Symmetry Breaking in Escaping Zebra Fish

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Abstract: It has been predicted theoretically that individuals confined to a room can produce a nonsymmetrical use of two identical exit doors under emergency. To some extent, the existence of that phenomenon has been demonstrated experimentally by using ants as a model of pedestrians. However, we found something different when the object changed to be zebra fish, which belongs to non-social animal but do well in schooling. We show that zebrafishes confined to a tank compartment with two symmetrically located exits prefer one of the exits to abandon it in normal conditions and the total number of escaping fishes increases as the volume of the compartment decreases. In addition, compared with normal conditions, the non-symmetry became weaken if panic is created by adding a repellent fluid. To some extent, our experimental results suggest some similarities between the behavior of humans and fishes since human beings are both of social and individual characteristics. Humans do not like being trapped in a crowded room and tend to own adequate living space. Under emergency, a throng of people will always lose their previous order and be scared away in disarray.

Keywords: zebra fish, behavior, escape, symmetry breaking

Introduction

Many researchers paid much attention on the collective behavior of many social animals to explore some inner connection between a person and another person since the connection is hard to be quantitative analysed. They gave much emphasis to the “follow -the-crowd” effect when animals are in danger. Helbing et al. (2000) made an important step forward in a theoretical article where simulations of individuals escaping from a closed room were presented. That work has been rapidly followed by further theoretical models and by experiments in rats (Saloma et al. 2003), humans (Helbing et al. 2003), and ants (Ernesto Altshuler et al.2005). One of the most obvious achievements in these theoretical or experimental articles is the panic-induced symmetry breaking in the escape from a room with two exits.

However, we can observe symmetry breaking even without panic when we use zebra fish as a model of fugitives. In addition, we show that more fishes tend to abandon the compartment as the space decrease and behave less orderly under emergency. This result seems to suggest some similarities to the behaviors of human: pursuing enough living space, being scared away in disarray.

In nature, fish may detect and respond to altered water quality and avoid detrimental chemical conditions (1). In response to detrimental conditions, fish prefer to make movement rather than sustain the given condition (2) (3). Thus, the behavior of fish in response to altered water may be an important factor that will help human to monitor the water quality.

Materials and Methods

The zebrafish was used as test fish throughout because it is not a social species. Though zebra fishes belong to non-social animals, they do well in schools. They also thrive as shoals of six or more. The zebrafish can be easily bought in the fish market since it is a popular aquarium fish, frequently sold under the name zebra danio. And it is an important vertebrate model organism in scientific research. Their ease of keeping and breeding, price, playful nature and broad availability all contribute to their appropriateness for the experiment.

The size of the zebra fish used in the escape experiment ranged from 3 to 4 cm standard length. The fishes were maintained in a big round porcelain crock(Figure1A) in the laboratory by feeding them tropical fish food, which is made up of nutrition food fish and fish derivative, vegetable protein extracts, wheat germ etc. Tap water was used as a water source and a submersible pump was used to improve the oxygen content of the water. During the process of testing, drugs can be administered by adding directly to the tank, which is another advantage for choosing zebrafish as a material for the experiment.

A cuboid glass trough (Figure1B) 31cm long, 17cm wide, and 20cm deep was used to measure the behavior of fishes under different situations. Two froth boards of 0.5cm thickness, 22cm height were also needed to divide the trough into three compartments. The width of the boards should be the same with the tank. In order to make two escaping exits, two circular holes of 3cm diameter were made on the two boards, respectively. The distance between the center of the hole and the bottom of the board is 5cm.

Four types of experiments were performed. In each experiment, the tank contained water of 10cm depth. In order to avoid the effect that the outside environment may have on the fishes, we pasted white paper on all the faces of the tank except the above one. In the first(experiment 1), the two froth boards divided the tank into three compartments, both the left and the right 12.5cm long, the middle 5cm long. We simultaneously introduced a group of 200 fishes from the big porcelain

B

A

Figure 1: A, A big round porcelain crock to keep the fishes. B, A glass trough and two froth boards for the experiment. White papers were pasted on all faces except the above one. The two thin plastic boards were not presented here.

crock into the middle compartment with two exits symmetrically situated at left and right, which were initially blocked by two thin plastic board. Once the fishes were introduced, we pulled up the two plastic boards synchronously so that the fishes were able to escape through the two holes. After four minutes, we blocked the two holes again with the two plastic boards. Then we counted the number of fishes that escaped into the left and the right compartment and make records.

In the second and the third kind of experiment (experiment 2 and experiment 3)，everything took place as in experiment 1, with the important difference being that the distance between the two froth boards changed into 10cm,15cm, respectively. So the length of the left and the right compartment changed into 10cm in experiment 2 and 7.5cm in experiment 3.

In the fourth kind of experiment, just as in the experiment 2, the two froth boards divided the trough into three equal areas, each 10cm long. What was different was that before the fishes were introduced into the middle, a little ball of toilet paper which had been soaked in toilet water which contain 5% DEET for several minutes was rapidly dropped into the middle compartment. DEET is perhaps one of the most effective fish repellents known to man. We also tried a kind of fish medicine and citronella oil. The former seemed to make no

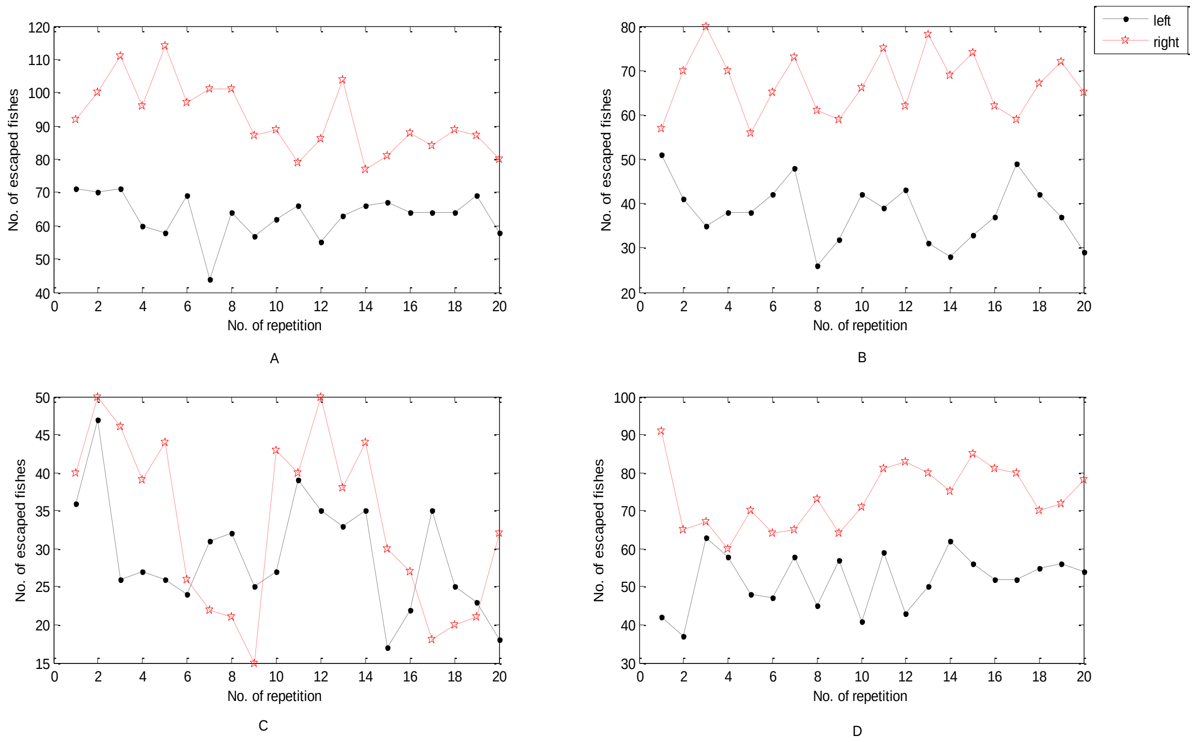

Figure 2: Graph A、B、C、D present the number of escaping fishes in experiments 1,2,3 and 4, respectively, with number of repetition as abscissa and number of escaping fishes as ordinate. The five-pointed star represent the number of fishes escaping right while the solid point represent the left.

difference while the later turned out to kill the fishes. So we chose the DEET as the repellent.

Every type of experiments were repeated 20 times. It should be noted that it was important to keep the total fishes 200. In the middle of the tests, some fishes may be injured or dead. Under the circumstance, we should replace them with living and healthy fishes to keep the total number 200.

Results and Discussion

On the basis of size measurements of 200 fishes, we got some data. Tables A1, A2, A3 and A4 present the number of fishes escaping to the left and the right area in the 20 repetitions of experiments 1, 2, 3 and 4, respectively. The final column of each table reports the percentage difference in use between the two holes, calculated as

$$\frac{|\text{total of fishes escaping right} - \text{total of fishes escaping left}|}{\text{total of escaping fishes}} \times 100.$$

On the basis of the number of fishes escaping right and left, we drew four scatter diagrams in Figure 2. If both exits are used with equal probability, we say that there is no symmetry breaking. If one of the doors is used preferentially, we say that there is symmetry breaking. A simple inspection of the four scatter diagrams suggest there is symmetry breaking since the number of fishes escaping right is generally larger than the other side. The fish prefer the right door to abandon the middle compartment. We can also use t test to investigate the result, if there is no symmetry breaking, the percentage difference in use between the two holes is expected to approximate to 0. In the case of experiment 1, 2, 3 and 4, the calculated value of t was 11.04, 12.02, 7.37 and 8.42, respectively, well above 2.86, which was the value of $t_{0.005}(19)$.So, the null hypothesis is rejected and the percentage difference in use between the two holes is greater than 0(p<0.01). Thus, there is symmetry breaking in the four types of experiment. With easy calculation, we got the mean value of the total number of escaping fishes in experiment 1, 2, 3: 155, 105, 63 (Figure3). 17cm multiplied by 10cm and then by 5cm, 10cm, 15cm make the volume of water in the middle compartment in experiment 1, 2 and 3, respectively. So it is obvious that the total number of escaping fishes increases as the volume of the compartment decreases. What is interesting is that the total number of escaping fishes increases by nearly 40 as the distance between the two boards decreases from 15cm to 10cm and 10cm to 5cm.

The mean value of the total number of escaping fishes in experiment 4 is 125, which is larger than that in experiment 2 by 20. However, the mean value of the percentage difference in use between the two holes in experiment 2 is 27.70, which is larger than that in experiment 4 by 10.10(shown in Figure4). Thus, we can conclude that though the repellent indeed make more fish swim away, the non-symmetry become weaken.

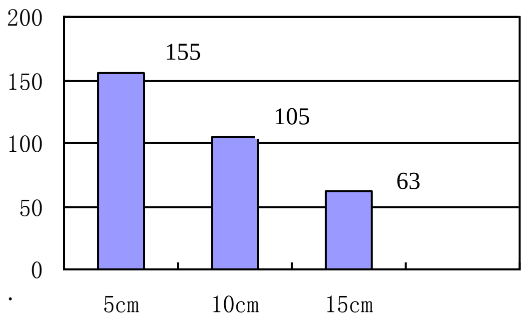

Figure3: The mean value of total number of escaping fishes in experiment 1, 2 and 3

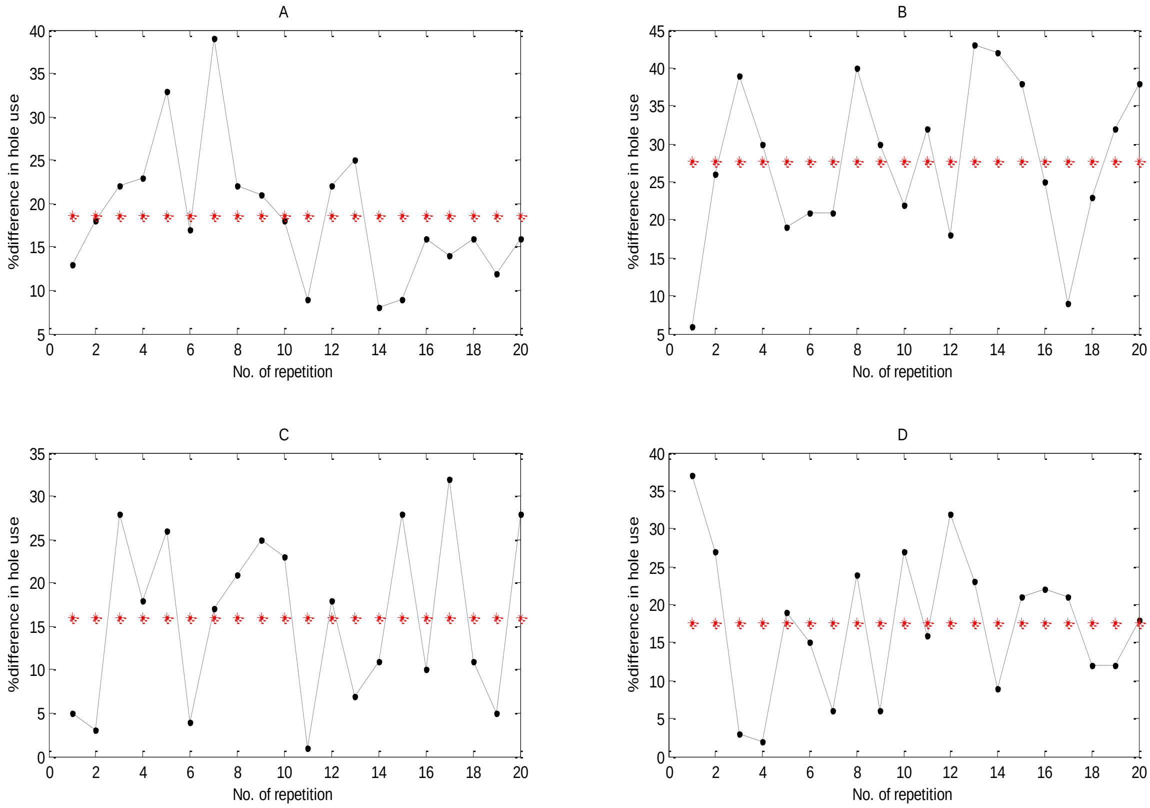

Figure 4: Graph A、B、C、D present the percentage difference in use between the two holes in experiments 1,2,3 and 4, respectively, with number of repetition as abscissa and % difference in holes use as ordinate. The marked line that parallels with the X-axis represent the mean value of the percentage difference in holes use and the numerical value is: 18.65, 27.70, 16.05, 17.60. The standard deviation of the percentage difference in the two holes use is: 7.55, 10.30, 9.74, 9.35. So the Coefficient of variation in the four types of experiment is: 40%, 37%, 60%, 53%.

The first column of tables B1, B2, B3 and B4 present the ratio between the number of fishes in the middle compartment and the total number, calculated as

$$MP = \frac{\text{number of fishes staying in the middle}}{\text{total number of fishes}(200)}.$$

The second column reports the density of fishes in the middle, calculated as

$$MD = \frac{\text{number of fishes staying in the middle}}{\text{the volume of the water in the middle}},$$

while the third column reports the density of fishes in the other two compartments, calculated as

$$LRD = \frac{\text{total number of escaping fishes}}{\text{the total volume in the left and the right compartment}}.$$

The volume of the water in the middle in experiment 1, 2, 3 and 4 is 0.85dm^3, 1.70 dm^3, 2.55 dm^3 and 1.70 dm^3, while the total volume of water in the left and the right compartment is 4.25dm^3, 3.40 dm^3, 2.55 dm^3 and 3.40 dm^3, separately. The last column reports the difference between MD and LRD.

The mean value of MP in the four types of experiment is 0.23, 0.48, 0.69 and 0.37, separately. The mean value of the difference between MD and LRD is 16.12, 25.25, 29.45 and 6.91, respectively. We can observe that the density of fishes in the middle is generally bigger and the difference between MD and LRD increases as the volume of the middle increases while it decreases very much when repellent is introduced. So maybe we can conclude that schooling fishes tend to stay where they are first introduced and live together, only when they are faced with some emergency, such as living space limit and water pollution will they prefer to escape to a better habitat. Why the fishes prefer to one hole when escaping is still a phenomenon that confused us.

Expansion

After we finished the four types of experiment mentioned above, we are surprised by the phenomenon that fishes prefer the right hole to abandon the middle compartment. In order to explore whether light have an influence on the preference of the fish, we repeated the second type of experiment 12 times in the evening and put the tank under a box that guarantee no light. What was different from experiment 2 was that the observation time lengthened from 4 minutes to 10 minutes. The reason why we length the time is that we want to know whether the fish will spread equally like gas when the time for escaping is long enough. Another difference was that the total number of fishes introduced into the tank was 90 in this fifth type of experiment since we had no enough fishes because a large proportion of fishes died over the one-month duration of the experiment. Anyway, we got some data that means something. Table A5 present the related data. The results that the fish still could find the hole to abandon the middle compartment without light really amaze us. The calculated mean value of the percentage difference in holes use in experiment 5 is 28.12, approximating to 27.70 in experiment 2.Thus, light seems to make no great difference to the symmetry breaking.

From Table B5, we can calculate that the mean value of the difference between MD and LRD is 7.79 and it is easily observed that the observation time lengthen to 10 minutes, the density of fishes in the middle is also bigger than that in the right and left side. This phenomenon seems to provide an evidence for our speculation.

Acknowledgements

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APPENDIX A
Experimental Data

Table A1: Results of 20 repetitions of experiment 1

Repetition	Left	Middle	Right	Percentage difference in door use(%)
1	71	37	92	13
2	70	30	100	18
3	71	18	111	22
4	60	44	96	23
5	58	28	114	33
6	69	34	97	17
7	44	55	101	39
8	64	35	101	22
9	57	56	87	21
10	62	49	89	18
11	66	55	79	9
12	55	59	86	22
13	63	33	104	25
14	66	57	77	8
15	67	52	81	9
16	64	48	88	16
17	64	52	84	14
18	64	47	89	16
19	69	44	87	12
20	58	62	80	16

Table A2: Results of 20 repetitions of experiment 2

Repetition	Left	Middle	Right	Percentage difference in door use(%)
1	51	92	57	6
2	41	89	70	26
3	35	85	80	39
4	38	92	70	30
5	38	106	56	19
6	42	93	65	21
7	48	89	73	21
8	26	113	61	40
9	32	109	59	30
10	42	92	66	22
11	39	86	75	32
12	43	95	62	18
13	31	91	78	43
14	28	103	69	42
15	33	93	74	38
16	37	101	62	25
17	49	92	59	9
18	42	91	67	23
19	37	91	72	32
20	29	106	65	38

Table A3: Results of 20 repetitions of experiment 3

Repetition	Left	Middle	Right	Percentage difference in door use(%)
1	36	124	40	5
2	47	103	50	3
3	26	128	46	28
4	27	134	39	18
5	26	130	44	26
6	24	150	26	4
7	31	147	22	17
8	32	147	21	21
9	25	160	15	25
10	27	130	43	23
11	39	121	40	1
12	35	115	50	18
13	33	129	38	7
14	35	121	44	11
15	17	153	30	28
16	22	151	27	10
17	35	147	18	32
18	25	155	20	11
19	23	156	21	5
20	18	150	32	28

Table A4: Results of 20 repetitions of experiment 4

Repetition	Left	Middle	Right	Percentage difference in door use(%)
1	42	67	91	37
2	37	98	65	27
3	63	70	67	3
4	58	82	60	2
5	48	82	70	19
6	47	89	64	15
7	58	77	65	6
8	45	82	73	24
9	57	79	64	6
10	41	88	71	27
11	59	60	81	16
12	43	74	83	32
13	50	70	80	23
14	62	63	75	9
15	56	59	85	21
16	52	67	81	22
17	52	68	80	21
18	55	75	70	12
19	56	72	72	12
20	54	68	78	18

Table A5: Results of 12 repetitions of experiment 5

Repetition	Left	Middle	Right	Percentage difference in door use(%)
1	13	23	54	61
2	18	30	42	40
3	21	36	33	22
4	21	32	37	28
5	26	32	32	10
6	36	30	24	20
7	34	38	18	31
8	18	47	25	16
9	19	47	24	12
10	17	52	21	11
11	28	52	10	47
12	30	47	13	40

APPENDIX B

Table B1:

Repetition	MP	MD	LRD	MD-LRD
1	0.19	43.53	38.35	5.18
2	0.15	35.29	40.00	-4.71
3	0.09	21.18	42.82	-21.65
4	0.22	51.76	36.71	15.06
5	0.14	32.94	40.47	-7.53
6	0.17	40.00	39.06	0.94
7	0.28	64.71	34.12	30.59
8	0.18	41.18	38.82	2.35
9	0.28	65.88	33.88	32.00
10	0.25	57.65	35.53	22.12
11	0.28	64.71	34.12	30.59
12	0.3	69.41	33.18	36.24
13	0.17	38.82	39.29	-0.47
14	0.29	67.06	33.65	33.41
15	0.26	61.18	34.82	26.35
16	0.24	56.47	35.76	20.71
17	0.26	61.18	34.82	26.35
18	0.24	55.29	36.00	19.29
19	0.22	51.76	36.71	15.06
20	0.31	72.94	32.47	40.47

Table B2:

Repetition	MP	MD	LRD	MD-LRD
1	0.46	54.12	31.76	22.35
2	0.45	52.35	32.65	19.71
3	0.43	50.00	33.82	16.18
4	0.46	54.12	31.76	22.35
5	0.53	62.35	27.65	34.71
6	0.47	54.71	31.47	23.24
7	0.45	52.35	35.59	16.76
8	0.57	66.47	25.59	40.88
9	0.55	64.12	26.76	37.35
10	0.46	54.12	31.76	22.35
11	0.43	50.59	33.53	17.06
12	0.48	55.88	30.88	25.00

13	0.46	53.53	32.06	21.47
14	0.52	60.59	28.53	32.06
15	0.47	54.71	31.47	23.24
16	0.51	59.41	29.12	30.29
17	0.46	54.12	31.76	22.35
18	0.46	53.53	32.06	21.47
19	0.46	53.53	32.06	21.47
20	0.53	62.35	27.65	34.71

Table B3:

Repetition	MP	MD	LRD	MD-LRD
1	0.62	48.63	29.80	18.82
2	0.52	40.39	38.04	2.35
3	0.64	50.20	28.24	21.96
4	0.67	52.55	25.88	26.67
5	0.65	50.98	27.45	23.53
6	0.75	58.82	19.61	39.22
7	0.74	57.65	20.78	36.86
8	0.74	57.65	20.78	36.86
9	0.8	62.75	15.69	47.06
10	0.65	50.98	27.45	23.53
11	0.61	47.45	30.98	16.47
12	0.58	45.10	33.33	11.76
13	0.65	50.59	27.84	22.75
14	0.61	47.45	30.98	16.47
15	0.77	60.00	18.43	41.57
16	0.76	59.22	19.22	40.00
17	0.74	57.65	20.78	36.86
18	0.78	60.78	17.65	43.14
19	0.78	61.18	17.25	43.92
20	0.75	58.82	19.61	39.22

Table B4:

Repetition	MP	MD	LRD	MD-LRD
1	0.34	39.41	39.12	0.29
2	0.49	57.65	30.00	27.65
3	0.35	41.18	38.24	2.94
4	0.41	48.24	34.71	13.53
5	0.41	48.24	34.71	13.53
6	0.45	52.35	32.65	19.71
7	0.39	45.29	36.18	9.12
8	0.41	48.24	34.71	13.53
9	0.4	46.47	35.59	10.88
10	0.44	51.76	32.94	18.82
11	0.3	35.29	41.18	-5.88
12	0.37	43.53	37.06	6.47
13	0.35	41.18	38.24	2.94
14	0.32	37.06	40.29	-3.24
15	0.3	34.71	41.47	-6.76
16	0.34	39.41	39.12	0.29
17	0.34	40.00	38.82	1.18
18	0.38	44.12	36.76	7.35
19	0.36	42.35	37.65	4.71
20	0.34	40.00	38.82	1.18

Table B5:

Repetition	MP	MD	LRD	MD-LRD
1	0.26	13.53	19.71	-6.18
2	0.33	17.65	17.65	0.00
3	0.40	21.18	15.88	5.29
4	0.36	18.82	17.06	1.76
5	0.36	18.82	17.06	1.76
6	0.33	17.65	17.65	0.00
7	0.42	22.35	15.29	7.06
8	0.52	27.65	12.65	15.00
9	0.52	27.65	12.65	15.00
10	0.58	30.59	11.18	19.41
11	0.58	30.59	11.18	19.41
12	0.52	27.65	12.65	15.00

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