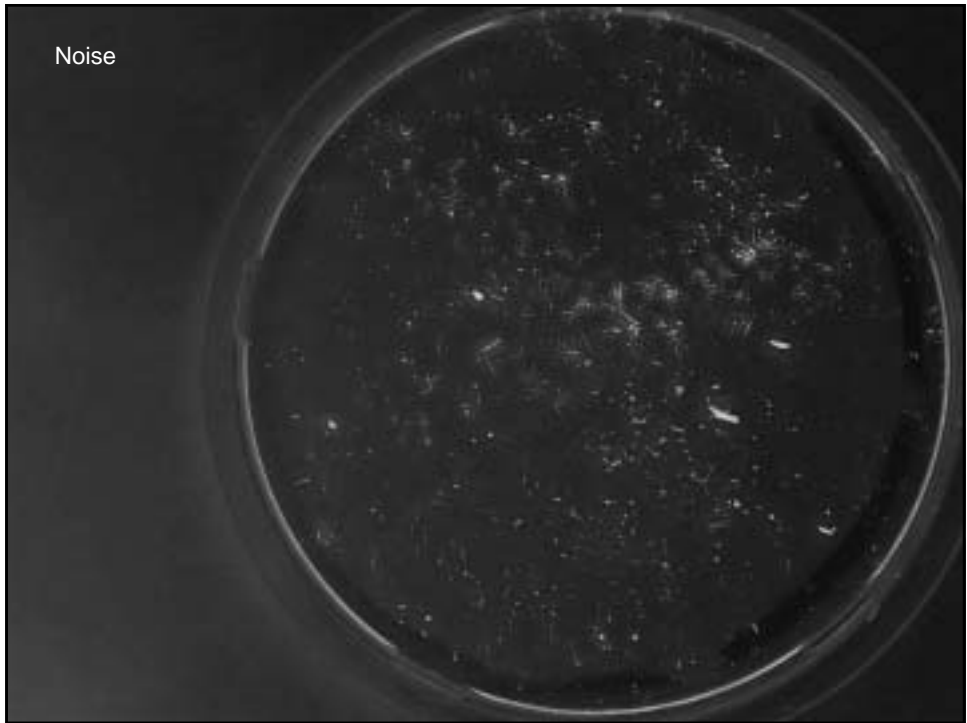
Enchytraeus tox test

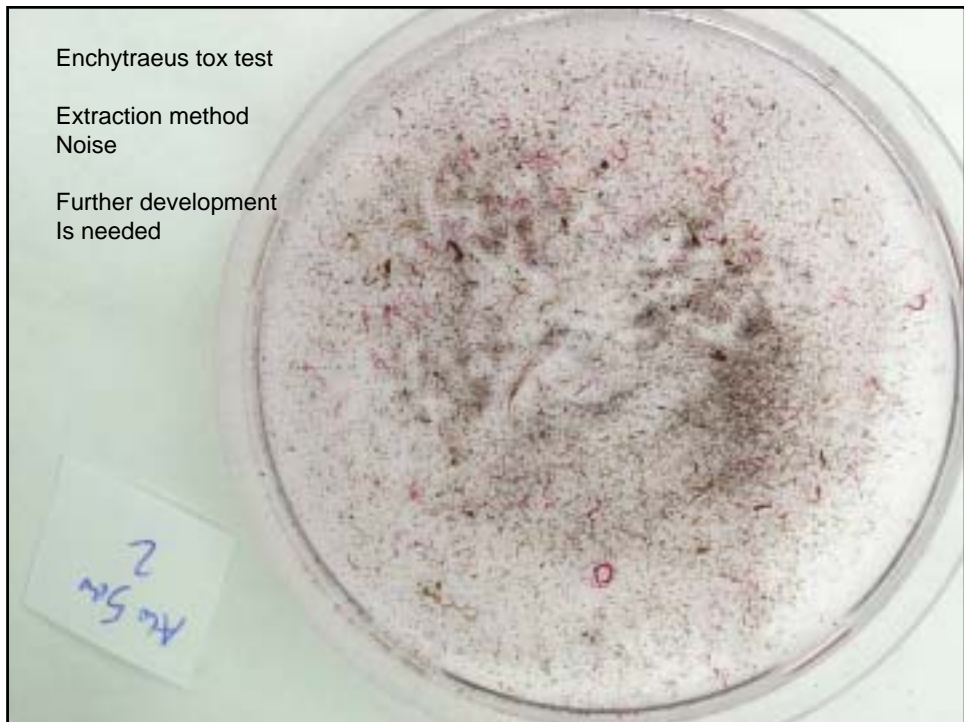
Miriam Leon Paumen



Magenta channel







Conclusions

- New counting tools for Daphnia are available
- Reliable counts of numbers, size distributions
- Monitor population dynamics in lab and field situations, using living and fixated individuals
- Useable for other organisms. E.g. birds, springtails

