On the Snapping of Carp, *Cyprinus carpio* LINNE, under Experimental Conditions

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"What kind of sense guides the food-searching behavior in fishes and induces this activity?" This question has attracted much attention for long years as being one of the most fascinating problems both from theoretical and practical view point, in the field of fisheries research.

In fact, many approaches to the functions of sensory system of fishes from the physiological view point have been reported by various researchers.¹⁾²⁾ Investigations along other lines have also been made about the possible relationship between some morphological properties of the nervous system and ecological characteristics of fishes revealed by field observations in natural conditions.³⁾⁴⁾

A large amount of those analytical data point to the fact that some of the ecological aspects of fishes are characterized by physiological and morphological characteristics of the nervous system. These relationships have proved to be of great value for interpreting results obtained in physiological and morphological investigations. However, they do not seem to be satisfactory for the solution of certain practical problems; as for example, the development of the technique of attracting and feeding fish in culture ponds, particularly in case of large ponds.

The present authors feel that many difficulties for the progress of scientific knowledge in practical fields stem from the scarcity of investigations dealing directly with the inducing and guiding mechanisms of food-searching behavior in fishes.

The purpose of this present research is to analyse under experimental conditions the effective stimuli inducing the food-searching behavior in various species of fish. And the present paper, as a part of this line of research, describes some effects of stimuli originating from the food itself or from other environmental factors, upon the frequency of snapping made by the carp, *Cyprinus carpio* LINNE, at the tip of a glass funnel through which the stimulant was introduced.

MATERIALS AND METHODS

The carps used for the experiments were obtained from concrete culture ponds at the Faculty of Fisheries and Animal Husbandry, Hiroshima University. They

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had been spawned in May of 1970 and reared in the same ponds until they attained 2.0 to 6.5 cm in standard length by July to August of the same year. Then they were transfered into aquaria for the experiments. The fish were fed on fish meal diet of appropriate size (Cramble No. 3, Pelet No. 5 and Pelet No. 6; products of Nihon Haigō Shiryō K. K.) except during the first month of their growth when they had fed on a diet of water flea.

About one month prior to the experiments the fish were transfered from the outdoor ponds into an aquarium $(21 \times 36 \text{ cm})$ containing 16.3l of well-water. The aquarium was placed in an isolated dark room in order to eliminate all other stimuli than those that were to be examined, and to regulate the light condition freely. Water of the aquarium was kept at almost constant temperature, ranging from 23.5° to 25.5° C, by an equipped thermostat-heater system (100W). The aquarium was also equipped with an air-lift circulating system with filter, in order to keep the quality of water in a good condition as well as to keep it clean. One third of the amount of water in the aquarium was renewed from time to time taking adequate care that fish were not disturbed.

Daily illumination and feeding schedules were strictly maintained. The light source consisted of two fluorescent tubes fixed on the ceiling of the room, about 2 m above the aquarium. They were switched on at 09:00 am and turned off at 05:30 pm everyday, except for the period from September 24 to October 10 of 1970. During this period the time interval of illumination was extended to 08:30 am and 06:00 pm for the observation of daily fluctuation in the snapping activities of the carps.

Feeding was made around 11:00 am and 05:30 pm everyday. For the feeding a fish meal diet was introduced into the aquarium through a glass funnel. The funnel was placed during the feeding time at a fixed corner of the aquarium. The lower tip of the funnel was placed everytime at a fixed depth in the water, 25 mm beneath the surface.

After their transfer to the aquarium, for a while the fish did not take the food particles until they had dropped onto the bottom of the aquarium. However when they got used to the new conditions the fish were observed to take food particles on the way to the bottom. After being well conditioned, the fish seemed excited by the placement of the funnel and they took food particles even just beneath the tip of the funnel.

At this stage of conditioning the fish were also seen snapping at the tip of the funnel even before the introduction of food through it. In the following description this pattern of behavior will be referred to as "snapping".

The well-conditioned fish only were used in the experiments. The experiments were carried out during the period from September 1970 to February 1971. For experimental purposes waters of different qualities were introduced instead of food materials during a period of 10 to 40 minutes in which the snappings were recorded by an observer. The observer was positioned 2 m away from the aquarium in order to avoid possible disturbance in the fish activity. The snappings were recorded

also for 5 to 10 minutes before and/or after the introduction of the waters

The snapping-activity in fish is expressed in this present paper in the form of the mean value of the frequency of snappings per minute calculated from the data recorded for different time intervals. This mean value will be refered to as "the rate of snapping" or simply "snapping rate".

The water used as stimulant was prepared as follows; Five grams of the fish meal was mixed in 100 m/ of destilled water and stirred well. The residual material was then filtered away in order to get a solution containing some water-soluble components of fish meal. This solution will be hereafter referred to as "the extract solution" or simply "the extract".

The water sampled from the aquarium was also used as "control water".

The extract solution and the control water came from the beaker placed at a level of 1 m above the aquarium, to the funnel through a vinyl tube having an inner diameter of 7 mm and equipped with a screw-cock to regulate the amount of inflow of the water.

RESULTS

Daily Fluctuation of Snapping Behavior

A series of experiments aimed to determine the most appropriate time of the day for making the observations on the snapping of carp. So the experiments were carried out at various times of the day from 08:30 am to 06:00 pm during a session from September 24 to October 10 in 1970.

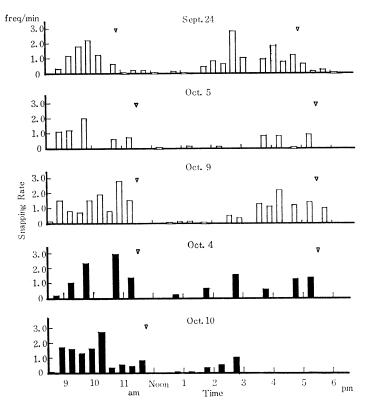
Four types of experiments were included in this series. In the first type (Type 1), the funnel was placed during 10 minutes for each observation and the snapping was recorded during this period. The experiments of this type were carried out on 24th of September, 5th and 9th of October. The second type (Type 2) consisted in the observation of the snapping response of the carps to the introduction of control water, initiated after fixing of the funnel and lasting for 10 minutes. The experiments of this type were carried out on 4th and 10th of October.

In the other two types of experiments, the funnel was placed during 20 minutes for each observation. However, in one of these types of experiments (Type 3), the control water was introduced through the funnel during the latter half of the period. The recording was made during both periods separately. Experiments of this type were carried out on 25th, 28th, 29th and 30th of September, 3rd and 8th of October. In the last type of experiments (Type 4), the introduction of control water was made during the first 10 minutes period and the funnel was left in its place during these two periods. The experiments of this type were carried out on 7th of October.

All the experiments of this series have been carried out on a group of 22 carps. The snapping rates were calculated for the responses of the group as a whole,

The results on the daily fluctuation of the snapping rate, obtained through the experiments of Types 1 and 2 are depicted in Fig. 1 and those obtained through the experiments of Types 3 and 4, in Fig. 2. It clearly appears evident from the figures that there are two periods of higher snapping activity in one day, one in the morning and the other in the afternoon, interspaced by a period of very low activity around noon.

For more detailed analyses on those data, the times of maximal occurrence of the snapping responses both to the fixed funnel alone and to the introduction of control water, are given in Table 1. No clear difference in the time of maximal snapping activity can be observed in the table among the different experimental conditions for both the kinds of reponse, that is the one to the fixed funnel alone and the other to the introduction of the control water.



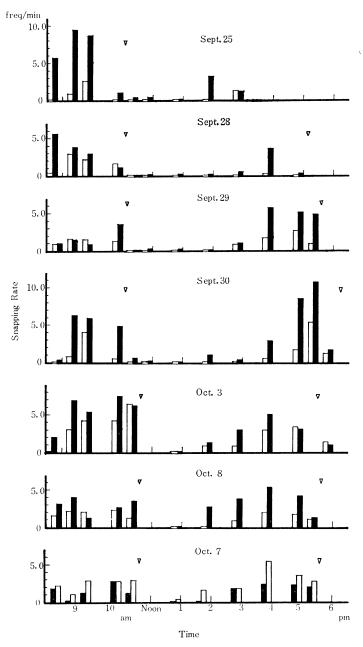


Fig. 2. Daily fluctuations of the snapping rates of the responses; to the fixed funnel alone (white columns) and to the introduction of control water (black columns) in the carp under the experimental conditions of Type 3 and Type 4, ∨ mark indicates the time of feeding.

Response to	Ехрег Туре	riment Date	Time (Snapping rate**)		
	1 Septem	ber 24	09:50 am (2.2)	02:50 pm (2.8)	
	October	5	09:40 (2.0)	indistinct	
		9	10:50 (2.8)	04:20 (2.2)	
	3 Septem	ber 25	09:40 (2.5)	02:40 (1.4)	
the funnel alone		28	09:20 (2.8)	indistinct	
		29	09:20 (1.6)	04:50 (2.6)	
		30	09:40 (4.0)	05:10 (5.4)	
	October	3	11.10 (6.4)	04:40 (3.4)	
		8	indistinct	03:40 (2.0)	
	4 October	r 7	indistinct	03:50 (5.4)	
	2 October	r 4	10:40 am (2.9)	02:40 pm (1.5)	
		10	10:20 (2.7)	indistinct	
	3 Septem	ber 25	09:20 (9.5)	01:50 (3.3)	
		28	08:40 (5.6)	03:50 (3.6)	
the introduction of		29	10:50 (3.5)	03:50 (5.7)	
control water		30	09:20 (6.3)) 05:20 (10.6)	
	October	r 3	10:50 (7.4)	03:50 (5.1)	
		8	09:20 (4.0)	03:50 (5.4)	
	4 Octobe	r 7	10:40 (2.8)) indistinct	
Mean and Standard deviation			09:58 am±:43	03:49 pm±:57	

Table 1. Times of the highest occurrence of the snapping response in the carps.*

* A group of 22 carps was used for this series of experiments.

** The snapping rates were expressed in the form of the frequency of snapping per minute.

The maximal activity in the morning concentrated to the zone between 09:20 am and 09:50 am, in 9 cases out of 17; at 09:58 am on an average with the standard deviation of 42 minutes. This means that under those experimental conditions the carps make snapping most frequently in the morning during the period from 20 minutes to 1 hour and 40 minutes prior to the regular feeding time.

On the other hand the maximal activity in the afternoon can be seen in a broader zone between 01:50 pm and 05:10 pm without any clear concentration. It is also seen that the activity reaches its maximum in the afternoon at 03:49 pm on an average with the standard deviation of 57 minutes, that is during the period from 4 to 6 hours after the regular feeding time of the morning.

In order to compare the levels of snapping from experiment to experiment, the mean rates of snapping and their 95% confidence limits in each experiment are presented in Table 2 for the response to the fixed funnel alone, and in Table 3 for that to the introduction of control water. It is clear from the tables that the confidence intervals for the mean rates of snapping are overlapping within the same type of experiments for both the kinds of response.

However, the comparison of the mean rate of snapping among Types reveals

Туре	Experiment	Date	Number of observations	Mean Snapping Rate*	Confidenc upper	e Limits** lower
1	September	24	26	0.76	1.04	0.46
	October	5	11	0.75	1.17	0.33
		9	22	1.11	1.45	0.77
	Type 1 in	total	59	0.89	1.08	0.70
3	September	25	9	0.64	1.29	0.01
		28	11	0.75	1.39	0.10
		29	12	0.99	1.51	0.48
		30	13	1.45	2.29	0.60
	October	3	11	2.43	3.76	1.01
		8	11	1.43	1.96	0.90
	Type 3 in total		67	1.23	1.61	0.84
4	October	7	11	2.47	3.43	1.52

Table 2. Mean snapping rates and the confidence limits of the responses to the fixed funnel alone in the carp.

* as same as in Table 1.

** at the probability of 95% level.

Туре	Experiment	Date	Number of observations	Mean Snapping Rate*	Confiden upper	ce Limits** lower
2	October	4	12	1.16	1.64	0.68
		10	14	0.95	1.40	0.50
	Type 2 in	total	26	1.05	1.42	0.68
	September	25	9	3.37	6.28	0.46
	1	28	11	1.70	3.00	0.41
		29	12	2.03	3.39	0.66
		30	13	3.47	5.63	1.31
	October	3	11	3.84	4.60	3.00
		8	11	2.92	3.38	2.46
	Type 3 in	total	67	2.87	3.62	2.12
4	October	7	11	1.43	2.04	0.82

Table 3. Mean snapping rates and the confidence limits of the responses to the inflow of the control water in the carp.

* as same as in Table 1.

** at the probability of 95% level.

that the mean value on Type 3 for the response to the introduction of control water is significantly higher than the mean values of the rate of snapping on the other Types (Table 3), while in the case of the response to the fixed funnel alone there is no significant difference in the mean value of snapping rate from Type to Type (Table 2).

According to the results obtained here, the following points must be heeded in further experiments on the effects of the extract solution upon the snapping activity of carps:

(1) Experiments should be carried out during the period from 09:40 am to 10:20 am, one of the periods of maximal snapping activity of carp in one day.

(2) The snapping activity upon the fixed funnel alone should be measured for reference everytime of the experiments.

Snapping Activity in the Case of the Introduction of Extract Solution

Three groups of carps were used in this series of experiments; the group 1 was consisting of 22 carps, the group 2 of 6 carps and the group 3 of 14 carps. And so the snapping rates are presented in this section in terms of the frequency of snappings per minute per capita in order to make it possible to compare the results obtained on the groups consisting of the different number of fish mutually.

The experiments were carried out at about 10:00 am everyday, one of the times of maximal snapping activity in carp, during the sessions of several successive days. In each experiment the frequency of snappings was recorded separately for three sectioned parts of observation. In the first part, the snappings at the fixed funnel alone were observed for about 10 minutes following the setting of funnel. In the next, the recording was made on snappings while the extract solution or the control water was being introduced. Thereafter, for 10 minutes again the snappings of fish at the funnel alone were recorded.

The extract solution and the control water were supplied to the aquarium alternatively every two days, and the rates of snappings were compared between the responses, the one to the inflow of the extract solution and the other to that of the control water. In spite of the care taken to regulate the amount of inflow

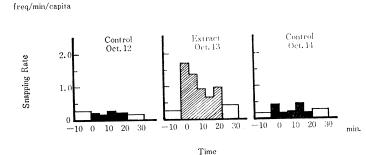


Fig. 3. Snapping rates of the response to the inflow of the extract solution (Extract) and of the control water (Control) in the carp (Group 1).

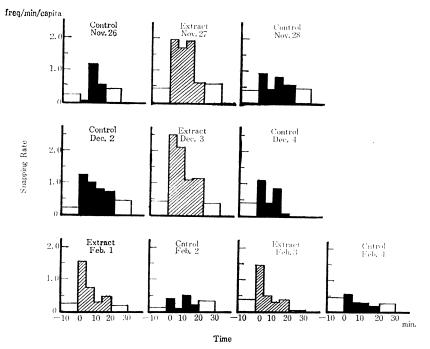


Fig. 4. Snapping rates of the response one inflow of the extract solution (Extract) and of the control water (Control) in the carp (Group 2).

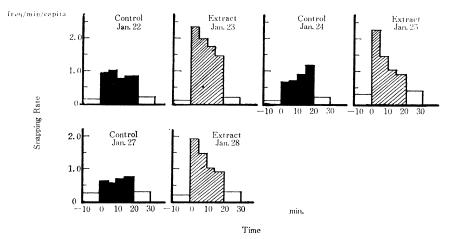


Fig. 5. Snapping rates of the respons to the inflow of the extract solution (Extract) and of the control water (Control) in the carp (Group 3).

by means of a screw-cock equipped to the leading tube, the duration of inflow of the almost fixed amount of the solution or the control water varied from experiment to experiment, by 20 to 25 minutes.

The fluctuation of the snapping rate in the course of each experiment are visualized in Fig. 3 for the group 1, in Fig. 4 for the group 2 and in Fig. 5 for

the group 3 respectively.

As shown in these figures, in the case of application of the extract solution, the snapping activity of carps always occurs in its maximal frequency at the time of initiation of inflow of the solution. And so it can be stated that the fluctuation of the snapping response to the inflow of the extract solution shows characteristically a tendency to decrease in the activity in the course of the inflow. The initial maximal values of the snapping rate, in other words, indicate properly the level of snapping response to the inflow of the extract solution.

On the other hand, it is evident from the figures that the snapping activity of

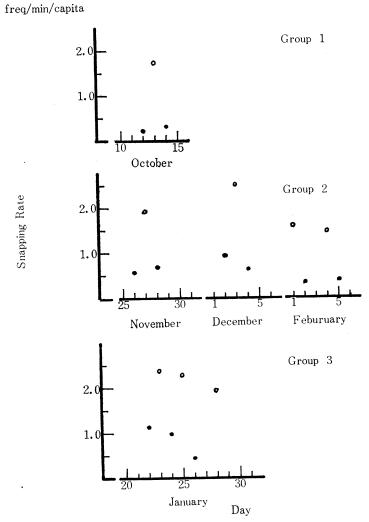


Fig. 6. The snapping rates of the response to the initiation of inflow of the extract water (white circles) and the mean snapping rates of the response to the inflow of control water (black circles) in carp.

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the carps in case of the inflow of the control water fluctuates very irregularly in its rate during the inflow; sometimes showing a steady tendency of decreasing as shown in the results obtained on the group 2 on December 2 for example, but sometimes showing an opposite tendency as the results of experiment on the group 3 on January 24 show. This situation allows us to represent the level of snapping response to the inflow of the control water in terms of the mean values of the obtained rates through the observation.

The activity of the responses to the inflow of the extract solution always stayed at higher levels than that of the responses to the inflow of the control water, as shown in Fig. 6.

It is also seen that the magnitude of dominance of the response to the inflow of the extract solution over that to the inflow of the control water remains almost fixed in a range from 1.7 to 1.0 freq/min/capita.

In conclusion, the above-mentioned analyses indicate that the inflow of the extract solution raises the level of snapping activity by 1.7 to 1.0 freq/min/capita when compared with the activity induced by the inflow of the control water.

DISCUSSION

The fact observed in the present study, that the frequency of snappings of the carps lowers very much during 4 to 7 hours following the real feeding (Table 1), seems to be in accordance with the findings of KARIYA $(1960)^{5}$ that the ration of the goldfish, a kind of stomachless fishes like the carp, does not attain its maximal value again, that is the ration for satiation without the starvation long enough, for longer than 5 to 6 hours after the previous feeding. The broader variation observed in the present study in the duration of low activity of snapping following the real feeding may be ascribed to the variation in the ration less than that for satiation, from experiment to experiment. This accordance assured that the snapping behavior observed here may be based on the appetitive motivation.

However, the presence of a maximal activity of snapping prior to the regular feeding time in the morning, suggests that the other factors may be involved in this behavior, because the long period of starvation over a night makes it very difficult to relate the fixed occurrence of the high snapping activity within a restricted times in the morning, to the appetitive factor alone. DAVIS and BARDACH (1965)⁶) have reported "the pre-light-pre-feeding activity of locomotion" in many species of fish reared under various schedules of feeding and illumination. The authors have concluded that the the high locomotory activity of this type is a consequence of conditioning the act of feeding to endogeneous cue. The snapping activity in the carp also is considered as being a consequence of the rearing of the fish under a fixed schedule of feeding and illumination during the pre-experimental period of about 1 month. In other words, the peaks of snapping activity prior to the regular feeding time in the carp may not be an essential element of the rythm of feeding activity as reported on the goldfish HIRATA and KOBAYASHI (1956)⁷) and HIRATA

(1957)⁸⁾ or on the amber-fish by HATANAKA, TAKAHASHI and MURAKAWA (1958),⁹⁾ but may be developed by conditioning.

Another evidence of the snapping behavior as being a consequence of conditioning is seen in the description on the change in the function of the funnel to induce this behavior in a course of the pre-experimental rearing (p. 40). In the earliest preexperimental period, the funnel still stays as a neutral stimulus. Later on, it becomes an effective stimulus inducing the snapping activity at the mean rate of 1.25 freq/min for a group of 22 fish, that is 0.06 freq/min/capita (Table 2).

The observed increase of the snapping rate, when the control water was introduced through the funnel, by 0.26 freq/min/capita (Figs 2–5) indicates that the inflow of the control water also functions as an inducer of the snapping behavior in the carp. Considering that there is no possible difference in the chemical and physical properties between the control water and the aquarium water as described in the previous section, Materials and Methods, the role of chemical sense as well as of a visual sense or thermal sense in detecting this inflow should be eliminated. Although the other possibility of using some mechanical senses by fish to the movement of water caused by the inflow of the control water is still existent, at the present time the further discussion on this problem is difficult to be developed because of lacking of the data on the movement of the water in the aquarium.

The further increase in the snapping activity of the carp is caused by the inflow of the extract solution by 1.17 freq/min/capita over that of the response induced by the inflow of the control water (Fig. 6). Since no difference is detected in physical properties, neither optical nor thermal, between both liquids, the increase in the rate of snapping by the extract solution should be ascribed to the difference in chemical properties between both the liquids. Furthermore the observed tendency of decrease in the rate during the inflow of the extract solution seems to indicate "the olfactory adaptation in the central integration".¹⁰ Anyway some chemical substances inducing the increase in the snapping of the carp must have their origin in the fish meal diet and must be soluble in water, functioning as an attractant. However, it still remained unsolved whether their attractive function is a consequence of conditioning or should be ascribed to an innate preference of the carp.

SUMMARY

The present study was made in order to analyse the factors which release the behavior of the carp, *Cyprinus carpio* LINNE, that is snapping at the tip of a glass funnel. The fish being conditioned to feed a fish meal diet introduced through the glass funnel were used.

The results can be summarized as follows;

1) The snapping behavior is released by setting of the funnel alone, as well as by the introduction of the water sampled from the aquarium (control water) or by that of the water containing some chemical components from the food (extract solution). 2) The snapping response to the fixed funnel alone or the infrow of the control water becomes most active during the period of 50 min. to 1 h. 40 min. prior to the regular feeding time, but lowers very much after the real feeding. And then they icrease again after the period of 4 to 7 hours of this low activity.

3) The frequency of snapping activities is observed to be the lowest in the case of the response to the funnel alone. Those in the response to the inflow of the control water are higher by 0.26 freq/min/capita than the previous ones. And finally the frequency of the response to the inflow of the extract solution is the highest, remaining at a higher level than that of the response to the control water by 1.17 freq/min/capita.

4) The comparison of the frequencies of the snapping activities under various experimental conditions with each other make it possible to analyse the stimulus signal releasing the food-searching behavior in the fish.

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コイの"つつき"に関する実験的研究

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漏斗を通して餌をとるように条件附けられたコイを用い,漏斗に対する"つつき行動"(snapping)を解 発する要因を解析して,次の結果を得た.

(1) つつき行動は、漏斗の設置、水槽水(対照水)又は餌の浸漬濾液(エキス液)の流入により解発される.

(2) 漏斗の設置又は対照水の流入に対して魚が行なうつつき行動は、定った給餌時刻の50分乃至1時間40分の間に著しく活発となり、給餌後は急激に低下する.然し、摂餌してから4乃至7時間たつと、つつき行動 は再び活発となる.

(3) 設置された漏斗に対するつつき行動の頻度は低く,0.06回/分/個体である.漏斗に対照水を流すとつつ き行動の頻度は0.26回/分/個体高まり,エキス液を流すと,更に1.17回/分/個体高まる.

(4) つつき行動の頻度を比較することにより、魚の索餌行動を解発する刺戟信号が解析出来る.