SOME PHYSIOLOGICAL OBSERVATIONS ON THE FRY OF *CHANOS CHANOS* (FORSKOAL) FOR THEIR TRANSPORT IN PLASTIC CONTAINERS

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INTRODUCTION

The problem of the transport of live fish, especially spawn and fry, has been a subject of great interest since last few decades. The main object in the transport of live fish, is to carry as many fish in as little water as possible with a minimum of mortality. This objective inevitably brings in several problems such as, size and weight of the fish to be transported, quantity of water medium, metabolic products of the fish, size and the shape of the container etc.

REVIEW OF THE PAST WORK

The live-fish transport was known to ancient Chinese and Romans (Norris, 1874). Gradual progress in the technique of the fish transport, has been the use of variety of containers from earthen pots and metal containers, to most modern transport tankers with their complicated circulating system. In India, the practice of transporting the fry of fresh water fishes like *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* in open earthenware hundies has been in vogue since time immemorial. Basu(1951) has given a review of this mode of transport. Mitra (1942) suggested that the fish fry could be better transported in sealed vessels with oxygen. Khan(1946) described an oxygenated container for transporting fish seeds, while Sundara Raj and Cornelius (1949) designed and used successfully, a tank of galvanised sheeting with oxygen container for the transport of mirror carp. Later, Ranade and Kewalramani (1956) used polythene bags filled with oxygen to transport the fry of *Labeo*, *Catla* and *Cirrhina*. Saha and Sen (1958) have studied the suitability of three different types of containers for the fish transport. Small polythene bags were put in use by the Fisheries Extension Department of the Government of India, for quite some time (Rao, 1961).

The methods given above take into consideration the physiological problems of fish transport, such as oxygen intake by the fish and the temperature variation in the water. However, besides these two, there are other factors like release of carbon dioxide and metabolic products and also the bacterial and fungal pollution of the medium of transport. Good amount of work has been carried out by Basu(1950 1951), Srinivasan *et al* (1955), Saha and Chowdhury (1956) and Saha *et al* (1956 a & b) in this direction. Work of Norris and others (1960) on the survey of fish transportation methods touches all aspects of this subject.

SCOPE OF THE PRESENT WORK AND MATERIAL AND METHODS

The present work was undertaken with a view to studying the basic physiological problems, such as the lethal level of oxygen and carbon dioxide, and also the optimum density of the fry of *Chanos chanos* of different sizes, in plastic containers, with and without oxygen pressure.

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The containers used in the present investigation are of transparent plastic, manufactured by M/s. Indian Plastics Ltd., Bombay, under the name *Bestolite new container* 759 (Fig. 1).

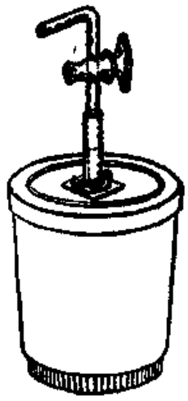

Fig. 1

Fig. 1—Plastic container (Bestolite new container 759) used in the present investigation.

This container has the advantage of a broad mouth with a screw cap and a resting ring at the bottom which keeps the actual bottom a centimeter above the floor. The broad mouth is convenient for pouring the water and fry and also for cleaning the containers. The screw-cap prevents the spilling of the contents. The resting ring at the bottom affords insulation. Moreover, the container is also pleasing to look at. However, it has a disadvantage that, since the cap is only about a centimeter in height and slightly bigger than the container itself (Fig. 1), it is difficult to make the container air-tight. These containers were preferred to polythene bags as the latter has the disadvantage, that they are delicate and weak and hence give way under stress (Saha and Sen, 1958). The polythene bags also cannot be used for high pressure packing and even the slight pressure given at the time of packing is lost in course of time due to diffusion, especially when thinner bags are used.

A small hole of 5 mm. diameter was bored in the centre of the cap of the container and a cycle valve chuck with a double soft rubber padding at the base was fixed. The possible leakage at the base was prevented by a coat of adhesive cement. To the chuck was fixed a glass stop-cock with the help of a rubber tubing, as shown in Fig. 1. The arms of the stop-cock were suitably cut and bent.

The *Chanos* fry were collected from Pamban and Mandapam lagoon and brought to the laboratory in conventional tin carriers. They were released in shallow wooden tanks and slowly acclimatised to fresh water. The fry were fed on the bottom mud layers, especially brought from their natural habitat. The experiments were started after about twelve hours. Required number of fry of a definite size was put in each container, in 100 ml. of well aerated water. The container was closed and the cap made air-tight by applying plasticine or soft paraffin wax all round the rim. This was further covered by two layers of cellophane tape. This made almost all the containers completely air-tight. Each container was then slightly evacuated with a low pressure vacuum pump. The amount of vacuum was judged by the depressed screw-cap of the container. The stop-cock was closed and then attached to an oxygen cylinder, Oxygen was passed through a fine adjustment valve supplying oxygen in litres per minute. The speed of the gas was maintained at half a litre per minute. This enabled the gas to pass very slowly. When the depressed lid raised to its normal size and even slightly bulged, the gas supply was stopped, the cock was closed and the container was left in a cool place at room temperature.

The fry were observed at the periodic intervals to note their swimming movements, general behaviour and mortality. Some containers developed cracks on the cap and leakages at the cap rim. In these containers, the mortality was invariably heavy and they were, therefore, discarded.

Several sets of experiments were performed with and without oxygen pressure. In each set, about 10 containers were employed. Experiments were carried out with the density of fry at the levels of 50,75,100,125, 150, 175 and 200, for the fry of the transparent stage (size, 13-14 mm.). For the bigger fry (15-21 mm.), only two experiments at the density levels of 100 and 125 were performed. While the fry of the largest size (22-25 mm.) were used at the density level of 50 only. In all the cases, the quantity of the water was kept constant at 100 ml. Totally, 16,275 fry were used in this series of experiments. The time limit was fixed at 48 hours in all cases. Only in some cases, where the mortality was high, the experiment was concluded before 48 hours.

Fresh water used in the experiments had the pH of 8·0 and the temperature ranging between 28·0 to 29·0° C. Average air temperature during the period of experiments was 30·4°C.

The dissolved oxygen content of the water in the containers was estimated before and after the experiment. The free carbon dioxide was estimated only at the end of the experiment. The standard methods by Ellis *et al* (1948) were employed for these estimations.

RESULTS AND DISCUSSION

Results of the experiments with the different densities of fry of three size-groups, with and without oxygen pressure, have been shown in Tables I to V. Reference to Table I will indicate that 100 fry of transparent stage could survive in 100 ml. of water for 48 hours, without oxygen pressure. The mortality in these three experiments is low, except in two containers *i.e.*, 2 and 4 in the second experiment. However, if the density of fry is increased to 125, the mortality is more, and in many cases it is very heavy (Table II). The exceptions are only three containers namely 8, 10 and 12. The same number of fry kept under oxygen pressure show cent per cent survival in all the containers. From this, it appears that *Chanos* fry of transparent stage could be transported in the density of 100 fry in 100 ml. of fresh water, without the oxygen pressure. The higher density shows appreciable mortality. To find out, how far the density of fry could be increased under oxygen pressure, further experiments were conducted.Table 3 shows that this density level could be increased under oxygen pressure, up to 200 fry per container. It would be seen that even at this level, the mortality is not appalling. Only in four containers *i.e.*, 2, 3, 4 and 5 the mortality appears more or less heavy.

It is further interesting to see from Table III that old stock of about 7 to 9 days shows heavy mortality, even at the low level of 175 fry per container, in less than 48 hours time. This indicates that fry should be transported as early as possible after their collection, and should not be kept in artificial surroundings for more than 3 to 4 days.

Table IV indicates that larger fry of average size 16·23 mm. could easily be transported under oxygen pressure, at the density levels of 100 to 125 fry per container. Owing to non-availability of the fry of this size, no further experiments could be carried out. However it is felt that in this case, the density level could not possibly be increased beyond 150 fry per container, under oxygen pressure. The largest fry of the average size of 22·98 mm. show cent per cent mortality within 10 hours even at the level of 50 fry per container, without oxygen pressure. Their viability could be increased to some extent under oxygen pressure (Table V). It is suggested that the problems relating to the physiological aspects for the transport of the fry of over 21·0 mm. size, should be studied more intensively.

From the experimental results given above, it was possible to calculate the optimum water volume with or without oxygen pressure, for the fry of a definite average size and weight for their transport up to 48 hours. Table VI gives the results of this study. It may be mentioned in this connection, that though 200 fry of the transparent size could remain with low mortality for 48 hours under oxygen pressure, it is felt that this density level is too high for the transport. The fry show overcrowding in the containers and hence the density level of 150 or 175 fry per container appears more suitable . Saha *et al* (1956 a) have given the maximum number of carp fry of different size-groups and weights, that could be kept in a closed system under oxygen pressure. They have also determined the minimum water volume required per carp fry, which comes to 2·0 c.c. for the fry of 17·7 to 19·1 mm. size and the average weight of 0·076 g. However, the authors have not mentioned the total period of the experiment. It will thus be seen that in the case of *Chanos* fry, the volume of water required per fry is slightly less for the period of 48 hours (Table VI).

Regarding the oxygen values in tables, it will be seen that in no container the final oxygen value is excessively low except in those with heavy mortality. In the Table IV, the oxygen value is low even where there is no mortality. In Table II, container 7 in the first experiment, shows oxygen level at 0·76 ppm. with a mortality of only 39·2%. This suggests that the lethal limit of oxygen may be below 0·7 ppm. at least for the fry of transparent stage. Saha *et al* (1956 a) observed that oxygen concentration above 0·5 ppm. is sufficient for the major carp of 5 cm. length.

It could also be seen from the data presented here, that in some containers the mortality was heavy but the oxygen level had not fallen below the probable lethal limit. This brings in the possibility that carbon dioxide in the water may perhaps be responsible for this mortality. It is of course evident, that the containers where mortality is heavy, the carbon dioxide content is also higher. From the data, it is difficult to establish a critical level of carbon dioxide. However, it may be said roughly, that the carbon dioxide level of more than 20·00 ppm. is harmful to *Chanos* fry. The containers where the carbon dioxide level is more than 30·00 ppm., there is invariably a very heavy mortality irrespective of final oxygen level. Ivlev (1936) and Vass (1951) observed that the accumulation of carbon dioxide may be a cause of fish mortality. To counteract the toxic effects of carbon dioxide, Vass (1951) and Srinivasan *et al* (1955) used secondary sodium phosphate while Saha *et al* (1956b) employed pulverised earth, activated charcoal, amberlit and the secondary sodium phosphate for the purpose. These authors further observed that spawn of major carps stand a free carbon dioxide concentration of 20 ppm. Basu (1950) has made a very interesting observation that the toxic level of carbon dioxide could be increased by increasing the oxygen level in the water. Thus, more is the oxygen content of the water, higher is the toxic limit of the carbon dioxide.

There was found to be a certain amount of post-experimental mortality. This occurred within 15 minutes, after the fry were released in the glass trough, after experiment. Maximum mortality was found to be 27·00% while the minimum was 0·33%. The causes of this post experimental mortality are not known.

SUMMARY

The experiments with the fry of *Chanos chanos* (Forskal) were conducted to find out the optimum density of the fry of different sizes for transport with and without oxygen pressure.

It was found that 100 fry of the transparent stage could be put in the containers without oxygen while 150 to 175 fry could be packed in a container with oxygen pressure. The fry of the larger size demand lesser density. The effects of oxygen and carbon dioxide levels on the survival of the fry and also the lethal levels of oxygen and carbon dioxide have been discussed.

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TABLE I.

Viability of Chanos *fry of transparent stage at different density levels without oxygen pressure.*

Number of fry :—50. Size : 13·0—14 mm. Avg. Weight : 0·010 g.

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality at 48 Hrs. %
1	5·97	4·80	12·79	Nil
2	5·97	4·52	17·06	12·0
3	5·97	5·09	21·33	16·0
4	5·97	4·66	17·06	Nil
5	5·97	4·80	17·06	Nil
6	5·97	5·55	8·53	Nil
7	5·97	5·17	8·96	4·0
8	5·97	5·02	10·67	4·0
9	5·97	5·17	13·22	2·0
10	5·97	4·66	11·09	6·0
Number of fry : 75				
1	6·55	3·64	17·49	9·54
2	6·55	2·69	21·76	20·00
3	6·55	2·98	20·48	9·54
4	6·55	3·20	24·31	30·66
5	6·55	3·72	16·21	6·66
6	6·55	3·72	13·65	5·33
7	6·55	4·59	9·81	8·00
8	6·55	4·52	9·39	1·34
9	6·55	3·72	17·06	10·67
10	6·55	3·93	11·09	10·67

TABLE I—*Contd.*

Number of fry : 100

1	6·70	2·62	25·60	2·00
2	6·70	3·42	19·63	8·00
3	6·70	2·62	27·73	9·00
4	6·70	2·91	20·90	3·00
5	6·70	2·98	21·76	2·00
6	6·70	3·28	21·76	Nil
7	6·70	2·40	21·76	Nil
8	6·70	3·20	23·03	Nil
9	6·70	3·06	20·04	12·00
10	5·09	3·08	6·60	2·00
11	5·09	4·02	6·60	2·00
12	5·09	2·78	6·60	5·00
13	5·09	2·26	6·60	Nil
14	5·09	1·96	6·60	1·00

TABLE II

Viability of Chanos *fry of transparent stage without and with oxygen pressure at the density level of 125 fry.*

Without Oxygen

Size : 13·0—14.0 mm. Avg. Weight : 0·010 g.

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality at 48 Hrs. %
1	6·55	1·53	33·28	96·81
2	6·55	1·09	20·48	78·39
3	6·55	1·31	27·30	97·61
4	6·55	0·36	35·83	98·40
5	6·55	1·75	21·33	68·79
6	4·93	3·02	17·60	11·20
7	4·93	0·76	22·00	39·20
8	4·93	1·86	15·40	0·80
9	4·93	1·10	17·60	13·60
10	4·93	1·86	15·40	1·60
11	4·90	2·36	11·00	56·01
12	4·90	2·46	8·80	5·60
13	4·90	2·96	8·80	8·80
14	4·90	2·46	8·80	16·00
15	4.90	2·41	8·80	20·80
With Oxygen				
1	5·20	3·18	15·40	Nil
2	5·20	3·56	13·20	Nil
3	5·20	3·04	15·40	Nil
4	5·20	2·56	13·20	Nil
5	5·20	3·76	13·20	Nil
6	4·94	3·94	15·40	Nil
7	4·94	3·80	15·40	Nil
8	4·94	2·56	15·40	Nil

TABLE III

Viability of Chanos *fry of transparent stage at different density levels with Oxygen pressure.*

Number of fry : 150. Size : 13·0—14 mm. Avg. Weight : 0·010 g.

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality at 48 Hrs. %
1	5·12	3·14	13·20	0·67
2	5·12	3·54	13·20	0·67
3	5·12	3·14	17·60	5·33
4	5·12	2·56	13·20	0·67
5	5·12	2·56	15·40	3·33
6	5·10	3·60	15 40	5·33
7	5·10	3·28	15·40	5·33
8	5·10	3·28	13·20	0·67
9	5 10	3 08	15 40	5·33
10	5·10	2·72	15·40	5·99
Number of fry : 175				
1	5·09	4·12	17·60	Nil
2	5·09	3·68	17·60	1·14
3	5·09	2 68	15 40	Nil
4	5 09	2·10	15·40	Nil
5	5·09	3·28	15·40	Nil
6	5·09	1·44	17·60	4·00
7	5·09	2·06	17·60	4·00
8	5·09	2·88	17·60	10·29
9	4·90	1·92	15·40	12·57
10	4·90	2·76	17·60	17·72
11	4·90	1·80	19·80	73·14
12	4·90	2·60	17·60	32·00
13	4·90	3·72	15·40	15·43
Number of fry : 200				
1	4·80	3·28	22·00	18·50
2	4·80	1·20	22·00	27·00
3	4·80	1·48	22·00	55·00
4	4·80	1·42	24·20	71·00
5	4·80	3·12	19·80	22·00
6	4·80	3·62	15·40	1·00
7	4·80	3·06	15·40	3·50
8	4·80	2·84	15·40	5·50
9	4·80	2·94	17·60	11·00
10	4·80	3·12	15·40	6·50

TABLE III—*Contd.*

Old stock of fry

Number of fry : 175. Size : 13·0—14 mm. Avg. Weight: 0·010 g.

1	4·90	2·46	15·40	51·42	Experiment concluded at 43 Hours.
2	4·90	2·68	17·60	49·72	
3	4·90	2·88	17·60	34·85	
4	4·90	2·88	19·80	44·58	
5	4·90	3·36	17·60	32·58	
6	4·94	2·16	13·20	52·58	Experiment concluded at 24 Hours.
7	4·94	3·08	15·40	40·58	
8	4·94	2·92	15·40	37·71	
9	4·94	3·02	15·40	45·14	
10	4·94	2·48	15·40	38·29	
11	5·09	1·48	30·80	79·43	Experiment concluded at 48 Hours.
12	5·09	1·80	26·40	56·57	
13	5·09	0·21	33·00	99·42	
14	5·09	0·36	33·00	97·72	

TABLE IV

Viability of Chanos *fry of size* 15-21*mm. at different density levels with Oxygen pressure*

Number of fry : 100. Avg. Size : 16·23 mm., Avg. Weight: 0·021 g

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality at 48 Hrs. %
1	4·94	1·18	19·80	3·00
2	4·94	1·08	17·60	Nil
3	4·94	2·46	19·80	3·00
4	4·94	1·24	15·40	Nil
5	4·84	1·08	11·00	Nil
6	4·84	1·02	13·20	Nil
7	4·84	0·92	15·40	10·00
8	4·84	1·18	19·80	11·00
9	4·84	1·24	15·40	6·00
Number of fry : 125				
1	4·90	2·52	15·40	1·60
2	4·90	3·62	15·40	3·20
3	4·90	2·68	15·40	8·00
4	4·90	1·60	19·80	31·20
5	4·90	4·10	13·20	0·80
6	4·90	2·68	15·40	4·00
7	4·90	3·54	15·40	7·19
8	4·90	4·14	15·40	3·20
9	4·90	4·44	13·20	0·80

TABLE V

Viability of Chanos *fry of average size* 22·98 *mm. without and with Oxygen pressure.*

Without Oxygen.

Number of fry : 50 Avg. Weight : 0·086 g·

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality %	
1	4·64	Nil	52·80	100	All began surfacing within two hours and died within ten hours.
2	4·64	Nil	52·80	100	
3	4·64	Nil	52·80	100	
4	4·64	Nil	52·80	100	
5	4·64	Nil	52·80	100	

With Oxygen.

Number of fry : 50

Container No.	Initial Oxygen ppm	Final Oxygen ppm	Carbon dioxide ppm	Mortality %	
1	4·90	1·54	17·60	18·00	Experiment concluded at the end of 25 hours.
2	4·90	2·78	19·80	6·00	
3	4·90	3·90	19·80	12·00	
4	4·90	1·90	24·20	32·00	
5	4·90	4·02	22·00	12·00	

TABLE VI

Optimum water volume and density for the transport of Chanos *fry of a definite size and weight without and with Oxygen pressure.*

No.	Average length mm.	Average weight g.	Water volume per fry ml.		Optimum density of fry for 100 ml. water
			Without Oxygen	With Oxygen	
1	13·50	0·010	1·0	..	100
2	13·50	0·010	..	0·67	150
3	13·50	0·010	..	0·57	175
4	16·23	0·021	..	0·80	125
5	22·98	0·086	..	2·00	50