How can different diets and different population densities induce sexualisation of Yamatohime-mimizu (*Enchytraeus japonensis*)?

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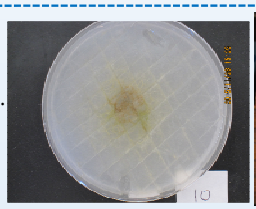
Research Question: *Enchytraeus japonensis known as* Yamatohime-mimizu is a creature that can reproduce sexually and asexually. The worm might choose either way of reproduction responding to the environment. Hokkaido University E. japonensis research group suggests possible factors inducing sexualisation, among which I focused on food and population. The research question is formulated as: How can different diets and different population densities induce sexualisation of Yamatohime-mimizu?

Experiment 1: Different diets

Oatmeal and freshwater fish food were used.
- Region 1: Oatmeal is mixed with culture.
- Region 2: Oatmeal is mixed with culture which was coloured blown with barley tea.
- Region 3: Oatmeal is put on culture daily.
- Region 4: Fish food is put on culture daily.

0.26g of food was given per petri dish. 4 worms were placed per petri dish with 90mm diameter. I made some petri dishes' duration longer than others.



A petri dish from experiment 1 region 4

Experiment 1 region 2

Experiment 1: Results

Experiment 1 Week 4

Region	Total	Asexual	Sexualised	Sexualised ratio(%)
2	8	8	0	0
	19	17	2	10.5
	0	0	0	0
	25	25	0	0
	0	0	0	0

Experiment 1 Week 5

Region	Total	Asexual	Sexualised	Sexualised ratio(%)
1	51	51	0	0
3	48	48	0	0
	50	50	0	0
4	26	24	2	7.7

Experiment 1 Week 6

Region	Total	Asexual	Sexualised	Sexualised ratio(%)
3	158	158	0	0
4	88	81	7	8

Experiment 1 Week 7

Region	Total	Asexual	Sexualised	Sexualised ratio(%)	Average sexualised ratio(%)
1	134	132	2	1.5	1.65
	27	27	0	0	
	66	65	1	1.5	
	55	53	2	3.6	
3	171	171	0	0	0
	108	108	0	0	
	85	85	0	0	
	116	116	0	0	
	134	134	0	0	
	78	78	0	0	
4	59	59	0	0	3.57
	116	116	0	0	
	75	67	8	10.7	

Experiment 2: Population density

- Region1: Population density was 0.168 worm/㎠using 55mm diameter petri dish.
- Region2: Population density was 0.063 worm/㎠using 90mm diameter petri dish.
- Region3: Population density was 0.024 worm/㎠using 145mm diameter petri dish.

0.19g fish food was given per petri dish. 4 worms were placed in each petri dish. Duration of this experiment was 36 days.

Experiment 2: Results

Region 1

Petri dish	Total	Asexual	Sexualised	Sexualisation ratio(%)
1	6	6	0	0
2	19	19	0	0
3	3	3	0	0

Region 2

Petri dish	Total	Asexual	Sexualised	Sexualisation ratio (%)
1	26	26	0	0
2	24	24	0	0
3	28	28	0	0

Region 3

Petri dish	Total	Asexual	Sexualised	Sexualisation ratio (%)	Average sexualisation ratio (%)
1	93	72	21	22.6	22.3
2	109	94	15	13.8	
3	82	57	25	30.5	

Petri dishes of experiment 2

Results: Different foods had effects on sexualisation differently. Fish food induced more sexualised worms than oatmeal had. For example, region 4 where fish food was fed got the highest average sexualisation ratio compared to other regions. The fish food contained protein much more oatmeal had (about 50 and 15% respectively). The oatmeal's main ingredient was carbohydrate. So it may be that the protein in higher ratio of fish food caused higher ratio of the sexualisation. However, t-test value does not support this conclusion. Population density had influence on sexualisation. Worms in the lowest population density were sexualised. The sexualisation ratio was 22.3%. T-test value supports this result and also corresponded with Hokkaido University research group's study.

Further study: Repeating the experiments under laboratory condition is vital to have more reliable results. At the same time, doing field work to observe Yamatohime-mimizu is crucial to make understanding of its reproductive strategy, which has never been found since described as a new species in 1993.

Asexual *Enchytraeus japonensis*

Sexualised *Enchytraeus japonensis*

A cocoon